## OKLAHOMA STATE UNIVERSITY

BODY COMPOSITION OF BROILER BREEDER HENS DURING PRODUCTION CYCLE, AN EVALUATION OF PANTOTHENIC ACID, AND MAGNESIUM SUPPLEMENTS ON BROILER PERFORMANCE

Ву

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Submitted to the Faculty of the Graduate college of the Oklahoma State University in partial fulfillment of the requirements for the degree of MASTERS OF SCIENCE May, 1999

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#### ACKNOWLEDGEMENTS

This thesis is dedicated to my family who helped me to reach this goal and every step of the way, and Vjollca with whom I would never of accomplished this task. I thank the Lord for his blessing in my life.

I would like to extend my thanks to my colleagues for their help and to Dr. Teeter for his considerable patience.

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## Chapter I Introduction

Nutritional management of modern broiler breeder strains has become very complex, in terms of amount, composition, and timing of feed provided to grow pullet and ' subsequent hen. In addition, it is unclear as to precisely what the pullet body condition is desirable throughout the growth curve for maximal egg production, fertility, and hatchability. It was previously thought that body weight at onset of puberty was the best variable for predicting hen performance, (Pearson and Herron 1980). Recent studies have reported that fat free empty body mass (lean and bone tissue) is more important in predicting broiler breeder performance, (Pearson and Herron 1980; Pearson and Herron 1981; Pearson and Herron 1982). Broiler breeders that are not restricted fed, enter onset of lay in an obese condition that results in decreased feed efficiency, laying cycle length, fertility, hatchability, and egg production

(Robinson, 1993; Yu et al, 1992). Broiler breeder nutrition is important to insuring a healthy productive flock and vibrant young broiler offspring.

Modern broiler strains are susceptible to heat distress due to high ambient temperature-relative humidity. Heat distressed broilers suffer from reduced weight gain, feed efficiency, and increased mortality and fat percentage. Metabolites needed for oxidative phosphorylation become sparsely distributed in the body. Repartitioning agents and supplements, such as pantothenic acid, are added to broiler diets to help the bird's metabolism compensate for the increased stress, and remain productive. The over all goal is to help the bird lower its heat production.

The study reported herein was conducted to further refine the knowledge base regarding nutritional effects on broiler breeder performance, to evaluate the pantothenic acid requirement in broilers exposed to heat stress and dirty litter challenge, and finally to evaluate the effects of magnesium supplements on broiler performance.

Chapters are prepared as manuscripts in style required by the Journal of Poultry Science to facilitate publication of experimental results.

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#### Chapter II Literature Review

#### Introduction

In modern broiler industry every aspect of live production is influenced by nutritional guidelines that presumably promote optimum performance. The broiler breeder, and subsequent progeny are provided nutrients to prepare the animal for efficient production be it egg or meat production. Genetic improvements made in today's poultry flocks, compared to birds earlier in this century have produced birds that must now be closely managed and provided proper nutrition, in order to assure optimal performance. This review focuses on few basic nutrient and therapeutics used in the poultry industry to help birds adapt to life cycle challenges.

#### Protein effects

Excess dietary protein has been shown to increase calcium excretion, however Heaney (1998) found that as long as dietary calcium is adequate skeletal structure in human females is not harmed, broilers most likely exhibit similar responses to protein. Laying hens are unique in that they secrete grams of calcium per day in an egg shell mass and therefore have a much higher calcium requirement than that of broilers. Rennie (1997) and associates showed that

despite calcium supplementation modern, laying hens become osteoporotic at cessation of lay.

Waldroup (1976) reported that broiler breeder hens consuming corn-soybean based diet require between 20-22 grams of protein per day. Lopez (1995) found improved performance and reduced body-weights with broilers fed a 10% CP diet compared to a 16% CP diet as long as synthetic amino acid are supplemented to prevent deficiency.

Energy is stored and utilized in the body primarily in chemical energy and thermal forms. Metabolism is fueled by the release of this stored energy from the hydrogen atoms of certain reduced carbon compounds. Practical diets for poultry are formulated on a metabolizable energy (ME) basis. Broiler ME is equal to gross energy value supplied by the ingested food minus losses due to fecal and urinary excretion, (Scott 1982). The primary sources of energy for poultry are fats and carbohydrates, both ready supplies of reduced carbons, (Stryer 1995). The energy level of the diet is the most significant factor, provided protein levels are adequate, in determining growth. Teeter and Smith (1985), observed that carcass growth rate was limited by physiological processes beyond feed intake. They found that above 140% of ad-libitum feed intake no further weight gain was noted due to genetic effects on appetite.

Body Composition

Body composition (BC) is commonly used to describe the amount of different types of cellular tissues (lean tissue, fat tissue, bone tissue) either as grams of tissue or a percentage of the total live body-weight.

Dual energy x-ray absorptiometry (DEXA) is a non invasive method of determining the BC in living animals, utilizing a constant potential x-ray. The DEXA system generates a high and low kVp x-ray beams that pass through the body tissue, and calculates the body tissue based on the amount of x-ray absorbed by the tissue, (Brunton et al, 1993). The DEXA system allows for repeated measurements on a subject, which may offer researchers a more accurately measure BC over many life stages. Proximate analysis requires a different bird be used for each BC measurement, which increases variation due to differences between birds.

DEXA devices were originally developed to measure bone mass in human patients, recent studies (Brunton et al 1993; Svendsen et al, 1993; Mitchell et al, 1997) have shown that DEXA is a potentially acceptable means of measuring bone mass content, bone density, fat tissue, and lean body mass in chickens.

#### Pantothenic Acid Metabolism

Nutrient deficiency of dihydroxy- $\beta$ - $\beta$ -dimethylbutyrl- $\beta$ alanine or pantothenic acid was first described by Norris and Ringrose, (1930). Symptoms of deficiency in chickens,

as described by Scott (1982), include subcutaneous hemorrhage and severe edema in embryos, severe dermatitis, broken feathers, perosis, reduced growth, reduced feed intake and mortality. Chicks appear emaciated and definite crusty scabs, like lesions appear in the corners of the mouth and eyes. The outer layers of skin (feet toes, footpad) peel off and cracks and fissures appear, protuberances develop on the balls of the feet. The liver is hypertrophied and yellow in color, nerves and spinal cord fibers show myelin degeneration to the lumbar region. PA deficient chicks exhibit necrosis in the bursa of fabricius and the thymus, and a lymphatic paucity in the spleen. The national research council (NRC) sets Pa requirement at 10 ppm to achieve optimum performance for all poultry species (1994).

Pantothenic acid is an essential vitamin, used as a component of coenzyme A, however pantothenic acid is not stored in significant amounts in the body. The highest concentrations of PA are found in the liver and kidneys. Serum pantothenic acid is transported primarily in the erythocytes as Coenzyme A. Synthetic PA occurs in two forms d and L and is sold as dL pantothenate or as racemix. The racemix has approximately half of the availability of dl-pantothenate. PA is typically supplemented in broiler diets as d-calcium -pantothenate (Hoffman-La Roche).

Pantothenic acid is an important B vitamin involved in the synthesis of co-enzyme A. Co-enzyme A functions as a carrier of acyl groups as thioesters and is important in fatty acid and acetate metabolism. (Mclure 1996)

Studies have shown that supplementing pantothenic acid above 10 ppm does not improve broiler performance under normal stress conditions. (Deyhim et al 1992; Harms and Nelson, 1992)

#### Magnesium

Magnesium (Mg) is the eleventh most abundant element in the body and the fourth most abundant cation, (Britton et al. 1989). Mg is an essential metal ion catalyst in many phosphorylation reactions. Serum concentrations of Mg when supplemented for maximal growth is 0.72 mg/dL, (Scott 1982). The NRC (1994) suggests that 600 ppm dietary Mg is sufficient to support maximal growth.

Newly hatched chickens consuming diets devoid of Mg live only a few days, further Mg deficiency in hens results in rapid decline of egg production. Phosphorus and calcium have been shown to antagonized Mg absorption, and exacerbate deficiency. Mg deficient rats had 125% the BMR of control rats. Serum contains only about 50mg/L compared to soft tissue such as liver, striated muscle, kidney and brain which contain 430-540 mg Mg/kg. The concentration of serum mg increased in non-hibernating and cold-blooded animals

when body temperature is artificially lowered, and in hibernating animals during hibernation. About one-half of the total body Mg is in the bones (Scott 1982).

Magnesium aspartate hydrochloride has been shown to reduce weight loss in heat stressed hens, (Donoghue et al 1990). The mechanism by which the magnesium has acts has still not been defined. Hypermagnesemia has been shown to cause hypotension, and peripheral paralysis. Magnesium is thought to act as a necessary chelating agent for various enzymes which depend on mg for proper function. These enzymes include ribosome, acetylcholine receptors, Adenosinetriphosphate, (Aikawa, 1971; Wacker, 1980; Britton et al, 1989).

#### Restricted Feeding

Unrestricted feeding of broiler breeder hens results in poor egg quality, poor egg production and overall poor performance. While limit fed breeders initiate daily lay at first light, full fed hens lay in erratic patterns during the day. Feed restricted hens lay in sequences of 40 consecutive days on average before skipping a day. Full-fed hens experience shorter laying cycles than restricted fed hens and go into first molt at a younger age. Fertility and embryonic survival rates are lower for

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#### CHAPTER III

### INFLUENCE OF STARTER PROTEIN AND FEEDING CURVE ON BODY COMPOSITION OF BROILER BREEDER HENS

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#### Abstract

Two experiments were conducted on Cobb broiler breeder hens to examine the effect of energy, protein, feeding curve, and daily feed intake during lay on body weight, body composition and egg production. In the first experiment higher starter protein increased body weight 3978.2g vs. 3936.5g (P < 0.05) and reduced fat tissue mass 545.8g vs. 586.3 (P < 0.05), increased lean tissue mass 2897.3g vs 2849.9g (P < 0.05), and higher bone tissue mass 77.5g vs 75.0g (P < 0.05), at the end of the laying period. In the second experiment birds receiving 24,804 kcal by 20 weeks had higher body weight 2232.9g vs. 2113.6g and 1946.9g (P <

1538.5g (P < 0.05). These studies suggest that dietary energy, protein, and feeding regimes delivered to the pullet prior to laying play an important role in determining the bird composition.

(Keywords: X-ray, protein, energy, body composition, fat, lean, bone, and egg production)

#### Introduction

The breeding hen is one of the most crucial factors influencing today's poultry industry. Previous studies have examined changes in body-weight and proximate analysis of broiler breeder hens at various stages of production. (Attia, 1995; Pearson and Herron, 1979; Pearson and Herron 1980; Pearson and Herron, 1981; Pearson and Herron 1982) However, limited information is available relating live body composition changes to performance and subsequent egg production. Studies on body composition (lean tissue, bone, and fat tissue) have difficulty in interpreting the actual effects of each tissue accumulation percentage on production and reproductive performance of the chicken. The energy buffering effect of body fat tissue acts to masks dietary and feeding effects (Kwakkel 1997).

