THE EFFECT OF NUTRIENT DENSITY ON PERFORMANCE AND PROCESSING

YIELD IN COBB 700 × MV MIXED-SEX BROILER CHICKENS

A Dissertation

by

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ABSTRACT

The objective of this research program was to determine the dietary amino acid and energy density required to optimize performance and yield in the Cobb $700 \times MV$ broiler in a series of 4 experiments. Experiment one consisted of 2×3 factorial design with a high or low level of apparent metabolizable energy (AME) and a low, medium, or high level of digestible essential amino acid density (dEAA). Broiler performance was incrementally improved with increasing dEAA and AME. Increasing dEAA increased breast fillet and tenderloin yield and reduced fat pad yield. Feeding the high AME diet reduced feed intake and increased fat pad yield leading to a reduction in breast meat yield. No treatment interactions were observed, suggesting that the low level of AME was adequate to support increased muscle accretion.

Experiment two consisted of five dietary treatments: a control (CON) and a CON plus 5%, 10%, 15%, or 20% of dEAA. Broiler performance was improved with each stepwise increase in dEAA. Feeding the 20% treatment led to the greatest increase in breast fillet yield and decrease in fat pad yield. Regression analysis confirmed a positive linear relationship with digestible lysine intake and total breast meat yield. A breakpoint in breast meat yield response was not observed, suggesting that yield may be improved further with a dEAA above +20%.

Experiment three and four consisted of two identically designed experiments, each utilizing the same seven dietary treatments: a low (L), medium (M), or high (H) dEAA during the starter phase followed by a L, M, H, or High + (H+) dEAA during the grower, finisher, and withdrawal phases. Increasing dEAA improved performance and led to incremental increases in breast fillet and tenderloin yield and decreases in fat pad yield. Additionally, increasing dEAA from 0-12 d increased breast meat yield at harvest irrespective of the density fed from 12-49 d. In totality, these data demonstrate the beneficial impact of increasing dEAA on Cobb 700 x MV broiler performance and yield and define expected improvements associated with these increases, allowing producers to conduct economic evaluations to determine their most profitable nutrient density.

DEDICATION

To my wife, Katherine. Your unconditional love is a pillar on which I can lean. An immeasurable force, giving me the strength to achieve anything.

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Thank you Dr. Jason Lee for giving me the opportunity to join your lab and obtain my doctoral degree. Your dedication is unmatched by any other scientist that I have encountered in either industry or academia. Working with you enabled me to conduct numerous studies, engage with my peers at many scientific meetings, and build my professional reputation. I am looking forward to continuing to collaborate with you as I enter the industry.

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NOMENCLATURE

EAA	Essential Amino Acid
СР	Crude Protein
dEAA	Digestible Essential Amino Acid
NEAA	Non-Essential Amino Acid
dLys	Digestible Lysine
dArg	Digestible Arginine
Gly _{equiv}	Glycine Equivalent
BW	Body Weight
FCR	Mortality Corrected Feed Conversion Ratio
FI	Feed Intake
na/na	Homozygote Naked Neck Chicken
Na/na	Heterozygote Naked Neck Chicken
IGM	Ivey Growth Model
BPHL	Bromley Park Hatcheries Limited
IGF-1	Insulin-Like Growth Factor 1
CP:AME	Crude Protein to Apparent Metabolizable Energy Ratio

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CHAPTER I

INTRODUCTION

Amino acid nutrition in animal agriculture is a vital factor in the efficient production of meat, eggs, and milk. The essentiality of dietary amino acids for growth and maintenance was first demonstrated by Osborne and Mendel (1916) when they fed rats diets deficient in lysine and tryptophan. Thereafter, requirements for arginine (Klose et al., 1938), methionine (Briggs Jr et al., 1942), tryptophan, threonine (Almquist, 1947), lysine (Grau, 1948), isoleucine, leucine and valine (Grau and Peterson, 1946) were determined for maximal growth rate in chicks. The concept of a diet containing a perfect balance of amino acids was first theorized by Mitchell and Block (1946). This concept would ultimately be put into practice as an ideal amino acid profile, where a diet provides a precise blend of essential amino acids (EAA) (Baker and Han, 1994). The widespread use of computer based least cost formulation by broiler producers and the implementation of digestible amino acid values greatly improved precision feeding (Rostagno et al., 1995). Additionally, the commercial availability of several crystalline amino acids has led to the reduction in dietary crude protein (CP) and nitrogenous waste. Collectively, these historic advances in amino acid nutrition have aided producers in their ability to precisely and economically feed broilers.

The selection of broilers for improved growth rate, feed efficiency, and meat yield perpetuates the need for the re-evaluation of amino acid requirements. Optimal amino acid density can vary depending on sex, age, strain, environmental conditions, and market objectives. Additionally, the amino acid density of a diet contributes largely to the overall cost of production. Therefore, the economic viability of poultry production is dependent upon feeding ingredients with known quantities of digestible essential amino acids (dEAA) and understanding the dEAA responses of a broiler strain. The literature contains an immense amount of information on factors that affect the amino acid responses of broiler chickens. In the absence of available research for a particular strain, a nutritionist must rely on data derived from other strains, which can lead to overor under-feeding amino acids and reduce profitability. At a minimum, research should be conducted to assess the response of feeding increased dEAA density on performance and meat yield.

The ultimate goal in amino acid research is to develop factorial models that can predict strain responses in varying environmental, dietary, and economic scenarios. Determining the ideal dEAA ratio, digestibility of feedstuffs, maintenance requirements, accretion rate, and the efficiency of utilization above maintenance are all important elements in modeling amino acid nutrition (Fatufe et al., 2004). The application of modeling can allow nutritionists to implement feeding strategies based on the cost of ingredients, the response of a particular broiler strain to amino acid density, and market conditions. The purpose of this review is to determine how these factors can be incorporated into strategies to aid nutritionists in profitable poultry production.

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CHAPTER II

LITERATURE REVIEW

Amino Acid metabolism, utilization, and partitioning

An adequate supply of dietary amino acids is necessary to sustain the normal physiological mechanisms that occur in a growing animal. As such, an amino acid requirement is made up of two components, the requirement for maintenance and the requirement for growth. The two components are a function of the catabolic and anabolic processes that occur within the tissues, making the pattern of amino acids required for each component quite different (Fuller et al., 1989). In growing chickens, the anabolic processes that accrete lean tissue are dependent upon a constant and balanced supply of dEAA. A deficiency in any essential amino acid affects protein turnover by reducing the synthesis and increasing degradation. Defining the dEAA requirements for maintenance and growth and determining the effect of amino acid intake on metabolism, utilization, and partitioning are important elements to develop accurate requirement models.

Maintenance Requirements. Determining the maintenance of amino acid requirements for a particular genetic strain of broiler is necessary as the selection for greater amounts of lean mass changes metabolic and physiological processes. For example, obligatory amino acid losses, which occur through increased oxidation or losses in the digestive tract, account for a portion of the maintenance requirement and differ between genetic strains (Fuller, 1994). Collectively, increased obligatory amino acid loss reduces the efficiency of amino acid utilization. Maintenance amino acid requirements are often assessed by defining the nitrogen balance, which is the point where nitrogen intake exactly equals the sum of nitrogen losses so that the nitrogen content of the body remains constant (Sakomura et al., 2015). Therefore, nitrogen balance implies the state in which there is no growth, protein accretion, or amino acid accretion. There is evidence that whole-body amino acid accretion is a preferred method in defining dEAA maintenance requirements as opposed to measuring nitrogen balance. Animals can adapt to low amino acid intake and maintain nitrogen balance while depleting lean body mass (Laflamme and Hannah, 2013). Additionally, nitrogen balance does not account for changes in body composition or health in response to low amino acid intake, prior nutritional status, or current energy status. Collectively, these effects lead to an underestimation of the dEAA maintenance requirements when assessed via nitrogen balance.

Measuring total body protein to determine the point of zero protein accretion may be a better method in defining dEAA maintenance requirements. Although, some have argued that this method also underestimates the maintenance requirement because total body protein may be accreting while the amino acid in question is still limiting (HM Edwards 3rd and DH Baker, 1999). The counter to this assertion is that if amino acid intake is increasing and there is positive protein accretion, then, by definition, this level cannot be maintenance even if the accretion of the amino acid in question is zero (Sakomura et al., 2015). However, these different methods produce maintenance values that are within 10% of one another. There is not a direct relationship between amino acid requirements and body weight (BW) because the maintenance of lipids, water, and minerals do not have an amino acid cost. Nonetheless, several reports have suggested that maintenance amino acid requirements expressed as a function of metabolic BW and daily intake allow for more effective comparison between different studies and genetic strains (Gous, 2007; Bonato et al., 2011; Sakomura et al., 2015).

Amino Acid Utilization. The linear relationship between amino acid intake and protein accretion suggests that the efficiency of utilization above maintenance for a limiting amino acid is constant over a wide range of intakes, but varies between individual amino acids. For example, Baker et al. (1996) fed 10-20 d old chicks diets with seven graded levels of valine ranging from 5 to 95% of the ideal level for maximal weight gain and feed efficiency. The efficiency of utilization for valine was 73%, and utilization did not decline with increasing intake. Edwards III et al. (1997), HM Edwards III and DH Baker (1999) and HM Edwards III and DH Baker (1999) conducted identical experiments for threonine, lysine, and methionine plus cystine or total sulfur amino acids (TSAA). The efficiency of utilization above maintenance was 82, 79.3, and 68% for threonine, lysine, and TSAA, respectively. In all four experiments, linear regression analysis showed that the level of intake for each amino acid did not affect the efficiency of utilization above maintenance. A unique aspect of these studies is that utilization was accessed for amino acid accretion as opposed to protein accretion. Interestingly, several studies have shown that there is a decline in the efficiency of protein utilization with increasing protein intake (Jackson et al., 1982; Deschepper and DeGroote, 1995; Swennen et al., 2004). Studies with pigs and rainbow trout show that lysine utilization over a wide range of intakes is not linear and follows the law of diminishing returns (Gahl et al., 1994; Rodehutscord et al., 1997). Therefore, the reduction in protein utilization observed by feeding increasing levels of CP may indicate that the rate of oxidation increases for a particular dEAA as the dietary supply approaches or surpasses the requirement. Furthermore, these findings suggest that measuring amino acid accretion versus protein accretion will provide varying results on the efficiency of utilization for an individual amino acid.

TSAA Use and Accretion. The effects of dietary methionine and cystine on protein turnover and body composition have been studied extensively in the chicken. Methionine is the first limiting amino acid in corn-soybean meal broiler diets and is involved in a variety of metabolic functions. The requirement for methionine is often combined with cystine and expressed as TSAA. Neither cystine nor methionine can be synthesized endogenously, but methionine can be readily converted to cystine through reverse transsulfuration in the liver. This conversion allows for cystine to supply up to 50% of the TSAA requirement (wt:wt) (Baker, 2006). Feathers and their associated structures make up approximately 3-6% of BW and are predominately composed of protein with a high concentration of cystine (Leeson and Walsh, 2004). From 0-6 weeks, approximately 10% of the daily dietary protein intake is partitioned to feather formation (van Emous and van Krimpen, 2019). Thus, feather growth may represent a significant proportion of the total dietary TSAA requirement. Studies that compare full sibling homozygote chickens with normal plumage (na/na) to heterozygote naked neck chickens (Na/na) have evaluated the effects of feather growth on TSAA requirements. Interestingly, these studies have found that either the TSAA requirement does not differ between na/na and Na/na chickens or the difference is too small to be practically applied in diet formulation (Pesti et al., 1996; Yalcin et al., 1999).

Furthermore, Pesti et al. (1996) found that weight gain was impacted by changes in dietary content of TSAA more readily than feather growth. The methionine composition of skeletal muscles is among the lowest of all essential amino acids (Hamm, 1981; Murphy, 1994). Nonetheless, a methionine deficiency compromises the efficiency of amino acid utilization and reduces protein accretion. Broilers fed methionine- and cystine- free diets have reduced growth and lower rates of whole-body protein synthesis. Conversely, methionine supplementation improves muscle growth and increases protein synthesis and accretion in the pectoralis major (Barnes et al., 1995).

Lysine Use and Accretion. Lysine has the greatest impact on carcass composition and muscle growth. A lysine deficiency has the potential to reduce breast meat yield due to the high concentration of lysine in the tissue (Hamm, 1981; Vieira and Angel, 2012). The maintenance requirement for lysine is low relative to other EAA, which is likely due to the low rate of degradation and predominate use in muscle deposition as opposed to metabolism. Sakomura et al. (2015) found that the maintenance

lysine requirement was 81 mg/kg^{0.75}/d based on zero protein accretion. HM Edwards 3rd and DH Baker (1999) determined the maintenance requirement for lysine in fastgrowing broilers as 114 mg/kg^{0.75}/d and slow-growing broilers as 89.1 mg/kg^{0.75}/d based on zero lysine accretion. Leeson et al. (2001) suggested that the maintenance requirement is lower when measuring zero protein accretion because the bird may still be losing lysine even while other amino acids accrete. When chicks are fed lysine levels near maintenance, there is a shift in whole-body protein toward collagen production, which is rich in proline and glycine, versus lysine-rich contractile protein. This is evident by the inverse linear relationship in whole-body amino acid composition when birds are provided lysine at maintenance, where proline and glycine concentrations increase as lysine concentration decreases (HM Edwards 3rd and DH Baker, 1999).

The importance of lysine for muscle accretion is evident when the responses to increased dietary lysine are compared between broilers and layers. Fatufe et al. (2004) fed broiler and layer chickens gradient increases in lysine concentrations that ranged from 0.38% to 1.68% from 8 to 21 days. In general, the lysine requirement for optimal conversion or weight gain in the layer chicken was 20% lower than in the broiler chicken. In both genotypes, protein accretion increased with increasing dietary lysine. Interestingly, the study found that broilers had a 10% increase in the efficiency of lysine utilization compared to layers (61% vs. 71%). Dietary amino acid digestibility was found to be the same between the two genotypes, indicating that the improvement in utilization by the broiler was purely a response to differences in post-absorptive metabolism. Selection for high muscle growth in modern broilers has led to changes in

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protein synthesis and degradation rates compared to older genotypes (Tesseraud et al., 2001). Tamminga (1991) concluded that a reduction in protein degradation rate was responsible for the rapid protein accretion of modern strains. It seems logical that reducing protein breakdown would be more energetically favorable than increasing protein synthesis. As broilers are continually selected for increased breast meat yield and improved feed efficiency, there will likely be a corresponding improvement in the efficiency of amino acid utilization.

Ideal Protein and experimental methodologies

Researchers at the University of Illinois were the first to establish an ideal amino acid profile for chicks (Baker and Han, 1994). In the most basic form, the ideal protein concept states that all birds require dietary amino acids to be supplied in a precise balance to achieve optimum performance. Non-limiting amino acids are oxidized when they are provided in excess of the first limiting amino acid. Therefore, feeding a diet with an ideal protein ratio optimizes the amino acid balance because nitrogen utilization is maximized, and amino acid oxidation is reduced. Several scientific approaches, including the factorial, dose-response, and deletion methods, have been used to determine ideal amino acid profiles. These methods have led to the development of two concepts that are foundational in broiler amino acid nutrition. The first concept has shown that broilers fed imbalanced high amino acid density diets have reduced performance compared to those fed balanced low amino acid density diets. Secondly, the requirement for dEAA can vary depending on many factors, but the ratios between dEAA tend to remain relatively similar. The latter observation is the basis for stating the requirement for all dEAA as a ratio to lysine. The application of this ratio limits the need to continuously reevaluate every dEAA requirement, which varies depending on environmental, genetic, and dietary conditions. Instead, the requirement for lysine is determined, and the remaining essential amino acids are supplied following the ratio. Lysine is used as the reference amino acid because it is almost exclusively utilized for body protein accretion, has no metabolic interactions with other amino acids, and is relatively easy to analyze. The following sections describe the three methodologies used when determining the ideal protein ratio in animals.