Just how important increased protein is in the

increased egg size, Pearson and Herron (1981) did not observe any effect of increased protein on breeder reproductive performance. However, a decrease in hatchability was noted when the protein to energy ratio exceeded 15g to 1 MJ metabolizable energy(ME). A 20g to 1 MJ ME ratio was found to decrease egg production. (Pearson and Herron, 1982) Radcliff (1978) reported adequate protein content more important than protein intake, for maximal protein deposition in Zucker rats. Lopez (1994) detected no benefit to feeding above 21 g crude protein(CP)/day to breeders after 45 weeks of age for egg weight, and feeding 30g CP/hen/day reduced egg production. There is no benefit to excess protein after the hen has matured. Deficient dietary protein during early development may reduce egg production even though growth may appear normal. Bartov et al (1994) found hens had lower egg production when low protein diets were fed in the first 6 weeks.

Dietary energy content has a high influence on body composition of the broiler breeder hen. Dietary energy is closely correlated to both egg production and body-weight, as a result (Pearson and Herron 1981) body-weight is a good indicator of expected hen egg production. Information regarding relationships between body composition and breeder performance over the laying cycle is scarce. Pearson and Herron (1981) observed that increased dietary energy levels

resulted in increased body-weight, carcass fat, water content, and egg weight, but fertility was decreased. Too little energy in the diet can also reduce performance, for breeder hens receiving a diet containing 1.13 MJ apparent ME/bird/day, the onset of lay was delayed and occurred at a lower body-weight than birds with higher dietary energy levels (Pearson 1982).

Geneticists have labored to produce uniform breed lines, yet there remains a degree of individual variation among breeder lines. Maintenance requirement and heat production values vary between birds with higher percentage body fat and birds with lower percentage body fat, the latter having higher heat production values (Radcliff 1978). Pullar (1977) found heat production is closely related to lean tissue weight in Zucker rats. Increased starter energy levels to 4 weeks of age did not affect abdominal fat at 7 weeks in broilers (Kubena 1974). Birds raised on the same diets may appear uniform but have very different body compositions and production levels.

The role of dietary energy and protein on bone structure of the breeder hen is of critical importance for natural breeding operation. Excess dietary protein causes exogenous loss of calcium (Barzel 1995). Heaney (1998) found that excess protein intake is not harmful to bone development as long as dietary calcium is adequate and found

20mg calcium to 1g protein intake to be the optimum ratio in humans. Osteoporosis is a consistent problem in high producing laying hens. Several studies have examined the cause of bone loss in hens (Couch 1955; Urist and Duetsch 1960; Bell and Siller 1962; Wilson et al. 1992). Rennie (1997) found no response in bone loss to treatments of low phosphorus, low protein and high vitamin K, oystershell, fluoride, and 1,25-dihydroxycholecalciferol in Hisex laying hens. Past studies have relied on sacrificing breeders at various stages of production, new techniques may allow monitoring of changes in bone tissue throughout the life cycle.

Dual energy X-ray absorptiometry (DEXA) is a potentially accurate means of non-invasive determination of live body composition. Mitchell (1997) found that DEXA was most accurate for chickens, when the small animal scanning software was used. Fat percentage was overestimated by 4%, lean mass was highly correlated to body protein ( $R^2=0.90$ ) and body water ( $R^2=0.93$ ). Bone mass was correlated with total body ash ( $R^2=0.77$ ) for all chickens scanned. Svendsen (1993) found DEXA to be an accurate method for predicting soft tissue body composition, and total body bone mass. DEXA overestimated fat percentage and underestimated lean and bone tissue values for large pigs in the pediatric scanning mode (Brunton, 1993). Research continues to expand

the DEXA database allowing the effects of dietary energy, protein and feed restriction on the breeder performance.

Studies have been conducted to relate weight gain to egg production in broiler breeders. (Robinson, 1993; Yu et al, 1992). Yu (1992) observed that the breeder broiler must be limit feed during rearing phase to achieve optimum egg production and survivability. Pearson and Herron (1979), suggested that restricting feed to broiler breeder in early lay reduces body-weight gain, and subsequent maintenance requirement of hens during the later laying period. If the maintenance requirement is reduced, more of the dietary nutrients are available to be used for egg production instead of homeostasis.

This study was designed to determine the body composition of the hen throughout the production cycle. Hens were exposed to varying levels of nutrition, feed restriction and stocking density in an effort to evaluate performance.

#### Materials and methods

Two experiments were conducted simultaneously utilizing 2900 Cobb pullets in 56 floor pens. Experiment 1 contained eight treatments in a 2x2x2 factorial complete block design. Treatments (TRT) for experiment 1 were as follows: TRT 1 Stocking density 1.3 ft per bird (55

chicks/pen), TRT 2 Stocking density of 1.46 ft per bird (50 chicks per pen), TRT 3 Linear feeding curve, TRT 4 Sigmoid feeding curves, (linear vs Sigmoid feeding curves were designed by John May), TRT 5 21% protein starter ration to six weeks of age TRT 6 18% protein starter ration to six weeks of age. Experiment 2 contained seven treatments in a complete block 2x3x2 design. Treatments for experiment 2 were as follows: TRT 1. 21% Crude protein starter ration to six weeks of age, TRT 2. 18% Crude protein starter ration to six weeks of age, TRT 3. Cumulative calorie consumption of 21,996 Kcal per hen at 20 weeks of age, TRT 4. Cumulative calorie consumption of 23,400 Kcal per hen at 20 weeks of age, TRT 5. Cumulative calorie consumption of 24,804 Kcal per hen at 20 weeks of age, TRT 6. Cobb 500 recommended amount of daily feeding allotment after 20 weeks, and TRT 7. Five percent above Cobb 500 recommended daily amount of daily feeding allotment after 20 weeks. Variables monitored for both experiments included live body weight, DEXA<sup>1</sup> body composition scans, and egg production.

Three birds per pen were selected for body composition analysis and followed from 8 to 48 weeks of age. On a typical scanning day breeders were collected at approximately 3 o'clock each day and fasted over night. During the fasting period birds were monitored for oxygen consumption and carbon dioxide production in order to

<sup>&</sup>lt;sup>1</sup> Hologic Bone densiotometer QDR1000/w using rat whole body analysis algorythm.

estimate heat production as described by Belay and Teeter (1993). Birds were anesthetized using .4 mg per Kg ketamine intramuscular injection, and Isoflourene volatilized in medical grade oxygen gas at 5L per minute. The isoflourene was delivered via a cylindrical mask fasten around the head with elastic wrapping. The mask was darkened to minimize photo-stimulation and enhance the anesthetic effects. Birds were scanned using a Hologic® ODR1000/W Bone densitometer at 60 Hz, using the small rat whole body algorithms provided by Hologic® to calculate bone mass content, fat tissue mass, and lean tissue mass. Mechanisms for the Hologic® scanning system were described by Brunton, (1993). Scanning time was approximately 25 minutes per bird, after which birds were allowed a minimum of 30 minutes recovery time before being returned to pens. Daily egg production and egg weights were taken after onset of lay until the end of the trial. Eggs with double yolks were identified as being eggs whose weight exceeded 90 grams.

Statistical analysis was performed on the data using the general linear models of SAS (1997). Data was analyzed for correlations using Proc Corr procedures of SAS (1995) to provide a more extensive look at relationships between variable.

#### Results and Discussion

In experiment 1 all variables: SD, FC, and SP had significant effect on body weight up to 20 weeks of age (P < 0.01) (table 3). Birds raised at lower SD had greater body weights over all treatments (1314 vs 1289g; P < 0.01), birds fed high SP (1320g vs. 1283g; P < 0.01) had higher body weight, and birds fed sigmoid FC diets had higher body weights (1322g vs. 1282g; P > 0.01). The interaction between SD and SP was largely due to the high SP birds having heavier body weights. After 20 weeks of age SD, SP and FC had no significant effects on body weight.

Lean tissue mass in experiment 1, (table 5), was affected by SD (P > 0.1), with the low density having higher initially at week 8, the higher density birds had greater lean mass on weeks 16, 20, 24, and 28 with no significant differences between treatments beyond week 28. Birds on the sigmoid FC had higher lean tissue mass up to 20 weeks (P < 0.05), after onset of lay however the linear birds had numerically higher lean tissue mass on and after week 28, and significantly higher lean mass on weeks 32 and 36 (P < 0.05). Birds feed the starter ration had a numerically higher lean mass throughout the trial with significant differences on weeks 8, 12, 16 40, 44, and 48. The SP x FC interaction was significant with the high SP birds having the higher lean tissue mass for both linear and sigmoid

diets (2050 and 2054g) than birds fed the low protein starter (2015 and 2039 g) with (P < 0.05). Similarly the SP x SD interaction was significantly higher for the high SP ration (2056 and 2048 g) than the low SP (2014 and 2041g; P < 0.01), the high density and low density SD'S were not significantly different for the high SP diets. This suggests the importance of early nutritional protein on lean tissue accretion.

Grams of fat tissue were significantly higher for the high density treatment up to 24 weeks of age in experiment one, (table 6). Fat tissue mass was greater for the low density bird treatment at weeks 32, 36, 40, 44, and 48 (P < 0.05). Fat tissue mass for breeders on sigmoid FC was significantly lower for weeks 12, 16, 20, 24, 28, and 32 (P < 0.05), (table 7). Over all sigmoid FC treatments had lower fat tissue mass than the linear treatments (303 vs 319; P < 0.01). The high SP birds had significantly less fat tissue at weeks 20, 24, 28, 32, 36, 40, 44, and 48 (P < 0.05).

Experiment 1, bone mass content was significantly different for the high SP at 8 weeks (15.7 vs 14.6g; P < 0.05) and during weeks 44 and 48. (table 9) Sigmoid fed hens and low density treatment birds had higher bone mass content from week 12 until the onset of lay at week 24 (P < 0.05). Low SD birds did have higher bone mass at weeks 44

and 48 (P < 0.05), where as the sigmoid birds were not significantly different beyond week 24, (table 8). SD x FC interactions were significant, linear and sigmoid treatments were significantly different in the high density treatments (51.8 vs 51.6g; P < 0.53) not in the low SD (50.0 vs 51.1g; P < 0.01). In the SD x SP interaction, the low density SD treatments had a higher bone mass content regardless of SP, the high SP had significantly more bone mass than the low SP birds (P < 0.05). In the feeding x SP only the sigmoid birds on high SP had significantly higher bone mass (51.7 g; P < 0.01). These results indicate that protein has a less important role on bone mineral deposition than the amount of nutrition being fed to the bird.

Egg production was significantly increased in the low density SD trt at week 32 in experiment one, (Table 11). Significant SD x SP ration interactions indicate high SP decreased mean egg production in the low density treatments (48.1 vs 51.5%) and increased mean egg production in the high density treatments (54.2 vs 51.9%) with (P < 0.01). Starter nutrition decreased mean egg production for the sigmoid feeding treatments (50.0 vs 52.1%) with (P < 0.05). SP levels, and FC had no effect significant on egg weight. The Low SD increased egg weight significantly in the low SP birds (61.6 vs 60.8 g; P < 0.03). The high SD low SP birds

had the lightest egg weight (60.8g) the other treatments were not significantly different.