Factorial Method. The factorial method is derived from the concept that amino acid requirements can be divided into three components: the requirement for body protein accretion, feather growth, and maintenance. To define the digestible amino acid requirements for particular response criteria, such as egg output or breast meat yield, an investigator must determine the net amino acid retention for body and feather protein, maintenance requirement, and utilization of absorbed amino acids. Maintenance is defined as either the number of amino acids needed to achieve zero net amino acid accretion, protein accretion, or nitrogen equilibrium. The requirement for feather growth and lean mass accretion is determined by analyzing the tissues for amino acid content and dividing by a utilization coefficient (e.g., the sum of obligatory amino acid losses). Lemme (2003) argued that a potential shortfall in the factorial method is the assumption that the efficiency of utilization for the amino acid in question is constant across all

levels of intake above maintenance. This argument can also be described as the assumption that a linear dose-response relationship is present regarding the utilization of absorbed amino acids. Some assert that as the intake of an amino acid approaches the requirement, the more oxidation occurs, leading to a reduction in the efficiency of utilization. As mentioned in an earlier section of this review, studies conducted by Edwards III and Baker (1997, 1999a & 1999b) are not in agreement with Lemme (2003) and have shown that the efficiency of utilization of an dEAA is constant over a wide range of intakes above maintenance. There is evidence that the efficiency of protein utilization declines with each incremental increase in protein intake above maintenance, but this does not explain the observations with individual amino acids (Jackson et al., 1982; Swennen et al., 2004). The effect of protein intake on utilization above maintenance may be due to an amino acid imbalance.

It is paramount to consider the metabolism and physiological function of an individual dEAA when analyzing responses in factorial experiments. For example, lysine is used almost exclusively for contractile protein accretion, and the requirement has a positive linear-plateau function as the bird matures. Methionine + cystine are primarily utilized for feather growth and maintenance; thus, the requirement is a function of BW and, therefore, age. For example, the TSAA: LYS ratio increases from 65% at day 7 to 99% at day 63. When requirement values are graphically compared, lysine and TSAA follow a linear plateau function, indicating that both amino acids would be suitable as the reference in the ideal protein concept. The goal of amino acid nutrition in animal agriculture is to determine the optimum balance of amino acids that maximize

productive traits (e.g., breast meat yield or egg output). Therefore, the relationship of lysine with muscle growth versus TSAA with bodyweight, make the former the preferred choice as the reference dEAA in the ideal protein concept.

Dose-Response Method. Dose-response studies are conducted by doing a series of experiments concurrently to evaluate changes in growth and body composition from feeding varying levels of a single amino acid. Care is taken to reduce non-treatment variation between studies by closely maintaining environmental conditions, diet composition, and study design to enable comparison of multiple experiments. Mack et al. (1999) conducted a series of dose-response experiments to determine the optimum TSAA, threonine, tryptophan, arginine, isoleucine, and valine: digestible lysine (dLys) ratio for two commercial strains from 20-40 days of age. Broilers were fed an identical basal diet in all experiments, which consisted of corn and soybean meal with 17.2% CP and 3153 kcal/kg. The basal diet was fed with six graded levels of the dEAA under evaluation and was adequately supplemented with all other dEAA. For example, the experiment to determine the methionine requirement contained 0.9, 1.5, 2.1, 2.7 3.3, and 3.9 g/kg of added DL-methionine. The study measured meat yield and performance and determined the breakpoint in response using both the exponential and broken-line models. The study found that the ideal amino acid ratio relative to lysine was 75% TSAA, 63% threonine, 19% tryptophan, 112% arginine, 71% isoleucine, and 81% valine. For optimal feed conversion, 1.15% dLys was recommended. The primary limitation of using the dose-response method is the selection of a model to determine the appropriate dEAA level. The broken line regression model is dependent upon the flawed assumption that an individual animal or population has a linear response to a limiting nutrient which plateaus at the best inclusion (Pesti et al., 2009). This method tends to underestimate the optimal dEAA value. The primary limitation of the exponential model is that an arbitrary selection of the asymptote is needed to identify the optimal dEAA level. For example, if Mack et al. (1999) selected the optimum dLys value based off 95% or 99% of the asymptotic response, then the dLys value would be either 1.02% or 1.17%, respectively.

Deletion Method. The deletion method was first used in rats and subsequently swine and poultry. The procedure relies on the concept that the reduction of a non-limiting amino acid has no effect on nitrogen retention (Bender, 1965; Fuller et al., 1989; Wang and Fuller, 1989; Webel and Baker, 1999). After removing a proportion of a non-limiting amino acid, the changes in nitrogen retention are measured to calculate a dietary amino acid profile in which all the amino acids are equally limiting. By comparing the response to feeding a positive control diet with no dEAA deficiency versus the test diets with the removal of the dEAA under evaluation, the ideal protein can be determined. The effects of the test diets on N-retention are associated with the slope of the regression line between the test diet and the control group. For all dEAA except lysine, the regression lines are assumed to be the same. Therefore, a point can be determined where a reduction of a dEAA would not affect N-retention. For example, Gruber et al. (2000) fed control diets with 0.95% and 0.81% of lysine and TSAA, respectively. The control

dEAA inclusions were set to 100% and then reduced by 20% to 0.76% lysine or 35% to 0.53% TSAA. The nitrogen retention for the control diet was 56.2% of N-intake. Feeding the lysine and TSAA test diets reduced N-retention by 16.2% and 14.4%, respectively. Amounting to a 0.81% and 0.41% decrease in N-retention for each percentage point reduction in lysine and TSAA, respectively. These results show that TSAA could be reduced by 83% before a decline N-retention, indicating that 71% TSAA: 100% lysine is the appropriate ratio. The primary limitation of the deletion method is the assumption that the slopes of dose-response relationships are identical for all amino acids. This assumption does not account for the differences in amino acid requirements for maintenance, feather growth, and lean growth.

Feeding intact protein versus free amino acids

Substantial research over the past 60 years on dEAA requirements has allowed nutritionists to precisely feed broilers, thus reducing nitrogen excretion and improving economic efficiency. The continued expansion in the availability of feed grade crystalline amino acids has reduced the level of intact protein supplied in broiler diets. The implication of these factors has been a shift from using CP values to dEAA values as constraints in least-cost formulation, culminating in a dramatic reduction in dietary CP (Rostagno et al., 1995). For example, the 1960 Poultry NRC recommended feeding broilers 28% CP from 0 to 8 weeks of age. Conversely, the 2014 nutrition recommendations for the Ross 708 broiler state that from 0 to 8 weeks of age, diets should contain an average of 20.5% CP. This progression has allowed the broiler

industry to precisely feed protein ingredients and limit waste. The economic impact of feeding excess CP is derived from the overuse of expensive protein contributing ingredients. Furthermore, several reports have demonstrated a strong relationship between nitrogen intake and nitrogen excretion in broilers fed varying levels of CP (Ferguson et al., 1998a; Ferguson et al., 1998b; Bregendahl et al., 2002). This relationship, combined with a public concern of the environmental impact of nitrogenous waste, has increased the interest in lowering CP in broiler diets (Siegert and Rodehutscord, 2018).