In experiment 2, body weight increased with both Kcals consumed and SP level to 20 weeks of age (P < 0.01), (table 4). There was no significant SP x CAL interaction. After 20 weeks of age CAL intake had the only significant main effect with body weight increasing with CAL intake (P < 0.01). The amount x CAL interaction increased body weight for the 23,400 Kcal treatments (P < 0.01), but not for the higher of lower CAL value. It would seem that the bird receiving the 23,400 Kcal were best adapted to utilize the increased amount of feed. There was a significant SP by CAL interaction where the increased SP increased body weight for the 21,996 Kcal birds, and decreased body weight for the 23,400 Kcal birds. (3079 vs 3009g; P < 0.01 and (3130 vs 3198g; P < 0.01) respectively.

Fat tissue mass, in experiment 2, increased with CAL consumed to 20 weeks (P < 0.05) The 23,400 Kcal treatment hens had the highest amount of fat tissue during weeks 24, 28, 32, 36, 40, 44, and 48 (P < 0.05), (table 8). High SP had significantly lower fat tissue at 8 weeks and significantly higher fat content on weeks 12 to 28 (P < 0.05). The amount of feed after 20 weeks had no significant effect on body fat tissue mass.

In experiment 2, the birds with starter diets had significantly higher lean tissue mass at weeks (8, 12 and 16), lean tissue mass significantly increased with increase in CAL consumption to 20 weeks of age (table 6). Although the SP x CAL x week interaction was significant (P < 0.01), only birds on the highest CAL intake had significantly increased lean tissue due to SP. This indicates that growing birds must have sufficient energy from the diet to make efficient use of nutritional SP up to 20 weeks of age. After 20 weeks only the CAL treatments had significant effects on bird lean tissue mass. Birds consuming 21,996, 23,400, and 24,804 Kcal had (2326, 2411, and 2482g; P < 0.01) grams of lean tissue. The amount x CAL interaction was significant, the 23,400 CAL treatments had significantly higher lean tissue on the high amount ration than the low amount fed (2425 and 2398 g; P < 0.01). The 23,400 Kcal birds were able to utilize the increased amount of feed towards lean tissue accretion.

In experiment 2 high SP treatments had significantly higher bone mass up to week 12. (Table 9) Bone mass was increased at each level of CAL treatment throughout the growing stage (0-20 weeks) and up until week 42 with the 24,804 CAL treatment significantly higher than the middle and low CAL treatments at week 44, (P < 0.05). Only the 23,400 CAL treatment increased significantly with the high

SP starter (28.1 vs 26.5 g; P < 0.05). The high CAL and low CAL treatments showed no significant differences with the increased SP (28.5 vs 28.3 g) and (24.7 vs 24.4 g) respectively. It seems the low CAL birds did not have the energy resources to utilize the extra protein, and the high CAL birds saw no benefit to the extra protein. After 20 weeks the high SP increased the over all bone mass at the low and middle CAL levels (58.4 vs 56.3 g) and (60.3 vs 59.0 g) respectively at P < 0.05, increased SP decreased bone mass in the high CAL treatment birds (60.2 vs 62.3 g) at (P < 0.05).

In experiment two, the 23,400 kcal treatment birds had significantly higher egg production on weeks 24 and 28 compared to the 21,996 and 24,804 kcal treatments. (table 12.) The 21,996 kcal hens had significantly higher egg production at 48 weeks probably due to delayed peak production as they were the lowest producing treatments at onset of lay. Amount of daily feed increased mean egg production for the 21,996 kcal treatment (52.6% vs 48.7%; P < 0.05) and decreased egg production for the 23,400 Kcal treatment (53.3% vs 55.6%; P < 0.05). High SP treatments had significantly lower egg production on week 36, (table 11). High SP decreased mean egg production for Low CAL (49.5% vs 51.8%; P < 0.05) and high CAL (51.7% vs 54.6%; P < 0.05) treatments.

The Low SD increased egg weight significantly in the low SP birds (61.6 vs 60.8 g; P < 0.03). The high SD low SP birds had the lightest egg weight (60.8g) the other treatments were not significantly different.

Body weight from week 12 to week 32 was positively correlated to egg production to 32 weeks ( $R^2 = .51$ ; P < 0.01), at 40-44 weeks body weight was negatively correlated ( $R^2 = -.49$ ; P < 0.01).

Body weight gain up to 20 weeks was positively correlated with egg weight, however body weight at weeks 24-48. Lean tissue mass at 12-36 was positively correlated with egg production ( $R^2$ = .47; P < 0.05). Lean tissue gain from 12-20 weeks correlated with egg production ( $R^2$ = .54; P < 0.01). Pullets under go a pubertal growth phase at 19 weeks, of which 40-70% is reproductive tract related. A certain amount of fat free tissue (CP, ash and water) seems to be critical for the initiation of maturity and onset of lay (Kwakkel 1995).

Body fat content at 8 weeks was positively correlated  $(R^2 = .29; P < 0.04)$  to egg production at 20-28 weeks of age. Egg production was positively correlated  $(R^2 = .41; P < 0.01)$  with bone tissue mass gain at 16-20 weeks. SP, and FC had no significant effect on egg weight. Bone mass content gain from 12 to 20 wks was positively correlated to egg production (.40 P < .01) from 20 to 28 wks of age. Lean

production (.40 P < .01) from 20 to 28 wks of age. Lean tissue gain from 12 to 16 wks of age was positively correlated to egg production from 20 to 28 wks of age (R2= .66 P < .01). BW gain from 8 to 16 wks was positively correlated to egg production from 20 to 28 wks of age (R2= .62 P < .01). BW gain from 8 to 20 wks of age was positively correlated to egg production from 20 to 28 wks of age (R2= .56 P < .01).

Heat production data was collected on the breeder broiler hens during the fasting period.(table 19) It was assumed that broilers would be near their basal metabolic state. During weeks 10, 11, and 12 basal heat production was 24.9072, 26.7705, and 26.7445 Kilojoules per kilogram of body weight. During week 32 heat production was 43.4729 Kilojoules per kilogram of tissue.

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Ingredients	Starter	Developer	Layer
		(%)	
Com	64.3	68.345	72.81
Soy Bean Meal	18.9	15.5	17.0
Wheat Middlings	13.78	12.4	0.0
Dicalcium phosphate	1.8	1.85	7.5
Limestone	0.625	0.625	1.4
Vitamin Premix <sup>1</sup>	0.2	0.1	0.1
Methionine	0.05	0.08	0.09
Salt	0.25	0.1	0.1
Trace Mineral	0.1	1.0	1.0
Total	100.00	100.00	100.00
Calculated Analysis			
ME Kcal/Kg	2866.6	2917.2	2915
Crude protein (%)	16.5	15	16
Calcium (%)	.90	0.90	3.25
Phosphorus (% av)	0.45	0.47	0.42

### Table 1. Composition of low protein diets.

<sup>1</sup> Premix contained per kilogram: 22,050,000 I.U vitamin A, 7,452,900 I.U vitamin D3, 50,936 I.U vitamin E, 40 mg vitamin B12, 18,081 mg Riboflavin, 11,300mg Pantothenic Acid, 121,496 mg Niacin, 6,042 mg Menadione, 2,597 mg Folic Acid, 8,379 mg Pyridoxine, 5,557 mg Thiamine, 267 mg Biotin 1%.

Ingredients	Starter	Developer	Layer
		—— (%) —	
Corn	56.45	61.62	74.52
Soy Bean Meal	30.9	16.7	17.2
Wheat Middlings	9.5	18.6	0.0
Dicalcium phosphatel	1.5	1.2	0.7
Limestone	1.08	1.38	7.0
Vitamin Premix <sup>1</sup>	0.1	0.1	0.1
Methionine	0.17	0.1	0.18
Salt	0.2	0.2	0.2
Trace Mineral	0.1	0.1	0.1
Total	100.00	100.00	100.00
Calculated Analysis			
ME Kcal/Kg	3047	2794	2849
Crude protein (%)	21	18	16
Calcium (%)	1.00	1.00	1.00
Phosphorus (% av)	0.40	032	0.30

#### Table 2. Composition of high protein diets.

<sup>1</sup> Premix contained per kilogram: 22,050,000 I.U vitamin A, 7,452,900 I.U vitamin D3, 50,936 I.U vitamin E, 40 mg vitamin B12, 18,081 mg Riboflavin, 11,300mg Pantothenic Acid, 121,496 mg Niacin, 6,042 mg Menadione, 2,597 mg Folic Acid, 8,379 mg Pyridoxine, 5,557 mg Thiamine, 267 mg Biotin 1%.

				Bo	ody Mass (g	2			
4	16	20	24	28	32	36	40	44	48
ity									
375.4ª	2122.4ª	2182.7*	2545.2ª	2871.9°	3162.8°	3418.0ª	3637.4ª	3821.2 °	3969.1°
305.5 <sup>b</sup>	2151.5ª	2193.1ª	2564.3ª	2895.2ª	3185.8 *	3436.2 ª	3646.3 ª	3816.1 *	3945.6*
•									
303.6°	1771.1*	2182.2ª	2554.2°	2887.2ª	3181.2ª	3436.1 *	3651.9°	3828.7 *	3966.5 "
377.3 <sup>b</sup>	1795.0°	2193.6 ª	2555.2°	2879.8°	3167.5°	3418.1ª	3631.8 *	3808.5 °	3948.3 °
470.7°	2143.3 *	2196.9ª	2561.5°	2889.4ª	3180.5°	3435.0°	3652.8 *	3833.8*	3978.2 ª
402.2 <sup>b</sup>	2130.6 <sup>b</sup>	2178.9ª	2548.0ª	2877.7ª	3168.1 ª	3419.2°	3631.0 <sup>b</sup>	3803.4 <sup>b</sup>	3936.5 <sup>b</sup>
	4 ty 375.4 <sup>a</sup> 305.5 <sup>b</sup> 303.6 <sup>a</sup> 377.3 <sup>b</sup> 470.7 <sup>a</sup> 402.2 <sup>b</sup>	4 16   ty 375.4° 2122.4°   305.5° 2151.5°   303.6° 1771.1°   377.3° 1795.0°   470.7° 2143.3°   402.2° 2130.6°	4 16 20   ty 375.4° 2122.4° 2182.7°   305.5° 2151.5° 2193.1°   303.6° 1771.1° 2182.2°   377.3° 1795.0° 2193.6°   470.7° 2143.3° 2196.9°   402.2° 2130.6° 2178.9°	4162024ty $375.4^{a}$ 2122.4^{a}2182.7^{a}2545.2^{a} $305.5^{b}$ 2151.5^{a}2193.1^{a}2564.3^{a} $303.6^{a}$ 1771.1^{a}2182.2^{a}2554.2^{a} $377.3^{b}$ 1795.0^{a}2193.6^{a}2555.2^{a} $470.7^{a}$ 2143.3^{a}2196.9^{a}2561.5^{a} $402.2^{b}$ 2130.6^{b}2178.9^{a}2548.0^{a}	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	4162024283236ty 375.4° 305.5°2122.4° 2151.5°2182.7° 2193.1°2545.2° 2564.3°2871.9° 2895.2°3162.8° 3185.8°3418.0° 3436.2°303.6° 303.6° 377.3°1771.1° 1795.0°2182.2° 2193.6°2554.2° 2555.2°2887.2° 2879.8°3181.2° 3167.5°3436.1° 3418.1°470.7° 402.2°2143.3° 2130.6°2196.9° 	416202428323640ty 375.4°2122.4° 2151.5°2182.7° 2193.1°2545.2° 2564.3°2871.9° 2895.2°3162.8° 3185.8°3418.0° 3436.2°3637.4° 3637.4° 3646.3°303.6° 303.6°1771.1° 1795.0°2182.2° 2193.6°2554.2° 2555.2°2887.2° 2879.8°3181.2° 3167.5°3436.1° 3418.1°3651.9° 3631.8°470.7° 402.2°2143.3° 2130.6°2196.9° 2178.9°2561.5° 2548.0°2889.4° 2877.7°3180.5° 3168.1°3435.0° 3419.2°3652.8° 3631.0°	41620242832364044ty 375.4° 305.5°2122.4° 2151.5°2182.7° 2193.1°2545.2° 2564.3°2871.9° 2895.2°3162.8° 3185.8°3418.0° 3436.2°3637.4° 3646.3°3821.2° 3816.1°303.6° 303.6° 377.3°1771.1° 1795.0°2182.2° 2193.6°2554.2° 2555.2°2887.2° 2897.8°3181.2° 3187.8°3436.1° 3436.1°3651.9° 3651.9° 3631.8°3828.7° 3808.5°470.7° 402.2°2143.3° 2130.6°2196.9° 2178.9°2561.5° 2548.0° 2548.0°2889.4° 2877.7° 3168.1°3435.0° 3418.1°3652.8° 3652.8° 3631.0° 3803.4°