The historic decline in dietary CP has prompted research into defining the limits on replacing intact protein with free amino acids. Many studies have shown that broilers fed low CP diets do not perform as well as those fed diets with normal CP, even when both diets contain an equal concentration of dEAA. Edmonds et al. (1985) experimented with 0- to 8-day old chicks fed diets with either 24% CP or 16% CP. Both diets were formulated to contain the same level of all suspected limiting amino acids. Chicks that were fed the 16% CP diet had reduced weight gain compared to those fed the 24% CP diet. A similar experiment by Pinchasov et al. (1990) found that 7- to 21-day old chicks fed a 17% CP diet fortified with dEAA had increased FCR and reduced weight gain compared to those fed a 23% CP diet. That study also showed that keeping amino acid concentrations at a constant ratio with CP does not support optimal growth. Other potential remedies for the performance deficiencies observed with reduced CP fortified diets have been investigated with limited results. These include changing the percentage of CP provided by non-essential amino acids (NEAA) versus dEAA (Aletor et al., 2000), increasing or decreasing concentrations of dEAA (Fancher and Jensen, 1989a), and manipulating the dietary acid-base balance (Fancher and Jensen, 1989b). The potential for a non-essential nitrogen deficiency in low CP diets has been investigated by adding L-glutamic acid (Waguespack et al., 2009). The addition of L-glutamic acid did not improve performance in broilers fed low CP diets, indicating a deficiency in an unknown essential or semi-essential amino acid.

A breakthrough study by Dean et al. (2006) found that in broilers fed 16% CP diets, the addition of glycine plus dEAA was required to obtain the same performance as those fed 22% CP diets. Glycine is classified as a non-essential amino acid and has numerous metabolic functions, such as involvement in the formation of uric acid and excretion of N (Coon et al., 1975; Corzo et al., 2004). Interconversion of glycine and serine on an equimolar basis provide the need to express requirements based on glycine equivalence (Gly_{equiv}), where Gly_{equiv} is calculated as the glycine (%) + (0.7143 × serine (%)) (Dean et al., 2006; Kidd and Tillman, 2016). Based on the role of glycine in nitrogen metabolism, it seems that the requirement is likely associated with the level of dietary CP. This was observed by Heger and Pack (1996), who found that the glycine + serine requirement increased as dietary CP increased.

Conversely, there is evidence that the glycine + serine requirement varies depending on the protein source in chicks fed semi-purified diets (Waterhouse and Scott, 1961). Identifying the effects of adding glycine to low CP diets is further complicated by the numerous compounds that can be converted to glycine in-vivo, such as threonine, serine, sarcosine, glycolic acid, choline, betaine, and aminoethanol (Baker and Sugahara, 1970). The number of glycine precursors may explain why Waterhouse and Scott (1961) found that the protein source affected the glycine + serine requirement. The effects of amino acid precursors and the supply of dEAA on the glycine + serine requirement were evaluated in studies using a central composite design (Siegert et al., 2015a; Siegert et al., 2015b). The findings indicate that consideration of dietary cystine, threonine, and choline and the use of Gly_{equiv} values partly explain the large variation in glycine responses observed in the literature. Interestingly, Hofmann et al. (2019) found that a nutrient other than Gly_{equiv} was responsible for the growth limitations in broilers fed very low protein diets of CP at 14.7%. Further research is warranted to identify the responses of feeding very low protein diets in broilers.

Crystalline Amino Acids. The increasing availability of crystalline amino acids has attributed to the dramatic decline in the level of CP found in broiler diets. Several feed grade crystalline amino acids are commercially available for use in animal agriculture, including L-lysine, DL-methionine, L-threonine, L-valine, L-isoleucine, L-arginine, L-histidine, and L-tryptophan. The dietary inclusion rate of crystalline amino acids is dependent upon many factors. Meat yield objectives of a producer and the cost spread of soybean and animal by-product meals, which are the primary protein contributing ingredients, have the greatest effects on the inclusion rate. Additionally, the cost spread of fat and corn can also influence the inclusion of crystalline amino acids (Dozier III and Tillman, 2017). Furthermore, the age, sex, and strain of broiler can change amino acid requirements, making some diets utilize a greater variety or more of a

particular amino acid. Broiler nutritionists utilize shadow pricing in least-cost formulation as a technique that incorporates the cost of ingredients and desired dietary nutrient content to determine the economic viability of including a particular crystalline amino acid. Lastly, a shift to formulating diets on a dEAA basis instead of on a CP basis has increased the precision of formulation and the use of crystalline amino acids.

DL-methionine was the first feed grade amino acid to be readily available in the poultry industry and lead to a significant reduction in diet cost in the 1970s. Lysine was introduced in the 1980s, further reducing cost and improving precision formulation (Georgen and Tintignac, 1982). Use of lysine has increased over the past 20 years as more heavy broilers with high lysine requirements are produced (Dozier III and Tillman, 2017). Feed grade L-threonine was commercially introduced in the late 1990s but was not commonly used in broiler diets until the mid-2000s (Kidd et al., 2013). A dEAA deficiency is induced, and performance is negatively impacted when broilers are fed diets containing L-threonine with no constraint on the next limiting amino acid. This problem was mediated by the widespread utilization of ideal amino acid ratios in diet formulation, increasing the use of L-threonine from 0 to 15 million metric tons between 2000 and 2017, respectively (Dozier III and Tillman, 2017). The development of an accurate threonine estimate has allowed for the optimization of the fourth limiting amino acid, which varies depending on diet ingredient profile. Kidd and Hackenhaar (2006) found that the fourth limiting amino acid was either valine, isoleucine, tryptophan and isoleucine, or arginine depending on the grain source and type of animal by-product used.

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Consumer demand has increased the number of chickens raised in production systems that feed all-vegetable diets (Kidd and Tillman, 2016). In wheat or corn and soybean meal-based diets with no animal by-product meals, valine is the fourth limiting amino acid (Kidd and Hackenhaar, 2006). First introduced in 2008 (Kidd and Tillman, 2012), the production of feed grade L-valine has increased along with the number of broilers raised in all-vegetable production systems. The amount of L-valine used in traditional production systems dependent upon the market price and shadow value. Inclusion of L-valine can reduce CP by upwards of 2.5% (Corzo et al., 2011; Miranda et al., 2014) leading to reduced N excretion and improved precision feeding, but may induce a deficiency in dEAA or NEAA. Corzo et al. (2011) fed 28 to 42-day old broilers diets with six levels of added L-valine from 0 to 1.3 kg/metric ton, in 0.26-kg increments. Broilers fed diets with L-valine at 0.78 kg/metric ton and above had increased FCR and reduced breast meat yield. Conversely, those fed diets with 0.52 and 0.26 kg/metric ton had the same FCR and breast yield as those fed the positive control without L-valine. The author theorized that the negative performance in broilers fed Lvaline at or above 0.78 kg/metric ton was due to a deficiency in arginine, isoleucine, or both. Miranda et al. (2014) fed 0 to 43-day old broilers diets with or without minimum CP restrictions and added L-valine and L-isoleucine. Broilers fed diets without a CP restriction and added L-valine and L-isoleucine had the same FCR, BW gain, and breast yield as those fed diets without a CP restriction and no L-valine and L-isoleucine. These studies show that L-valine can be included and CP can be reduced, as long as the next limiting amino acid is supplied sufficiently.

Modeling amino acid requirements in broilers

Modeling in animal nutrition is an attempt to simulate the growth responses of an animal to varying dietary inputs while accounting for differences in feed intake, diet composition, environmental factors, and other variables. A model can be a combination of experimental findings, circumstantial evidence, and assumption. Thus, the accuracy of a model is dependent upon the reliability of the elements with which it is constructed. In general, a biometrical growth model must contain a "frame" that is formed from a priori knowledge and allows for the determination of relevant input and output variables, the time base, and subsystems. The "structure" of the model must define mathematical relationships independent of algorithmic constants and contain parameters that relate to biological findings of experimental data. Finally, a model must be validated on a replicative, predictive, and structural level (Zoons et al., 1991). Models are broadly described as either empirical or mechanistic, either deterministic or stochastic, or either static or dynamic. Models that produce only single values for a specific age or period are either static, deterministic, or empiric. Dynamic, stochastic, and mechanistic models can predict values as a function of time or age that correspond to the probability distribution of the data and describe the processes involved. Mechanistic models represent underlying mechanisms and are well suited to answer the "what if" questions involved in amino acid nutrition and nutrient partitioning (Zhang and Coon, 1994).

Mathematical modeling can quantitatively describe the effects of varying nutrient concentration while factoring the environmental, metabolic, and genetic inputs on the desired performance or yield response in a population. The Edinburgh Model Pig was the first attempt to simulate performance while considering animal, feed, and environmental parameters (Whittemore, 1976). Pigs are commonly raised in systems that control feed intake, so the development of a broiler model must consider the effects of voluntary feed intake. Variation in dietary amino acid density, environmental temperature, infection, and social stress can all induce changes in feed intake. The Edinburgh Broiler Model is an empirical model based on the Gompertz Equation and was the first to incorporate voluntary feed intake as a predicted variable (Emmans, 1981; Emmans, 1987). The Gompertz Equation describes a growth curve showing the maximum growth at any point in time, given the provided nutritional and environmental conditions and is expressed as:

$$\begin{split} W_t = & W_o e^{(L/K) (1-e(-K t))} \\ Where: \\ W_t = W eight at time t, \\ W_o = Hatch weight, \\ L = Initial growth rate, \\ K = Rate of exponential decay of L. \end{split}$$

The usefulness of the Gompertz Equation and other empirical models become limited when describing the realized growth of muscles, fat, and feathers as a result of genetic potential and the interactions of nutrient intake and environmental factors (Zoons et al., 1991).

Modeling in amino acid nutrition is particularly challenging due to the nonlinearity of growth in response to changes in dietary amino acid concentrations, interactions between amino acids, and variation in metabolism. Furthermore, several methodologies are utilized to determine dEAA requirements such as measuring plasma amino acids, growth variables, carcass analysis, rates of amino acid oxidation, and radioactive isotopes (Oviedo-Rondón and Waldroup, 2002). Ultimately, these studies lead to the development of a fixed requirement value for a particular dEAA based on the parameter set by the experiment (e.g., maximum breast yield, adequate plasma dEAA concentration). These values are useful as dietary minimum inclusion rates in the least cost formulation software, but cannot be implemented into economic and nutritional models. The effects of a range of dietary dEAA concentrations for varying genetic strains on carcass composition, meat yield, performance efficiency, and feed intake are needed to determine the optimal inclusion for the best economic return. To mechanistically model amino acid requirements, the digestibility of feedstuffs, maintenance requirements and, the efficiency of utilization for each amino acid on a strain-specific basis must be determined (HM Edwards 3rd and DH Baker, 1999; Fatufe et al., 2004).

The use of modeling in poultry production has shifted from creating standard growth curves and prediction equations to the development of software used by producers to make formulation decisions. There are several examples of commercially available models that can predict amino acid requirements and make economic decisions. These vary in the amount and type of input data needed for calibration, how closely the model estimates production data, how correctly the model predicts the distribution of data, and how the model handles bias (Harlow and Ivey, 1994). Hurwitz and coworkers developed software known as CHICKPOT TM that functions as an economic optimizer tied to a Gompertz style compartmental model. It utilizes computed

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determinations of daily amino acid and energy requirements for growth and maintenance. The most economic growth trajectory is determined using the feed cost for each increment of daily gain (Hurwitz et al., 1980; Jackson, 1987). The 1994 NRC used this model to produce amino acid requirements for which there was no data (Oviedo-Rondón and Waldroup, 2002). The Ivey Growth Model (IGM®) is a statistical model based upon a large series of feeding trials, tied to an optimizer. The model uses differential equations to determine growth while incorporating the effects of varying nutritional and environmental conditions (Harlow and Ivey, 1994). The Bromley Park Hatcheries Limited (BPHL) model is characterized as mechanistic, determinate, and dynamic. It defines protein retention from experimentally determined first-limiting amino acid coefficients based off the efficiency of utilization and diet concentration, determines the trajectory of feed intake and fat loss using mathematical expressions that are specific to each broiler strain, uses calibration to improve model accuracy, and uses simple multipliers acting on the model to account for changes in feed intake and heat output due to strain and environmental factors. Experimentally, these models have been shown to function with a high degree of accuracy, but have had limited use in commercial settings.

Response to increased amino acid density

Genetic selection for traits that improve meat yield and efficiency has led to the continual reevaluation of the optimum amino acid density needed by modern broilers. As broilers are selected for increased breast yield and weight gain, there is a consequential increase in the level of dietary amino acids needed to sustain growth (Gous, 2010). Furthermore, when determining changes in nutrient requirements of modern broiler strains, it has been observed that increased demand for amino acids is proportionately higher than that for energy (Morris and Njuru, 1990; Kidd et al., 2005; Cerrate and Corzo, 2019). Many studies have evaluated the effects of increasing amino acid density in various phases of production or through the entire life of the bird (Kerr et al., 1999; Kidd et al., 2004; Kidd et al., 2005; Zhai et al., 2013). Changes in metabolism (Kalinowski et al., 2003), allometric growth patterns (Schmidt et al., 2009) and feed intake (Dozier et al., 2008) in response to genetic selection provide the basis for the deviations in nutrient requirements observed between strains. The selection of broilers for increased growth rate and breast development has led to the emergence of some undesirable traits, like skeletal and circulatory disorders (Julian, 1998). Some have suggested that the appearance of woody breast and white striping are also a consequence of selection pressure for accelerated growth rate and increased breast yield. Therefore, the theory has been tested that lowering the nutrient density of a feed can potentially reduce the incidence of muscle myopathies (Meloche et al., 2018).

Performance and Yield Responses. The early post-hatch period provides a nutritional opportunity to enhance the physiological mechanisms that control protein deposition. Feeding suboptimal levels of lysine and other dEAA to chicks can impair muscle development and negatively impact growth. This effect is especially apparent in the pectoralis major, where the concentration of lysine is highest compared to all other

EAA (Hamm, 1981). Everaert et al. (2010) fed broilers high amino acid density diets for five days post-hatch and found that protein translation was upregulated in the pectoralis major. The efficiency of muscle protein deposition, which is the fractional difference between protein synthesis and protein degradation, is as high as 68% in the first-week post-hatch but reduces to 23% at six weeks. This effect is linear and leads to a net reduction in protein accretion and the efficiency of protein retention as the bird ages (Kang et al., 1985). Increasing dietary amino acid density can improve protein synthesis and retention by increasing the production of insulin growth factor 1 (IGF-1) (Urdaneta-Rincon and Leeson, 2004). The supply of dietary amino acids immediately post-hatch is crucial for the development of the pectoralis major and the expression of genes necessary for muscle cell proliferation and differentiation (Velleman et al., 2010).

The physiological improvements observed in young broilers fed high amino acid density diets has translated to improved yield and performance at harvest. For example, Kidd et al. (2005) found that feed efficiency was improved as early as five days posthatch in broilers that were fed increased amino acid density. Furthermore, Kidd et al. (1998) found that high amino acid density diets from 1-18 d improved final BW and breast meat yield at harvest irrespective of the amino acid density fed from 19-50 d. Bodyweight at day seven is the best predictor of BW at harvest (Willemsen et al., 2008). Allometric studies that have examined the relationship between breast weight and BW and have shown that the two increase proportionately (Zuidhof, 2005). During the first two weeks of post-hatch development, feed intake accounts for only 6% of the cumulative intake of a 55-day old broiler. The improvements in meat yield and

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performance along with the low level of feed intake, make the starter phase the opportune time to invest in increasing dietary amino acid density.