Table 3. Influence of starter protein, Stocking density, and feeding regime on body mass.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

	Body Mass (g)													
Age (week)	4	16	20	24	28	32	36	40	44	48				
Amount										•••				
High				2464.2 ª	2797.6°	3100.7 °	3373.5ª	3616.1 °	3828.3ª	4010.3ª				
Low				2457.9°	2790.2ª	3091.8*	3362.8 ª	3603.2 °	3813.0°	3992.2°				
Calories														
21996	324.6 <sup>b</sup>	1572.0°	1946.9 °	2301.3°	2635.2 °	2948.7 °	3241.6°	3514.2°	3766.2 <sup>b</sup>	3997.8°				
23400	337.8 <sup>b</sup>	1717.6 <sup>b</sup>	2113.6 <sup>b</sup>	2477.6 <sup>b</sup>	2809.7 <sup>b</sup>	3109.8 <sup>b</sup>	3377.9 <sup>b</sup>	3614.0 <sup>b</sup>	3818.2 <sup>b</sup>	3990.4ª				
24804	358.1ª	1822.6 ª	2232.9°	2604.3ª	2936.8ª	3230.4 ª	3485.0ª	3700.8ª	3877.6 *	4015.6 ª				
Starter Prote	in													
21%	360.8 "	1712.3 <sup>ª</sup>	2103.2°	2464.4ª	2795.7ª	3097.2ª	3368.9°	3610.8°	3823.0*	4005.3ª				
18%	319.5 <sup>b</sup>	1695.8 <sup>b</sup>	2092.4ª	2457.8ª	2792.1°	3095.4ª	3367.5ª	3608.5ª	3818.4ª	3997.2ª				

Table 4. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on body weight.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

	_					<u>Bod</u>	y Lean Tiss	ue Mass (g	2			
	Age (week)	8	12	16	20	24	28	32	36	40	44	48
	Stocking densit	y										
	1.40 ft <sup>2</sup> / bird	691.8ª	1015.2ª	1353.7 <sup>b</sup>	1678.8 <sup>b</sup>	1972.8 <sup>b</sup>	2225.7 <sup>b</sup>	2433.9°	2598.3ª	2722.7°	2812.7°	2874.7ª
	1.36 ft <sup>2</sup> / bird	646.6 <sup>b</sup>	1016.3ª	1377.9°	1711.4ª	2004.7 ª	2252.4 ª	2453.8 *	2611.4 ª	2729.9°	2815.1°	2872.5°
	Feeding											
	Linear	654.7 <sup>b</sup>	980.8 <sup>b</sup>	1333.4 <sup>b</sup>	1675.5 <sup>b</sup>	1983.6ª	2245.3 °	2456.0°	2617.1°	2733.8°	2813.5 ª	2864.0ª
	Sigmoid	683.7 ª	1050.8ª	1398.2 *	1714.8 ª	1993.9°	2232.8 ª	2431.7 <sup>b</sup>	2592.6 <sup>b</sup>	2718.8ª	2814.3 ª	2883.3 °
	Starter Protein											
35	21%	686.4ª	1034.2 *	1380.4 °	1704.7 ª	1994.3 ª	2242.8ª	2448.3ª	2612.3°	2738.6 <sup>b</sup>	2831.8ª	2897.3ª
	18%	652.0 <sup>b</sup>	997.4 <sup>b</sup>	1351.2 <sup>b</sup>	1685.6 ª	1983.2 ª	2235.4 ª	2439.4 ª	2597.3ª	2714.0ª	2796.0 <sup>b</sup>	2849.9 <sup>b</sup>

Table 5. Influence of starter protein, Stocking density, and feeding regime on body lean tissue mass.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05). <sup>C-D</sup> Means within columns with no common superscript differ significantly (P < .10).

	Body Lean Tissue Mass (g)												
Age (week)	8	12	16	20	24	28	32	36	40	44	48		
Amount													
High				1655.2ª	1956.6*	2215.8ª	2429.2ª	2597.2ª	2723.5°	2813.7°	2874.6ª		
Low				1651.4ª	1952.4ª	2211.2ª	2424.2ª	2592.3 *	2719.6 ª	2811.9ª	2875.9°		
Calories													
21996	580.5°	909.0°	1231.5°	1538.5°	1822.7 °	2079.5°	2305.8°	2500.5°	2663.6°	2796.4 <sup>b</sup>	2900.8ª		
23400	642.9 <sup>b</sup>	977.4 <sup>b</sup>	1330.3 <sup>b</sup>	1670.1 <sup>b</sup>	1976.1 <sup>b</sup>	2236.5 <sup>b</sup>	2446.4 <sup>b</sup>	2606.3 <sup>b</sup>	2720.5 <sup>b</sup>	2795.7 <sup>b</sup>	2840.1 <sup>b</sup>		
24804	681.5°	1027.6ª	1397.5 ª	1751.4ª	2064.6ª	2324.6°	2527.8 ª	2677.4ª	2780.5 ª	2846.4 ª	2885.0°		
Protein													
21%	646.4 ª	985.6ª	1329.3°	1655.7 °	1950.5 ª	2205.6ª	2417.7°	2587.4 *	2717.6*	2813.0°	2879.3°		
18%	623.5 <sup>b</sup>	957.0 <sup>b</sup>	1310.2 <sup>b</sup>	1650.9*	1958.4 *	2221.4ª	2435.7 °	2602.1ª	2725.4°	2812.6 ª	2871.2 ª		

### Table 6. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on lean body tissue mass.

<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

	_					Bod	ly Fat Tissu	e Mass (g)				
	Age (week)	8	12	16	20	24	28	32	36	40	44	48
9	Stocking Densit	ty	2007 F102 - 54174								141 (1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1-1	
	1.40 ft <sup>2</sup> / bird	69.3ª	131.9°	177.4ª	216.8°	257.0°	301.4 *	350.4 <sup>b</sup>	403.0 <sup>b</sup>	456.7 <sup>b</sup>	508.4 <sup>b</sup>	554.9 <sup>h</sup>
	1.36 ft <sup>2</sup> / bird	84.6 <sup>b</sup>	108.2 <sup>b</sup>	140.8 <sup>b</sup>	183.4 <sup>b</sup>	235.0 <sup>b</sup>	293.6°	356.1ª	419.1ª	479.1ª	532.8 ª	577.1ª
	Feeding Curve											
	Linear	71.5ª	133.9°	179.2°	219.2°	261.0ª	307.8ª	359.6ª	414.9ª	470.5ª	522.9°	568.4ª
	Sigmoid	82.4 ª	106.1 <sup>b</sup>	139.0 <sup>b</sup>	181.0 <sup>b</sup>	231.1 <sup>b</sup>	287.2 <sup>b</sup>	346.9 <sup>b</sup>	407.2ª	465.3ª	518.3 °	563.6ª
	Starter Protein											
L	21%	77.1ª	120.2 *	156.7ª	193.9 <sup>b</sup>	235.8 <sup>b</sup>	283.4 b	336.1 <sup>b</sup>	391.7 <sup>b</sup>	447.3 <sup>b</sup>	499.7 <sup>b</sup>	545.8 <sup>b</sup>
	18%	76.8ª	119.9ª	161.5 *	206.3ª	256.3ª	311.5ª	370.4ª	430.5ª	488.6ª	541.5°	586.3ª

Table 7. Influence of starter protein, Stocking density, and feeding regime on body fat tissue mass.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05). <sup>C-D</sup> Means within columns with no common superscript differ significantly (P < .10).

	Body Fat Tissue Mass (g)													
Age (week)	8	12	16	20	24	28	32	36	40	44	48			
Amount High Low					217.5° 216.3 °	258.4° 256.7°	313.0° 310.6°	380.4 ° 376.8 °	457.4 452.1	539.5° 531.9°	621.6° 611.0°			
Calories 21996 23400 24804	63.2 <sup>b</sup> 62.3 <sup>b</sup> 83.6ª	97.7° 124.6 <sup>b</sup> 167.9°	127.5° 164.1 <sup>b</sup> 196.7°	158.7° 197.0 <sup>b</sup> 206.9°	195.5° 233.9ª 221.4 <sup>b</sup>	240.4° 280.3° 251.9 <sup>b</sup>	294.9 <sup>b</sup> 338.3 ° 302.1 <sup>b</sup>	359.3 <sup>b</sup> 407.0 <b>*</b> 369.5 <sup>b</sup>	432.9 <sup>b</sup> 483.4 <sup>°</sup> 448.0 <sup>b</sup>	514.6 <sup>b</sup> 563.4 <b>*</b> 529.2 <sup>b</sup>	602.5 <sup>b</sup> 642.1 <sup>ª</sup> 604.3 <sup>b</sup>			
Starter Prote 21% 18%	in 67.9 <sup>b</sup> 71.6 °	134.4 ° 125.7 <sup>b</sup>	171.7 ° 153.9 ⁵	199.1 ° 176.1 °	228.9 ° 204.9 <sup>b</sup>	268.2 ° 246.9 b	319.6 ª 304.0 ª	382.6 ° 374.6 °	454.6° 455.0°	531.5° 540.0°	608.8 ° 623.8 °			

## Table 8. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on lean body tissue mass.

<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

	Body Bone tissue Mass (g)												
Age (week)	8	12	16	20	24	28	32	36	40	44	48		
Stocking Densit	ty												
1.40 ft <sup>2</sup> / bird	15.4°	24.5ª	33.2°	41.2°	48.7ª	55.4ª	61.3ª	66.4ª	70.8ª	74.4 ª	77.3°		
1.36 ft <sup>2</sup> / bird	14.8 ª	22.2 <sup>b</sup>	30.7 <sup>b</sup>	39.5 <sup>b</sup>	47.8 <sup>b</sup>	55.1 ª	61.3ª	66.4 <sup>a</sup>	70.3 ª	73.2 <sup>b</sup>	75.2 <sup>b</sup>		
Feeding													
Linear	15.0ª	22.6 <sup>b</sup>	31.1 <sup>b</sup>	39.6 <sup>b</sup>	47.7 <sup>b</sup>	55.0°	61.3ª	66.5 °	70.8ª	74.1 <sup>a</sup>	76.5ª		
Sigmoid	15.3 ª	24.1ª	32.9°	41.0°	48.8 ª	55.5°	61.4 ª	66.3 ª	70.3 ª	73.5ª	75.9ª		
Starter Protein													
21%	15.7 *	23.7°	32.0°	40.0ª	47.9ª	54.9°	61.1ª	66.4ª	71.0ª	74.6ª	77.5ª		
16%	14.6 <sup>b</sup>	23.0°	31.9ª	40.6 ª	48.6 ª	55.6ª	61.6ª	66.4ª	70.2ª	73.0 <sup>b</sup>	75.0 <sup>b</sup>		

Table 9. Influence of starter protein, stocking density, and feeding regime on body bone tissue mass.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05). <sup>C-D</sup> Means within columns with no common superscript differ significantly (P < .10).

	Body Bone tissue Mass (g)												
Age (week)	8	12	16	20	24	28	32	36	40	44	48		
Amount													
High	15.2°	22.9°	30.7ª	38.3 ª	45.6 ª	52.2ª	58.3°	63.8ª	68.6°	72.7 °	76.3ª		
Low	15.1 "	22.8ª	30.6 ª	38.2 ª	45.4 ª	52.1ª	58.2ª	63.6 ª	68.4 ª	72.6ª	76.2ª		
Calories													
21996	13.2°	21.1°	28.4 °	35.4 °	42.1 °	48.5 °	54.8°	60.9°	66.8 °	72.5 <sup>D</sup>	77.9ª		
23400	15.9 <sup>b</sup>	23.2 <sup>b</sup>	31.1 <sup>b</sup>	39.0 <sup>b</sup>	46.5 <sup>b</sup>	53.3 <sup>b</sup>	59.2 <sup>b</sup>	64.3 <sup>b</sup>	68.5 <sup>b</sup>	71.8 <sup>D</sup>	74.5°		
24804	16.3ª	24.4 ª	32.5°	40.4 ª	47.9 <sup>ª</sup>	54.7 ª	60.7 ª	65.9 ª	70.2 ª	73.7 <sup>c</sup>	76.4 <sup>b</sup>		
Starter Protein													
21%	15.8°	23.2°	30.9°	38.5°	45.7ª	52.4 ª	58.5ª	64.0ª	68.7ª	72.8ª	76.3ª		
18%	14.5°	22.6°	30.5°	38.1ª	45.3ª	51.9ª	57.9ª	63.4 ª	68.2 °	72.5ª	76.2*		

## Table 10. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on lean body tissue mass.

<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

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	Egg production (%)											
Age (week)	24	28	32	36	40	44	48					
Stocking Densi	ty											
1.40 ft <sup>2</sup> / bird	5.6*	56.6ª	62.4 <sup>b</sup>	60.2ª	54.9ª	52.7ª	47.5ª					
1.36 ft <sup>2</sup> / bird	8.6ª	62.6 ª	64.8ª	64.2ª	58.3°	55.1ª	49.7 ª					
Feeding Curve												
Linear	7.7ª	60.0ª	62.2ª	64.1ª	56.9ª	54.9°	47.8ª					
Sigmoid	6.5ª	59.2 *	65.0 °	60.3ª	56.3ª	52.9ª	49.4 ª					
Starter Protein												
21%	6.4ª	58.1ª	63.9ª	61.2°	56.7ª	54.5°	48.8ª					
18%	7.8ª	61.1°	63.3ª	63.3ª	56.5 ª	53.3 ª	48.4 ª					

Table 11. Influence of starter protein, Stocking density, and feeding regime on daily egg production.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

		Egg Production (%)											
Age (week)	24	28	32	36	40	44	48						
Amount													
High	7.8ª	58.3ª	61.9ª	60.9ª	57.0ª	53.2 °	47.1ª						
Low	5.2 ª	55.5ª	63.1ª	61.5 "	57.5°	52.3ª	48.7ª						
Calories													
21996	2.0°	49.4 °	62.7°	61.8ª	55.6°	52.9°	56.1ª						
23400	8.8ª	62.0ª	63.2°	61.6ª	58.2ª	54.7ª	48.9 <sup>b</sup>						
24804	8.7 <sup>b</sup>	59.1 <sup>b</sup>	61.0°	60.1 ª	57.9°	50.7 ª	38.7°						
Starter Protein													
21%	7.6ª	58.1 °	60.8 ª	57.4 <sup>b</sup>	56.4ª	52.3ª	47.7°						
18%	5.4ª	55.7ª	64.2ª	65.0 ª	58,0ª	53.3 °	48.2 ª						

Table 12. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on weekly egg production.

<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

	Egg Weight (g)								
Age (week)	24	28	32	36	40	44	48		
Stocking Dens	ity								
1.40 ft <sup>2</sup> / bird	48.6ª	55.8 <sup>b</sup>	59.6°	63.2ª	64.7°	65.7ª	65.6ª		
1.36 ft <sup>2</sup> / bird	44.8 <sup>b</sup>	59.0 ª	60.2 ª	63.9ª	65.5°	66.9ª	66.7ª		
Feeding Curve	9								
Linear	45.9ª	58.4ª	59.3°	63.4ª	64.9°	66.0ª	65.9ª		
Sigmoid	47.5ª	56.4ª	60.4ª	63.7ª	65.3 ª	66.6ª	66.4ª		
Starter Protein									
21%	44.6 <sup>b</sup>	59.1ª	59.8°	63.6ª	65.1ª	66.3ª	66.0ª		
18%	48.9 °	59.1°	60.0ª	63.5°	65.1°	66.2ª	66.2°		

Table 13. Influence of starter protein, Stocking density, and feeding regime on egg weight.

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<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

	Egg Weight (g)								
Age (week)	24	28	32	36	40	44	48		
Amount									
High	38.4ª	56.4ª	60.8ª	63.5ª	65.3ª	66.2ª	68.6ª		
Low	35.6 <sup>b</sup>	56.3 ª	59.8 ª	64.2ª	65.3ª	67.1 ª	67.6ª		
Calories									
21996	22.6°	56.2ª	59.6°	63.6 ª	64.5°	66.4ª	67.0ª		
23400	39.8 <sup>b</sup>	56.0ª	60.0ª	63.6ª	65.4 ª	66.6°	67.8ª		
24804	48.5ª	56.8ª	61.4ª	64.3ª	66.0ª	66.9°	69.5 °		
Starter Protein									
21%	34.7 b	56.2ª	61.3ª	63.9ª	65.6°	66.8ª	67.4°		
18%	39.3°	56.5ª	59.3ª	63.8ª	64.9ª	66.5ª	68.8ª		

Table 14. Influence of calories fed to twenty weeks of age and restricted feeding after twenty weeks on egg weight.

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<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

	BW to 20wks	BW after 20 wks	Lean Tissue	Fat tissue	Bone Mass	Egg Prod	Egg Wt.	Double Yolk
	(g)	(g)	(g)	(g)	(g)	(%)	(g)	(%)
Treatments								
Stocking Density								
1.40 ft <sup>2</sup> / bird	1314.8ª	3201 ª	2034.6 <sup>b</sup>	311.6°	51.7 <sup>b</sup>	49.8 <sup>b</sup>	61.4ª	.28 <sup>b</sup>
1.36 ft <sup>2</sup> / bird	1289.6 <sup>b</sup>	3210 °	2044.8 ª	310.0ª	50.6 ª	53.0ª	61.0ª	.33 ª
Feeding Curve								
Linear	1281.9 <sup>b</sup>	3211.0 *	2032.5 <sup>b</sup>	319.0 "	50.9 <sup>b</sup>	51.7ª	61.0ª	.32 ª
Sigmoid	1322.4 ª	3200.4 ª	2046.9ª	302.6 <sup>b</sup>	51.4 ª	51.1ª	61.3ª	.30 <sup>a</sup>
Starter Protein								
21%	1320.9*	3195.4 <sup>b</sup>	2051.9*	298.9 <sup>b</sup>	51.3 <sup>b</sup>	51.7°	61.2 *	.30 °
18 %	1283.4 <sup>b</sup>	3216.0 ª	2027.5 <sup>b</sup>	322.7 *	51.0ª	51.1°	61.2ª	.31ª

Table 15. Influence of starter protein, Stocking density, and feeding regime on mean body weight, Lean tissue, fat tissue, bone tissue, egg production, egg weight ,and Double yolks.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

	BW	Lean Tissue	Fat tissue	Bone Mass
	(g)	(g)	(g)	(g)
Treatments				
CAL				
21,996 Kcal	1156.2 °	1064.9°	111.8°	24.5°
23,400 Kcal	1257.7 <sup>b</sup>	1155.2 <sup>b</sup>	137.0 <sup>b</sup>	27.3 <sup>b</sup>
24,800 Kcal	1334.5 °	1214.6 ª	163.8 °	28.4ª
SP				
21%	1261.9 *	1154.3*	143.3 ª	27.1°
18 %	1237.1 <sup>b</sup>	1135.5 <sup>b</sup>	131.8 <sup>b</sup>	26.4 <sup>b</sup>

Table 16. Influence of cumulative calories fed to twenty weeks of age, and starter protein on mean body weight, Lean tissue, fat tissue, and bone tissue to 20 weeks of age.

<sup>ab</sup> Means within columns with no common superscript differ significantly (P < .05).

	BW	Lean Tissue	Fat tissue	Bone Mass	Egg Prod	Egg Wt.	Double Yolk
	(g)	(g)	(g)	(g)	(%)	(g)	(%)
Treatments							
AMT							
High	3161.4 ª	2403.4 *	372.0ª	59.47 ª	52.8ª	60.9ª	.34 ª
Low	3150.8 ª	2409.7 ª	367.8ª	59.34 ª	52.7 ª	61.1ª	.34 <sup>a</sup>
CAL							
21,996 Kcal	3044.0 °	2325.0°	349.9°	57.3°	50.6 °	60.1 °	.27°
23,400 Kcal	3163.9 <sup>b</sup>	2411.5 <sup>b</sup>	393.2ª	59.6 <sup>b</sup>	54.5ª	61.1 <sup>b</sup>	.33 <sup>b</sup>
24,800 Kcal	3660.4 ª	2482.2 ª	366.6 <sup>b</sup>	61.2 <sup>ª</sup>	53.1 <sup>b</sup>	61.8ª	.43 ª
SP							
21%	3158.6 °	2408.2ª	374.1 <sup>b</sup>	59.6ª	52.0°	61.1ª	.32 <sup>b</sup>
18 %	3153.6 ª	2404.9ª	365.6 *	59.2 <sup>b</sup>	53.5 <sup>b</sup>	61.0ª	.37ª

Table 17. Influence of cumulative calories fed to twenty weeks of age, amount of feed after 20 weeks, and starter protein on mean body weight, Lean tissue, fat tissue, bone tissue, egg production, egg weight, and Double yolks from 20 to 48 weeks of age.