For the duration of the growing cycle, broilers exhibit high allometric growth in the pectoralis compared to other muscles. For example, from 0-42 days, the allometric growth ratio of the breast muscle was 1.36 compared to 1.10 for drumsticks and 1.06 for thighs. Furthermore, the allometric growth ratio of the breast muscles from 14 to 35 days of age was 1.25 in birds selected for breast meat yield compared to 1.09 in unselected birds (Govaerts et al., 2000). This indicates that at later ages, a large portion of the amino acid intake above maintenance is partitioned to the growth of the breast muscles as opposed to other muscle groups. The increasing level of feed intake that occurs in broilers as they reach market age can drive producers to reduce the amino acid density in later phases. This may be an economically unfavorable feeding strategy, as protein accretion occurs in the breast muscle throughout the life of the bird. Zuidhof (2005) found that breast weight continued to increase linearly up to 112 days of age in broilers.

Furthermore, amino acids in breast muscles are more readily mobilized during times of dietary deficiency than those in the leg or thigh. Tesseraud et al. (1996a) measured protein fractional synthesis and breakdown rates in 2-, 3- and 4-week-old broilers that were fed lysine deficient diets. Lysine deficiency reduced tissue deposition at all ages in the pectoralis major, sartorius, and anterior latissimus dorsi. The pectoralis major was the most sensitive muscle to lysine deficiency. Another study found similar responses, when broilers that were selected for increased breast meat yield had greater fractional breakdown rates in the pectoralis compared to the unselected line. No differences were observed between lines in the metabolism of the sartorius muscle in response to lysine deficiency (Tesseraud et al., 2001). These studies indicate that reducing amino acid density at all ages can potentially reduce breast meat yield by negatively impacting protein metabolism.

A producer must continuously assess the economic viability of feeding a certain amino acid density and determine how it affects breast meat yield and performance. Therefore, establishing the responsiveness of a particular broiler strain to a variety of densities is essential for accurate decision making. A substantial amount of evidence exists that shows increasing amino acid density leads to improvements in breast meat yield and performance in various broiler strains. For example, Kidd et al. (2004) fed $Ross \times Ross$ 508 broilers three levels of amino acid density to 49 d and found that BW was increased by 607 grams, FCR was reduced by 42 points, and breast meat yield was increased by 3% in those fed high versus low amino acid density diets. Additionally, increasing amino acid density at later ages can improve performance and yield at harvest. Dozier III et al. (2007b) fed Ross 708 broilers increased amino acid density diets from 36 to 60 days of age and found that breast meat yield was increased by 0.53%. Conversely, Maynard et al. (2018) fed Cobb $700 \times MV$ broilers increased amino acid density and observed improvements in feed conversion but no changes in breast meat yield. Increasing amino acid density throughout the life of the bird leads to improvements in meat yield and performance in most strains. Therefore, it is essential to identify the responses of individual strains to increased amino acid density to ensure the economic viability of a feeding program.

Muscle Myopathies. Over the past decade, the prevalence of the breast muscle myopathies woody breast and white striping have increased (Meloche et al., 2018). Woody breast is characterized by an abnormal accumulation of collagen, other noncontractile proteins, and fluid in the pectoralis major, which gives the breast a palpable hardness (Sihvo et al., 2017). Microscopic evaluation of breast tissue afflicted with white striping indicates an increase in the deposition of intramuscular fat, which forms visible striations (Kuttappan et al., 2012). The myopathies have been anecdotally associated with increased growth rate and breast meat yield, leading to the theory that reducing dietary amino acid and energy density may ameliorate breast muscle myopathies. This theory was tested by Meloche et al. (2018) by feeding Yield Plus \times Ross 708 broilers treatment diets with 5% and 10% reductions in energy and amino acid density from 1 to 48 d of age. Broilers were fed either 100, 95, or 90% of the recommended nutrient density through the entire study or nutrient density was reduced to 95% or 90% in the starter alone or in both the starter and grower. The experimental design enabled the authors to test the effects of age and length of time in which the nutrient density was reduced on the incidences of breast muscle myopathies. Broilers that were fed the reduced density diets during the starter phase had increased severity of woody breast due to the effects of compensatory gain. Decreasing nutrient density to 90% of the recommendation from 1 to 48 d of age reduced the severity of breast muscle myopathies, but also worsened performance. Based off these findings it appears that reducing nutrient density is not a viable strategy for controlling the severity of breast muscle myopathies. This finding may be supported by the nature of amino acid partitioning in varying

tissues. Bailey and Light (1989) noted that collagen is high in glycine (21%) and proline (11%) and low in lysine (3.5%). When chicks are fed lysine levels near maintenance, there is an inverse relationship in whole-body amino acid composition, where glycine and proline are increased, and lysine is decreased (HM Edwards 3rd and DH Baker, 1999). This is indicative of a reduction in the synthesis of contractile proteins and an increase in the synthesis of collagen. Since woody breast is defined by the buildup of collagen in the pectoralis major, reducing amino acid density and dietary lysine content may exacerbate the incidence or severity.

Some have suggested that woody breast may be a symptom of increased oxidative stress and that increasing the dietary supply of an exogenous antioxidant or supplying more cofactors for endogenous antioxidant production will improve breast muscle quality. To test this theory, Bodle et al. (2018) evaluated the dietary effects of increasing the ratio of digestible arginine (dArg): dLys, supplementing with vitamin C, doubling the vitamin inclusion rate, reducing amino acid density by 15% in the grower phase, or a combination of all four previously stated treatments. No differences in 49day performance were observed. Feeding a higher ratio of dArg: dLys, higher vitamin C, or lower dEAA in the grower phase reduced the severity of woody breast. There has been no single dietary strategy to reduce the severity of breast myopathies. Improvements in breast quality due to feeding lower energy or amino acid density are more likely due to the reduction in growth rate or weight at slaughter as opposed to the effects of a particular nutrient. Beyond growth rate and final BW, the only finite association is that genetic strains selected for high breast yield have greater occurrences of myopathies (Petracci et al., 2019).

The continual selection of broilers for improved growth, efficiency, and yield make identifying the optimal nutrient density a dynamic decision. Several advances in amino acid nutrition have allowed producers to more precisely feed broilers to reduce nitrogen excretion and maximize profitability. These advances include the implementation of the ideal protein concept, the increased production of crystalline amino acids, and the development of models that can predict the economic outcome of different feeding strategies. However, experimentally determining the responsiveness of newly developed broiler strains to feeding varying nutrient densities continues to be an important facet in sustaining the economic efficiency of broiler production. The purpose of the research described herein was to evaluate the performance and processing yield responses of the Cobb $700 \times MV$ broiler to feeding diets with increasing amino acid and energy densities.

CHAPTER III

THE EFFECT OF NUTRIENT DENSITY ON PERFORMANCE AND PROCESSING YIELD IN COBB 700 \times MV MIXED-SEX BROILERS

Introduction

Over the last 20 years, the demand for breast fillets and value-added products has contributed to the large increase in the number broilers processed over 3.5 kg in the United States (Dozier III et al., 2007b). Producers must feed diets containing amino acid and energy dense ingredients in order to sustain the rapid growth of modern broilers. To maintain profitability and performance, it is vital to formulate diets with neither a deficiency nor excess in amino acids and energy. Furthermore, approximately 70% of the cumulative feed intake for a broiler occurs between 2.0 to 3.6 kg of live weight (Dozier III et al., 2007a). The high costs of feeding heavy broilers make identifying the nutrient requirements of newly developed strains essential to enhance economic performance.

The Cobb 700 × MV broiler is a strain selected to maximize feed efficiency and breast meat yield when raised to a heavier body weight (BW) (Cobb-Vantress, 2019). As broilers are selected for increased breast yield and weight gain, there is a consequential increase in the level of dietary amino acids and energy needed to sustain growth (Gous, 2010). When determining changes in nutrient requirements of modern broilers, it has been observed that increased demand for amino acids is proportionately higher than that for energy (Morris and Njuru, 1990; Kidd et al., 1998; Gous, 2010). This trend coincides with the goals of genetic selection, where heavy broilers are selected to more efficiently assimilate amino acids while reducing energy expended on maintenance and fat accumulation. Nonetheless, determining the optimum AME ensures efficient utilization of dietary amino acids (Jackson et al., 1982; Leeson et al., 1996; Gous, 2010; Gous et al., 2018). Changes to feed intake (Dozier III et al., 2008), allometric growth patterns (Schmidt et al., 2009), and metabolism (Kalinowski et al., 2003) in response to genetic selection provide the basis for the deviation in nutrient requirements observed between strains.

The United States broiler industry utilizes nutritional strategies on a strain specific basis to minimize diet cost and maximize meat yield. Studies that have evaluated the responsiveness of the Cobb 700 × MV to feeding diets with varying amino acid and energy densities have shown confounding results (Johnson, 2018; Maynard et al., 2018; Philpot, 2018; Maynard et al., 2019). Research presented by Philpot (2018) and Johnson (2018) found that increasing amino acid density improved breast meat yield and feed efficiency. Conversely, Maynard et al. (2018); (Maynard et al., 2019) found that feeding diets with increased amino acid density and AME content did not impact breast meat yield or feed efficiency. Research with other strains of heavy broilers have found that increased amino acid density improves breast meat yield, feed efficiency, and growth rate, while reducing fat yield (Kerr et al., 1999; Kidd et al., 2004; Kidd et al., 2005; Zhai et al., 2013; Zhai et al., 2016). The response of heavy broilers to changes in dietary AME has provided inconsistent results. Some studies have shown that increasing AME, which is achieved by feeding higher levels of dietary fat, leads to improved feed conversion without detrimental effect on meat yield (Saleh et al., 2004). Whereas other research has found that increasing AME negatively effects breast meat yield or has no effect on growth rate (Dozier III et al., 2007a; Dozier III and Gehring, 2014). It has been suggested that modern boilers are able to adjust feed intake in response to changes in dietary AME (Leeson et al., 1996; Hidalgo et al., 2004; Dozier III et al., 2007a). While this effect can lead to improved feed efficiency, it may also reduce amino acid intake. Reducing amino acid intake throughout the production cycle can negatively impact growth, feed efficiency and meat yield.

Response to amino acid density is dependent upon the strain and selection criteria in question. Studies evaluating dietary amino acid density show that breast meat yield is optimized at a higher lysine estimate than growth (Kerr et al., 1999). These estimates are further varied by the strain of broiler being evaluated (Acar et al., 1991; Bilgili et al., 1992). Lysine makes up approximately 8% of the crude protein in the pectoralis major, indicating that the amino acid composition of breast meat contributes to the high demand for dietary lysine (Hamm, 1981; Saleh et al., 2004). Primary breeders select heavy broilers for increased breast meat yield, growth and feed efficiency (Tesseraud et al., 1999). In order to meet the genetic potential and maximize profitability of newly developed strains, it is crucial to continuously evaluate the optimal dietary amino acid and energy density. Therefore, the objective of this study was to measure the impact of feeding diets with varying amino acids and energy densities on the performance and processing yield of the Cobb $700 \times MV$.

Materials and Methods

Diets and Dietary Treatments. Dietary treatments were formulated on a least cost basis and were primarily composed of corn, soybean meal, corn DDGS and pork meat and bone meal. Ingredients were acquired by the Texas A&M Poultry Science Department feed mill, where composite samples of corn, soybean meal, corn DDGS and pork meat and bone meal were analyzed by near infrared spectroscopy to determine amino acid content. The total amino acid content of each ingredient was multiplied by a digestibility coefficient to determine the digestible amino acid content. The digestible amino acid values for each ingredient were used to formulate all dietary treatments. Diets were manufactured at the research feed mill using a single-ribbon horizontal mixer and a steam pellet mill. During the starter phase, all dietary treatments were pelleted and then crumbled using a roller mill. In all feeding phases, triplicate feed samples were collected from each dietary treatment. A composite sample for each treatment was then submitted for proximate nutrient and amino acid analysis (Carat-Adisseo Analytical Laboratory, Antony, France)

Diets were manufactured in four-phases: starter (d 0-12), grower (d 12-26), finisher (d 26-36) and withdrawal (d 36-49). During the starter phase, all broilers were fed a common diet that contained 1.18% dLys. Beginning in the grower phase, broilers were fed one of six dietary treatments. Treatments were arranged in a 2×3 factorial, with two AME levels and three amino acid densities. Of the two AME levels, the low AME was based off current industry feeding practices for the Cobb 700 × MV. The high AME level consisted of low AME + 110 kcal/kg. Three amino acid densities were used and consisted of a medium level, a low level (medium -7.5% dLys) and a high level (medium +7.5% dLys). Treatments were formulated to maintain equivalent dEAA: dLys ratios across all diets within a given phase. All birds were fed a common starter from 0-12 d and then assigned one of six treatments: low amino acid and low AME (Laa \times Len), moderate amino acid and low AME (Maa \times Len), high amino acid and low AME $(Haa \times Len)$, low amino acid and high AME $(Laa \times Hen)$, moderate amino acid and high AME (Maa \times Hen) or high amino acid and high AME (Haa \times Hen). The Maa dLys concentration was 1.07, 0.96, and 0.89% for the grower, finisher and withdrawal feeding phases, respectively. Modifying the amino acid density of treatment diets was done by increasing or decreasing the amount of crystalline amino acids and soybean meal. In order to increase the energy density of the high AME diets, the amount of corn oil was increased compared to the control. Meat and bone meal and corn DDGS were included at a constant level across all treatments within a given phase. Diets were conditioned before pelleting and pellets were 4.4 mm in diameter.

Bird Husbandry. On the day of hatch, 2,160 Cobb $700 \times MV$ mixed sex broiler chicks were obtained from a commercial hatchery, where they were vaccinated for Newcastle and Marek's disease. Chicks were then feather-sexed and 18 males and 18 females were placed into each replicate pen at the Texas A&M University poultry research farm. The experiment consisted of six treatments with 10 replicates per treatment, arranged in a randomized complete block design. Broilers were raised in a

tunnel ventilated research barn and each pen contained two tube feeders, a nipple drinker line, and shaved pine litter. Feed and water were provided to broilers on an ad-libitum basis. Barn temperature was recorded daily and adjusted in response to bird comfort. The trial was conducted from November to December, which minimized heat stress. The lighting program consisted of 24 h of light at 2 foot candles on day 0-3; 23 h of light at 2 foot candles on d 3-8; 16 h light at 0.75 foot candle on d 9-18; 18 h of light at 0.1 foot candle on d 19-33; and 20 h light at 0.05 foot candle on d 33 to termination. Broilers were raised in accordance with procedures approved by the Texas A&M University Institutional Animal Care and Use Committee.

Measurements. Bird weight and feed intake were measured by pen at the end of each feeding phase on days 12, 26, 36 and 49. Mortality was recorded daily and used as an adjustment factor when calculating feed conversion. Average body weight (BW), g/bird/d feed intake, percent mortality and mortality adjusted feed conversion ratio (FCR) were determined by phase and on a cumulative basis. At 49 days of age, male and female birds were weighed separately in order to determine the average bird weight for both sexes of each pen. Average lysine intake (g/bird) was determined by multiplying average replicate feed intake (g/bird) by the calculated dietary dLys content for each feeding phase. The average lysine intake (g/bird) of each phase was then summed to determine the total lysine intake (g/bird). At 50 days of age, 4 males and 4 females were selected for processing from each pen based off the predetermined average bird weights. Individual live weight was determined, and birds were electrically stunned, bled,

scalded, mechanically plucked and manually eviscerated. Fat pad and carcass weight was measured for each bird. Carcasses were chilled in an immersion tank for 60 min at 1° C and manually agitated at regular intervals. After chilling, carcasses were deboned and the weight of legs, thighs, skinless-breast fillet and tender fillet were determined for each bird. The severity of woody breast (WB) was determined for each breast fillet using tactile evaluation as described by Tijare, et al. [30].