<sup>abc</sup> Means within columns with no common superscript differ significantly (P < .05).

Table 18. Mean (carbon dioxide production, oxygen consumption, body weight, and heat production) values for breeder broiler hens.

	CO2 Prod	02 Consumption	BW	Heat Prod.
Week	(L/min)	(L/min)	(g)	(Kj/Kg)
10	0.9078	0.8056	662.07	24.9072
11	1.3416	0.7308	794.26	26.7705
12	1.0119	1.2123	835.02	26.7445
32	5.4203	5.7821	2914.96	43.4729

Age	Body Weight	Age	Body Weight
Weeks of Age	Grams	Weeks	Grams
3-4	330	17-18	1680
4-5	460	18-19	1800
5-6	580	19-20	1930
6-7	690	20-21	2090
7-8	780	21-22	2240
8-9	870	22-23	2400
9-10	950	23-24	2540
10-11	1030	24-25	2770
11-12	1110	25-26	2900
12-13	1200	26-27	3000
13-14	1290	27-28	3040
14-15	1380	28-29	3130
15-16	1470	29-30	3180
16-17	1570	30-31	3220

Table 19.	Norma	I recommendation	for Cobb	500 broile	er breeder	body v	weight	t.
								-

Values based on Cobb 500 Breeder Management Guide

#### CHAPTER IV

### THE EFFECT OF PANTOTHENIC ACID FORTIFICATION LEVELS AND GROWTH PROMOTERS ON COMMERCIAL BROILERS DURING CYCLIC HIGH AMBIENT TEMPERATURE

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#### Abstract

One experiment was conducted utilizing 2,880 Cobb x Cobb broilers to evaluate the efficacy of supplemental dietary pantothenic acid (PA) 2.5, 7.5, 10, and 15 mg / Kg on blood and heart tissues, collected to see if serum components and heart tissue. The coccidiostat Salinomycin

(SAL) in broilers raised on dirty litter under cyclical ambient temperature stress. Four treatment levels of PA (0, 2.5, 10, and 17.5 mg/kg) and two treatment levels of antibiotics, an antibiotic rich treatment (SAL at 55g activity combined with a zinc-bacitracin and virginiamycin fortification) and a control (SAL alone at 40g activity). No significant differences were detected at any treatment level of PA. Birds fed diets with high levels of antibiotics had reduced calculated carcass fat by 1.6 % (P = 0.1). No significant differences were found between the antibiotic rich vs. control treatments.

(keywords: Pantothenic Acid, broiler, heat stress, Virginiamycin, salinomycin, zinc-bacitracin)

#### Introduction

Pantothenic acid (PA), also known as dihydroxy- $\beta$ - $\beta$ -dimethylbutyrl- $\beta$ -alanine, is a precursor of coenzyme A, a key acyl carrier in oxidative phosphorylation. (Mclure 1996; Stryer 1995; Combs 1992) Serum PA is transported primarily in red blood cells as Coenzyme A. Annous et al (1995) showed that pantothenic acid passively entered red blood cells suggesting that even under stressed conditions adequate therapeutic transport of PA to other tissues is possible. The NRC (1994) established the minimum PA requirement for growth at 10 mg per kilogram of diet. Heat

stress may effect broiler metabolic function in a way that elevated the need for PA above NRC guidelines for broilers.

Broilers are susceptible to heat stress (Adams 1962). Classical symptoms of heat stress include reduced feed intake, reduced growth, and poor feed consumption (Squibb 1959). Dale and Fuller (1980), found that birds were more likely to respond to nutrient supplementation during cyclical heat stress vs. constant heat stress. Beagle and Begin (1976) found birds consuming diets containing 7.5 mg/kg PA or lower had a higher heat increment than birds consuming diets at or above 10mg/kg PA. Harms (1992) found no response to pantothenic acid supplementation levels up to 14.4 mg/kg diet in broilers raised in a thermal neutral environment, but additional dietary pantothenic acid may improve performance of broilers under heat stress by reducing the heat increment.

Antibiotics such as BMD, VM and Sal (typically included in diets as a coccidiostat) have been shown to improve growth, and performance under stressful conditions (Miles et al 1984). In general, antibiotics are fed to reduce the incidence of subclinical levels of bacterial infections of the digestive and respiratory tract, (Jurgens, 1993). Salinomycin was shown to improve broiler performance during gram-positive microbial challenge (Woodbine 1984).

Commercial diets typically include both a coccidiostat, and an antibictic.

The present study was conducted to determine if supplemental pantothenic acid treatments improved broiler performance during cyclic ambient temperature stress in rations with and without drug treatments. The experiment also evaluated the effectiveness of drug rich, and drug lean treatments on broiler performance, when raised on use litter.

#### Materials and Methods

Experimental Design: The experiment was a 2 x 4 factorial with 9 treatment replications, in a completely randomized block design. The experiment utilized 2880 Cobb X Cobb chicks. On day 1 birds were randomly assigned to treatments, on used litter in 72 (152.4cm x 365.76cm) floor pens, to reduce variation. Water was provided by standard, commercial nipple drinker system, which transected the pen diagonally front to back. Feed was provided ab libitum via two suspended gravity feeders. Heat was provided for each pen in the experiment by natural gas brooders. The basal ration (table 1) was supplemented with four levels of pantothenic acid and two levels of antibiotic treatment. Each pen contained 40 chicks initially, birds were fed a starter diet to day 17, a grower diet to day 35, followed by a finisher diet to day 49. Temperature control was achieved

via vari-fan<sup>1</sup> thermostats. Ventilation was accomplished using three high-speed exhaust fans on the east-end of the building, negative pressure was relieved by airflow through two variable height curtains on the west-end of the building. Forced air mass flow was from west to east. On days 21-35 cyclical heat-stress conditions were applied, 12 hours during the day at 31.1°C and 21.1°C during the night.

Birds were monitored for mortality, feed consumption, average body weight, and live weight. At day 49 feed consumption, pen weight, and mortality were recorded and 2 birds per pen were transported from the growing house to the poultry-processing floor. Birds were slaughtered and analyzed for hot-weight, chill weight, breast yield, and specific gravity. Birds were transported from the growing house to the poultry-processing floor. Birds were sacrificed and processed as described by Belay (1994), except where special tissue was harvested. Chilled carcasses were weighed to obtain weight in air (WA) and weight in water (WW) to allow for calculation of specific gravity: Specific gravity was calculated according to Teeter and Smith's equation (1985) as specific gravity = (WA) / (WA-WW). Carcass fat was then calculated based on Teeter's predictive equation: % Carcass fat = 336.97008 - (310.19727 x specific gravity) (Wiernusz et al 1998). Whole breast muscle was removed, and breast yield was calculated as a

<sup>&</sup>lt;sup>1</sup> Vari-fan thermostats are a product of Multifan Inc.

percentage of whole carcass (Belay et al. 1991). Feed efficiency was calculated as ratio of feed consumed per kilogram of live weight gain during 49 days. The outer wall of the right ventricle was collected from each bird, prior to evisceration, placed in Teflon containers and analyzed by ion coupled plasma spectrum analyzer for calcium, sodium, potassium, and magnesium concentrations. Serum samples collected on day 40, were analyzed using a cobasmira<sup>2</sup> chemistry analyzer. The following serum component were analyzed using Roche reagent/electrode kits: magnesium using reagent # 44169, glucose using reagent # 47382 and 47383, sodium and potassium using reagents # 46997, 46998 and reference electrolyte # 46999, albumin using reagent # 42332, and protein using reagents # 44903 and 44026.

Due to high mortality rates adjusted feed efficiency was calculated to include the weight of dead birds to show total tissue produced vs total feed consumed. Analysis was preformed using the general linear model techniques of Statistical Analysis Software (SAS®).

#### Results and Discussion

Summarized results are shown in tables 2, 3, 4, and 5. Pantothenic acid treatments did not significantly affect average body weight, feed/gain ratio, mortality adjusted feed to gain ratio, survivability, and feed consumption. The results above agree previous studies (Harms, 1992; and

<sup>&</sup>lt;sup>2</sup> Cobas mira is a product of Roche.

Deyhim and Teeter, 1992) which found no increase in broiler performance due to excess dietary pantothenic acid supplementation. Body weight was higher for the 10 mg PA treatment (2.7910 g P < 0.05), however this was probably caused by increased feed consumption brought on by high mortality. There were no significant differences among the antibiotic treatments. (Table 2)

No significant treatment differences were detected in breast yield, carcass fat, specific gravity, and dressing percentage. Birds fed the higher antibiotic treatment had numerically decreased carcass fat. (Table 3) No treatment differences (P > 0.05) in right ventricle tissue concentrations of magnesium, calcium, potassium, or sodium, (Table 4). Similarly, no treatment differences (P > 0.05) were detected in serum magnesium, glucose, potassium, sodium, albumin or total protein. However, P value tended to range from 0.1 to 0.05.

Serum metabolites were correlated with heart tissue ion concentrations and performance data. Serum potassium was positively correlated with body weight (R=.22 P < 0.08). Heart tissue Na was negatively correlated with Serum protein (R=-.19 P < 0.05) and serum albumin (R=-.17 P < 0.09). The ratio of Na to K was negatively correlated with serum protein (R=-.17 P < 0.08) and serum albumin (R=-16 P < 0.1). The ratio of Na to Mg were negatively correlated with serum

protein (R=-.18 P < 0.06) and serum albumin (R=-.19 P < 0.06). The ratio of Mg to Ca were positively correlated with serum protein (R=.16 P > 0.09) and serum sodium (R=.18 P < 0.06).

Based on these results we can conclude that exceeding NRC dietary PA recommendation does not appear to provide benefit to broilers reared under conditions of cycling ambient temperature stress on used litter. Extra fortification of the SAL, BMD, and VM antibiotics failed to improve performance of broilers compared to the broilers whose diets contained SAL alone.

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Ingredients	Starter	Grower	Finisher
		(%)	
Com	51.65	60.06	59.81
Soy Bean Meal	37.0	31.0	31.0
Meat & Bone Meal	3.0	0.0	0.0
Fat	6.0	4.7	4.7
Salt	0.23	0.252	0.252
Trace Mineral	0.1	0.1	0.1
Methionine	0.15	0.1	0.1
Lysine	0.0	1.517	1.517
Deflorinated Phosphorus	1.4	1.298	1.298
Calcium Carbonate	0.36	1.02	1.02
Choline	0.05	0.05	0.05
Copper	0.03	0.03	0.03
Ethoxy	0.01	0.01	0.01
Vitamin Premix <sup>1</sup>	0.05	0.05	0.05
Selenium	0.06	0.06	0.06
Total	100.00	100.00	100.00
Calculated Analysis			
ME Kcal/Kg	3,159	3,175	3,175
Crude protein (%)	23	20	20
Calcium (%)	1.00	0.90	0.90
Phosphorus (% av)	0.68	0.47	0.47

#### Table 1. Composition of basal rations.

<sup>1</sup> Premix contained per kilogram: 22,050,000 I.U vitamin A, 7,452,900 I.U vitamin D3, 50,936 I.U vitamin E, 40 mg vitamin B12, 18,081 mg Riboflavin, 121,496 mg Niacin, 6,042 mg Menadione, 2,597 mg Folic Acid, 8,379 mg Pyridoxine, 5,557 mg Thiamine, 267 mg Biotin 1%.

	Ave Bv (k	Average Bwt. (kg)		Feed Efficiency (F/G)		Mortality Adjusted F/G ( F/G)		Survivability (%)		Feed Consumption (Kg)	
Pantothenic Acid (mg)	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	
0	2.7304	$\pm 0.02^{\text{b}}$	1.8840	$\pm$ 0.02 <sup>a</sup>	1.7823	0.01 <sup>a</sup>	86.9575	± 1.50 <sup>a</sup>	5.1412	$\pm 0.08^{b}$	
2.5	2.7910	$\pm 0.02^{a}$	1.9172	± 0.02 <sup>a</sup>	1.7977	0.01 <sup>a</sup>	84.1076	$\pm$ 1.50 <sup>a</sup>	5.4854	$\pm 0.08^{a}$	
10	2.7365	± 0.02 <sup>b</sup>	1.8726	$\pm$ 0.02 <sup>a</sup>	1.7674	0.01 ª	87.5159	$\pm$ 1.50 $^{a}$	5.0950	$\pm 0.08^{\rm b}$	
17.5	2.7133	± 0.02 <sup>b</sup>	1.8957	± 0.02 <sup>a</sup>	1.7849	0.01 <sup>a</sup>	85.1518	± 1.50 <sup>a</sup>	5.2220	$\pm 0.08^{b}$	
SAL	2.7327	± 0.01 <sup>a</sup>	1.9057	± 0.01 <sup>a</sup>	1.7766	0.008 <sup>a</sup>	86.0817	± 1.06 <sup>a</sup>	5.2521	± 0.04 <sup>a</sup>	
SAL w/ BMD & VM	2.7530	±0.01 <sup>a</sup>	1.8791	± 0.01 <sup>a</sup>	1.7895	0.008 <sup>a</sup>	85.7847	±1.06 °	5.2197	±0.04 <sup>a</sup>	

Table 2. Influence of pantothenic acid treatments on average bodyweight, feed to gain, adjusted feed to gain, survivability and feed consumption.

SE standard error of the mean

	Breas (9	t Yield %)	Carca (?)	Carcass fat (%)		Specific Gravity (g/cm <sup>3</sup> )		Percent
Pantothenic Acid (mg)	Mean	SE	Mean	SE	Mean	SE	Mean	SE
0	20.8750	$\pm 0.37$ <sup>a</sup>	12.2742	± 0.35 <sup>a</sup>	1.0468	± 0.001 <sup>a</sup>	75.8868	±0.45 <sup>a</sup>
2.5	21.4088	$\pm 0.39$ <sup>a</sup>	12.1727	$\pm 0.37^{a}$	1.0471	$\pm$ 0.001 <sup>a</sup>	75.7227	$\pm$ 0.48 <sup>a</sup>
10	21.0665	$\pm 0.36$ <sup>a</sup>	11.6816	$\pm 0.34$ <sup>a</sup>	1.0487	$\pm 0.001$ <sup>a</sup>	75.8607	$\pm$ 0.44 $^{a}$
17.5	20.3561	± 0.33 <sup>a</sup>	12.3954	$\pm$ 0.31 <sup>a</sup>	1.0463	$\pm$ 0.001 <sup>a</sup>	74.5749	$\pm$ 0.40 <sup>a</sup>
SAL	21.0357	± 0.26 ª	12.4347	±.25 <sup>C</sup>	1.0462	0.0008 <sup>a</sup>	75.7724	±0.32ª
SAL w/ BMD & VM	20.8175	±0.26 ª	11.8272	±.24 <sup>D</sup>	1.0482	0.0008 <sup>a</sup>	75.2502	± 0.31 <sup>a</sup>

Table 3. Influence of pantothenic acid treatments on breast yield, carcass fat, specific gravity, and dressing percent.

<sup>CD</sup> Means within columns with no common superscript differ significantly (P < .1). SE standard error of the mean

	Magnesium (ppm)		Calcium (ppm)		Potassium (ppm)		Sodium (ppm)	
Pantothenic Acid (mg)	Mean	SE	Mean	SE	Mean	SE	Mean	SE
0	1007.1	± 16.7 <sup>a</sup>	297.0	± 9.1 ª	14070.0	$\pm 241.0^{a}$	8241.8	± 185.9 a
2.5	983.7	±17.1 <sup>a</sup>	302.2	± 9.5 ª	13944.6	$\pm 247.0$ <sup>a</sup>	8156.7	± 196.4 <sup>a</sup>
10	998.5	± 16.6 <sup>a</sup>	279.6	± 9.1 <sup>a</sup>	14168.3	± 240.2 <sup>a</sup>	7850.5	± 185.2 <sup>a</sup>
17.5	987.1	± 15.8 <sup>a</sup>	290.8	± 8.6 ª	13607.9	± 227.6 ª	8218.1	± 179.1 <sup>a</sup>
SAL	986.0	±11.8 <sup>a</sup>	290.2	±6.5ª	13650.4	± 170.8 <sup>a</sup>	8081.3	± 132.7 ª
SAL w/ BMD & VM	1002.2	±11.9 ª	294.6	$\pm 6.5^{a}$	14245.1	$\pm$ 171.2 <sup>a</sup>	8152.2	$\pm$ 134.3 <sup>a</sup>

# Table 4. Influence of pantothenic acid on right ventricle tissue magnesium, calcium, potassium, and sodium concentrations.

<sup>ao</sup> Means within columns with no common superscript differ significantly (P < .1). SE mean standard error (+/-).

PA (mg)	New castle (Titer)		Magnesium (mcg/L)		Glucose (mg/dL)		Potasium (mmol/L)		Sodium (mmol/L)		Albumin (g/dL)		Protein (g/dL)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
0	0.9129	± 0.08°	1.8166	± 0.05 ª	232.83	± 5.33ª	4.6345	± 0.18ª	154.62	± 2.46 ª	1.1779	± 0.04 <sup>a</sup>	3.2346	± 0.11 <sup>a</sup>
2.5	0.9946	± 0.09 <sup>a</sup>	1.8019	± 0.06 ª	242.60	$\pm$ 5.97 <sup>a</sup>	4.9057	± 0.20 <sup>a</sup>	156.06	± 2.76ª	1.2728	± 0.05 <sup>a</sup>	3.3591	± 0.12°
10	0.8900	± 0.09°	1,7375	± 0.06°	240.81	± 5.86 <sup>a</sup>	4.6898	± 0.19ª	156.38	± 2.70°	1.2736	$\pm$ 0.05 <sup>a</sup>	3.4089	± 0.12 <sup>a</sup>
17.5	0.9518	± 0.07 ª	1.7856	± 0.05 <sup>a</sup>	234.98	± 5.21°	4.7289	± 0.17 ª	154.32	± 2.41°	1.2014	$\pm$ 0.04 <sup>a</sup>	3.3189	± 0.11 <sup>a</sup>
SAL	0.9217	± 0.06 ª	1.8040	± 0.04 °	242.9	± 4.1ª	4.877	± 0.12ª	155.1	± 1.9ª	1.247	± 0.03 ª	3.341	± 0.08 ª
BMD & VM	0.9067	± 0.06 ª	1.7147	± 0.04 ª	233.8	± 4.3 <sup>ª</sup>	4.445	± 0.13ª	153.0	± 2.0ª	1.198	$\pm$ 0.04 <sup>a</sup>	3.250	± 0.09 ª

Table 5. Influence of pantothenic acid on newcastle immunity titer and serum magnesium, glucose, potassium, sodium, albumin, and protein levels.

Means within columns with no common superscript differ significantly (P < .05).

SE standard error of the mean.

#### CHAPTER V

### INFLUENCE SUPPLEMENTAL DIETRARY MAGNESIUM PROTEINATE ON BROILERS DURING CYCLIC HIGH AMBIENT TEMPERATURE

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#### Abstract

One experiment was conducted to evaluate the effects of dietary magnesium proteinate (MgP) supplementation in birds reared under simulated summer conditions. Chicks were raised to 49 days of age and evaluated to see if supplemental dietary MgP impacts performance, survivability. Results show significant improvement in average live weight. Dietary Mg concentrations were positively correlated to serum and Cardiac tissue Mg concentrations (R=.3445 P < 0.01). Results indicate that broilers fed diets supplemented with 100 ppm Mg proteinate had higher body weight at 49 days.
(Key words: Magnesium, proteinate, performance, broiler, serum magnesium)

## Introduction

Poultry industry is in continual search for means of improving broiler performance. Though minerals are not normally deficient in commercial broiler diets, mineral bioavailability can vary depending on the ion form. Today advanced processing techniques have provided improvements in mineral processing have produce Magnesium proteinate<sup>1</sup> (MgP) is a result of is potentially more available to the body than metallic ion supplements. The current dietary magnesium requirement for broilers is 600 ppm., (NRC 1994), This requirement was established using metallic ion forms of magnesium such as: MgO (Atteh and Leeson, 1983), MgCO3 (Chicco et al, 1967, Nugara and Edwards, 1963, Stillmark and Sunde 1971), and MgSO4 (McWard 1967). Studies with dolomitic limestone, which contains at least 10% Mg, showed Mg caused reduced growth and decreased bone ash when present at .65% to .9% of the diet, (Griffith et al., 1963; Chicco et al., 1967)

Studies have demonstrated that organic MgP may prove to be less toxic, more bio-available, and have greater tissue specificity than ionic counter parts (Lyons 1993), which may be beneficial in stressful situations. Paripatananont

<sup>&</sup>lt;sup>1</sup> Magnesium Proteinate is a products of Chelated Minerals Inc.

(1997) found that tissue retention of Cu, Fe, Mn, Se, and Zn was increased 72.5, 39.1, 59.6, 56.6, and 23% respectively when birds were fed diets with chelated mineral supplements. Magnesium aspartate has been shown to improve meat quality and lower susceptibility to pale soft exudative syndrone(PSE) in swine under stressful conditions, (D'Souza et al 1998). This might be due to Mg's ability to decrease the liberation of acetylcholine from the neuromuscular junctions, and sympathetic ganglia. Mg is required by more than 300 enzymes, including phosphate transferring, glycolysis, and nucleic acid synthesis enzymes, (Britton et al. 1989; Aikawa, 1971; Wacker 1980). Elevated serum Mg, hypermagnesemia is known to cause hypotension, increased sensitivity of the carotid sinus, cutaneous vasodilatation, depression of the peripheral neuromuscular junctions, muscular weakness, decreased tendon reflexes, dysarithria, ataxia listlessness, lethargy, drowsiness and coma, depending on the concentration of Mg in the serum. (Aikawa 1971; Wacker 1980).

In a recent pilot study M. Daskiran (unpublished data) conducted a trial, at Oklahoma State University, using treatments of 0 and 200 ppm of added MgP, (table 2). Daskiran found no treatment effects (P > 0.05), however the numerical depression in body weight indicate a possible toxicity of magnesium proteinate at 200 ppm.

The following study was conducted to evaluate the effects of supplemental MgP treatments in exposed to cyclic high ambient temperature.

### Materials and Methods

Experimental Design: Two treatments were replicated in 8 blocks of a completely randomized block design. Analysis was preformed using the general linear model techniques of Statistical Analysis Software (SAS®). Nineteen hundred and twenty Cobb X Cobb birds were raised on litter in 48 (152.4cm x 365.76cm) floor pens contained in a 72 floor pen house, designated 72-House, located on the OSU Avian Climatology research farm. On day 1 birds were randomly assigned to treatments in order to reduce individual variation. Water was provided by standard, commercial nipple drinker system, which transected the pen diagonally front to back. Feed was provided in two suspended gravity feeders. Building was heated utilizing gas brooders. Each pen contained one brooder. The basal ration (table 1) was supplemented with 2 levels of magnesium chelate at 0 & 100 ppm. Each pen contained 40 chicks initially, birds were fed a starter diet to day 17 then a grower diet to day 35 followed by a finisher diet to day 49. Thermo-regulation distribution was achieved via

vari-fan<sup>1</sup> thermostats. On day 21 cyclical heat-stress conditions were applied, during the first 12 hours of the day it was maintained at 31.1°C and the remaining 12 hours it was maintained at 21.1°C. Heat-stress was terminated on day 35. Samples for serum chemistry analysis were taken on day 40. Two birds per pen were bled from the wing vein, (approximately 2 ml), into vacuum serum separating tubes. Blood was allowed to clot, tubes were centrifuged at 3000 rpm's for 25 minutes and serum collected in individually labeled collection vials for analysis. Serum samples were analyzed using a Roche Cobas Mira chemical analyzer for Na, K, Albumin, Protein, and Glucose. At day 49 feed consumption, pen weight, and mortality were recorded and 2 birds per pen were transported from the growing house to the poultry-processing floor. Birds were weighed then slaughtered and analyzed for hot-weight, chilled weight, specific gravity, breast yield. Birds were sacrificed and processed as described by Belay (1994), except where special tissue was harvested. Chilled carcasses were then weighed to obtain weight in air (WA) and weight in water (WW) to allow for calculation of specific gravity: Specific gravity was calculated, according to Teeter and Smith's equation (1985), as Specific gravity = (WA)/(WA-WW).

<sup>&</sup>lt;sup>1</sup> Varifan is a product of Multifan Inc

Carcass fat was then calculated based on Teeter's predictive equation: % Carcass fat = 336.97008 - (310.19727 \* specific gravity)(Wiernusz et al 1998). Whole breast muscle was removed, and breast yield was calculated as a percentage of whole carcass (Belay 1994). The outer wall of the right ventricle was collected from each bird, prior to evisceration, placed in Teflon containers and analyzed by ion coupled plasma spectrum analyzer for calcium, sodium and magnesium concentrations. Serum samples collected on day 40, were analyzed using a cobasmira<sup>1</sup> chemistry analyzer. The following serum component and their Roche reagent/electrode kits were analyzed magnesium using reagent # 44169, glucose using reagent # 47382 and 47383, sodium and potassium using reagents # 46997, 46998 and reference electrolyte # 46999, albumin using reagent # 42332, and protein using reagents # 44903 and 44026.

# Results and Discussion

Birds fed diets supplemented with of 100 ppm MgP showed tended to improved live body weight by 47.6 grams (P < 0.08). This is consistent with findings of Donoghue (1990) where Magnesium-Aspartate-Hydrochloride reduced weight loss in heat-stressed laying hens.

<sup>&</sup>lt;sup>1</sup> Cobas mira is a product of Roche

Serum magnesium levels were not significantly affected by supplemental MgP. Donoghue et al. (1990) and Kohnne and Jones (1975) saw a drop in serum Mg levels during heat stress. Heart tissue Mg concentration was significantly lowered by 31.4017 ppm (P<.08) in the supplement birds. Work done by Rodriguez-Zavala et al (1998) indicate that altering the concentration of Mg in rat mitochondria alters the rate of oxidative phosphorylation as well as the activity of certain enzymes. Heart tissue calcium was not significantly affected by the Mg treatment, even though it has been shown that excess dietary Mg can interfere with calcium absorption, (Scott, 1982).

Serum magnesium was positively correlated with heart tissue potassium (R= .3025 P<.03) and magnesium (R= .3445 P<.01). Heart tissue Na<sup>+</sup> to Mg<sup>++</sup> ratio was negatively correlated to serum protein (R= -.2364 P<.09) and serum albumin (R= -.2469 P<.07) concentrations. Heart tissue Na to K ratio was negatively correlated with serum albumin (R= -.2324 P< .09).

Results of this trial indicate that addition of 100 ppm MgP to basal poultry rations tends to improve body weight in heat stressed broilers to 49 days of age, possibly by inducing cardio hypotension or cutaneous vasodilation which might aid in heat dissipation. Current

research indicates the therapeutic level of supplemental magnesium proteinate, for broiler under heat distress, may lie between 0 and 200 ppm MgP in addition to 600ppm Mg requirement, (NRC 1994). Additional studies are needed to determine the optimal level of magnesium proteinate supplementation for broilers.

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- Wiernusz, C. J., B. C. Park, R. G. Teeter, 1998. Prediction of carcass fat, protein and energy from carcass dry matter and specific gravity of broilers. Australian J. of Animal Sci. (In Press)

				F	Performance						
	Feed Efficiency (f/g)		iency	Survivability		Feed (	Feed Consumption (Kg)			Body Weight (Kg)	
				(0							
Mg proteinate	Me	an	SE	Mean	SE	Mean	S	E	Mean	SE	
(ppm)											
200	2.13	302	0.01706	92.9274	0.8871	5.5714	0.0	443	2.6172	0.0135	
0	2.13	372	0.01716	93.2051	0.8871	5.5743	0.0	443	2.6092	0.0135	
P>T		0.770	3	0.8	257		0.9642		0.677	5	
				Carcas	ss Character	istics					
	Breast Yield		Abdon	Abdominal Fat (%)		Carcass Fa t (%)		Specific Gravity (%)		Dressing Percentage (%)	
	(?	(%)									
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	
200	19.9558	0.1359	27.3693	1.0569	12.1935	0.1551	1.0470	0.0005	76.0824	0.2063	
0	19.6864	0.1469	27.9303	1.1150	12.3176	0.1861	1.0466	0.0006	76.2871	0.2179	
P>T	0.1	0.1776 0		7149	.8526		0.5987		0.4951		
			Serum Cl	nemistry							
10	Serum M	aonesium	Serum T	rialvcerides	-						
	(mcg/dL)		(m	(mg/dL)							
	Mean	SE	Mean	SE							
200	1.5413	0.0233	39.4601	1.0115 <sup>b</sup>	-						
0	1.5334	0.2374	43.4386	1.0306*							
P>T	.8127		.0	064							

Table 1. Influence of dietary magnesium proteinate on broiler performance, carcass characteristics and serum chemistry analysis.

<sup>ab</sup> Means within rows with different superscripts are significantly different (P>.05) Printed with the approval of (Daskiran et al. 1994).

Ingredients	Starter	Grower	Finisher	
	·	—— (%) —		
Corn	51.62	61.39	61.39	
Soy Bean Meal	37.0	31.0	31.0	
Meat & Bone Meal	3.0	0.0	0.0	
Fat	6.0	4.7	4.7	
Salt	0.23	0.252	0.252	
Trace Mineral	D.1	0.1	0.1	
Methionine	0.15	0.1	0.1	
Lysine	0.0	1.517	1.517	
Deflorinated Phosphorus	1.4	1.298	1.298	
Calcium Carbonate	0.36	1.02	1.02	
Choline	0.05	0.05	0.05	
Copper	0.03	0.03	0.03	
Ethoxy	0.01	0.01	0.01	
Vitamin Premix <sup>1</sup>	0.05	0.05	0.05	
Selenium	0.06	0.06	0.06	
Total	100.00	100.00	100.00	
Calculated Analysis				
ME Kcal/Kg	3,159	3,175	3,175	
Crude protein (%)	23	20	20	
Calcium (%)	1.00	0.90	0.90	
Phosphorus (% av)	0.68	0.47	0.47	

Table 2. Composition of basal diet.

<sup>1</sup> Premix contained per kilogram: 22,050,000 I.U vitamin A, 7,452,900 I.U vitamin D3, 50,936 I.U vitamin E, 40 mg vitamin B12, 18,081 mg Riboflavin, 121,496 mg Niacin, 10 mg/Kg d-pantothenic acid, 6,042 mg Menadione, 2,597 mg Folic Acid, 8,379 mg Pyridoxine, 5,557 mg Thiamine, 267 mg Biotin 1%.

	Body Weight (Kg)		Survivability (%)		Feed Et (f/	fficiency (g)	Feed Consumption (Kg)			
Mg P (ppm)	Mean	SE	Mean	SE	Mean	SE	Mean	SE		
0	2.6945	.0189	85.2461	1.3068	1.8820	0.0163	5.1137	0.0412		
100	2.7421	.0189	87.5017	1.3068	1.8732	0.0163	5.1817	0.0412		
P>T	0.0844		0.2287		0.7056		0.2539			
	Carcass Characteristics									
	Breast Yield (%)		Carcass Fat (%)		Specific Gravity (%)		Dressing Percentage (%)			
	Mean	SE	Mean	SE	Mean	SE	Mean	SE		
0	20.9660	0.4340	12.1495	0.2895	1.0476	0.0013	77.7854	0.3560		
100	20.5925	0.4558	11.9356	0.2792	1.0493	0.0014	77.1366	0.3738		
P>T	0.5616		0.5969		0.3923		0.2226			

Table 3 Influence of 100 ppm Magnesium proteinate supplementation on broiler performance, carcass characteristics, serum chemistry, and heart tissue mineral concentrations.

<sup>ab</sup> Means within rows with different superscripts are significantly different (P>.05) SE stadard error of the mean

MgP (ppm)	Serum Magnesium (mcg/L)		Serum Glucose (mg/dL)		Serum Protein (g/dL)		Serum Sodium (mmol/L)		Serum Albumin (g/dL)	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
100	1.7514	.04185	237.4527	4.3964	3.2718	0.0913	153.3260	2.0894	1.2074	0.0365
0	1.7408	.04394	233.5404	4.5850	3.2082	0.0952	152.9215	2.1790	1.1871	0.0381
P>T	0.8616		0.542		0.6328		0.8943		0.7023	

Table 4 Influence of dietary Magnesium proteinate supplementation on serum chemistry values.

<sup>ab</sup> Means within rows with different superscripts are significantly different (P>.05) SE stadard error of the mean

	concentrations.							
	Magne (ppn	Calcium (ppm)		Potassium (ppm)		Sodium (ppm)		
MgP (ppm)	Mean	SE	Mean	SE	Mean	SE	Mean	SE
100	972.8869 <sup>D</sup>	12.5012	295.1217	6.8506	13673	192.48	8134.68	135.06
0	1004.2886 <sup>C</sup>	12.7255	291.7345	6.9736	14022.71	196.49	8190.73	136.31
P>T	0.0833		0.7310		0.2086		0.7719	

Table 5. Influence of dietary magnesium proteinate supplementation on right ventricle tissue mineral

<sup>ab</sup> Means within rows with different superscripts are significantly different (P>.05) <sup>CD</sup> Means within rows with different superscripts are significantly different (P>.1)

# VITA

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Masters of Science

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