Statistics. All data were analyzed by one-way ANOVA via the GLM model with means considered significantly different at $p \le 0.05$. Replicate pens were defined as the experimental unit. Means that were deemed significantly different were separated using Duncan's Multiple Range Test. Parts yield was evaluated based on percent live weight, where the weight of the part was divided by the live weight. The mean feed efficiency and percentage of breast fillet yield for each replicate pen was regressed with the average per bird lysine intake using linear regression analysis and considered significant at $p \le 0.05$. In both regression analyses, replicates were divided by AME treatment and a separate line was fitted to 30 replicates from either Len or Hen. The slope of the line represented the improvement in either breast meat yield or feed efficiency for each gram increase in lysine intake. The coefficient of determination (R-squared) was used to establish the goodness of fit for replicate means to the regression line.

Results

Dietary Analysis. The amino acid analysis (Tables 1, 2, 3 & 4) show that the experimental diets generally provided the desired nutrient variation between treatments. The goal of diet manufacture with respect to amino acid density was to increase or decrease the dLys content of M*aa* by 0.075% in H*aa* and L*aa*, respectively. The average percentage difference of analyzed lysine values between H*aa* vs. M*aa* and L*aa* vs. M*aa* was 0.071% with the lowest being 0.058% and the highest being 0.089%. Additionally, proximate analysis of the experimental diets indicates that the desired variation in AME was achieved. This is evident by the consistent increase in crude fat observed by H*en* vs. L*en* across all feeding phases.

	Starter
Ingredient (%)	Common for all treatments
Corn	59.51
Soybean meal	58.57
Meat and Bone	4.00
Corn DDGS ¹	5.00
DL- Methionine	0.29
L-Lysine HCL	0.22
L-Threonine	0.09
Corn Oil	0.90
Limestone	0.71
Phosphate	0.03
Vitamins ²	0.03
Trace Minerals ³	0.03
NSPase ⁴	0.03
Nicarbazin ⁵	0.04
Calculated Nutrient Content	
ME, kcal/kg	3003
CP, %	21.24
Crude Fat, %	4.60
dig-Met, %	0.60
dig-Lys, %	1.18
dig-TSAA, %	0.89
dig-Thr, %	0.77
dig-Val, %	0.91
dig-Arg, %	1.27
Avail. P, %	0.44
Ca, %	0.88
Na, %	0.18
Analyzed Nutrient Content (%)	
Ash	4.70
Crude Fat	3.10
Lysine	1.38
Methionine	0.62
Threonine	0.91
Arginine	1.40
Valine	1.11

Table III-1. Dietary formulations, calculated nutrient content, and analyzed nutrient content of diets fed to Cobb 700 X MV mixed-sex broilers during the starter phase.

¹Corn Distillers dried grains with solubles.

¹Corn Distillers dried grains with solubles.
 ²The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin.
 ³The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus*. ⁵Nicarbazin 25% was used for prevention of coccidiosis.
 ⁴The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase. 1765 U Subtilisin.

⁴The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

	Grower			Grower			
Ingredient	$Laa \times Len$	$Maa \times Len$	$Haa \times Len$	$Laa \times Hen$	$Maa \times Hen$	Haa imes Hen	
Corn	66.49	63.46	59.68	64.89	61.12	57.30	
Soybean meal	21.24	24.34	27.52	21.42	24.61	27.82	
Meat & Bone	4.00	4.00	4.00	4.00	4.00	4.00	
DDGS ¹	5.00	5.00	5.00	5.00	5.00	5.00	
DL-Met	0.21	0.25	0.28	0.21	0.25	0.29	
L-Lysine	0.21	0.21	0.21	0.21	0.21	0.21	
L-Threonine	0.07	0.08	0.09	0.07	0.08	0.09	
Corn Oil	0.77	1.12	1.73	2.58	3.19	3.81	
Limestone	0.83	0.83	0.82	0.83	0.82	0.82	
Phosphate	0.14	0.09	0.05	0.14	0.09	0.05	
Vitamins ²	0.02	0.02	0.02	0.02	0.02	0.02	
Trace Min ³	0.06	0.06	0.06	0.06	0.06	0.06	
NSPase ⁴	0.03	0.03	0.03	0.03	0.03	0.03	
Nicarbazin ⁵	0.04	0.04	0.04	0.04	0.04	0.04	
Calculated Nut	rient Content						
ME, kcal/kg	3062	3062	3062	3172	3172	3172	
CP, %	18.30	19.54	20.76	18.27	19.49	20.73	
Crude Fat, %	4.60	4.89	5.41	6.33	6.85	7.38	
dig-Met, %	0.48	0.53	0.58	0.49	0.53	0.58	
dig-Lys, %	0.99	1.07	1.15	0.99	1.07	1.15	
dig-TSAA, %	0.74	0.80	0.86	0.74	0.80	0.86	
dig-Thr, %	0.64	0.70	0.75	0.64	0.70	0.75	
dig-Val, %	0.78	0.83	0.89	0.78	0.83	0.89	
dig-Arg, %	1.06	1.15	1.23	1.06	1.15	1.24	
Avail. P, %	0.44	0.44	0.44	0.44	0.44	0.44	
Ca, %	0.92	0.92	0.92	0.92	0.92	0.92	
Na, %	0.17	0.17	0.17	0.17	0.17	0.17	
Analyzed Nutr	ient Content (%)					
Ash	5.0	4.6	4.7	4.3	4.6	4.7	
Crude Fat	4.4	5.2	5.7	6.5	7.5	7.3	
Lysine	1.15	1.25	1.27	1.14	1.22	1.30	
Methionine	0.53	0.56	0.61	0.50	0.55	0.61	
Threonine	0.76	0.84	0.90	0.80	0.89	0.93	
Arginine	1.17	1.26	1.36	1.25	1.31	1.40	
Valine	0.93	1.02	1.05	0.96	1.00	1.05	

Table III-2. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the grower phase.

¹Corn Distillers dried grains with solubles.

²The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B_{12} , 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin.

³The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus*.

⁴The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

⁵Nicarbazin 25% was used for prevention of coccidiosis.

	Finisher			Finisher			
Ingredient	$Laa \times Len$	$Maa \times Len$	$Haa \times Len$	$Laa \times Hen$	$Maa \times Hen$	$Haa \times Hen$	
Corn	70.88	68.61	65.06	69.66	66.27	62.60	
Soybean meal	17.93	20.66	23.64	18.07	20.93	24.02	
Meat & Bone	3.00	3.00	3.00	3.00	3.00	3.00	
DDGS ¹	5.00	5.00	5.00	5.00	5.00	5.00	
DL- Met	0.18	0.21	0.24	0.18	0.22	0.24	
L-Lysine	0.21	0.21	0.21	0.21	0.21	0.21	
L-Threonine	0.07	0.08	0.09	0.07	0.09	0.09	
Corn Oil	0.75	0.90	1.48	2.42	2.97	3.57	
Limestone	0.82	0.82	0.82	0.82	0.82	0.82	
Phosphate	0.07	0.03	0.00	0.07	0.03	0.00	
Vitamins ²	0.01	0.01	0.01	0.01	0.01	0.01	
Trace Min ³	0.04	0.04	0.04	0.04	0.04	0.04	
NSPase ⁴	0.03	0.03	0.03	0.03	0.03	0.03	
Calculated Nut	rient Content						
ME, kcal/kg	3106	3106	3106	3197	3197	3197	
CP, %	16.53	17.65	18.79	16.51	17.61	18.79	
Crude Fat, %	4.57	4.68	5.18	6.18	6.65	7.15	
dig-Met, %	0.43	0.48	0.52	0.43	0.48	0.52	
dig-Lys, %	0.89	0.96	1.03	0.89	0.96	1.03	
dig-TSAA, %	0.68	0.73	0.79	0.68	0.73	0.79	
dig-Thr, %	0.60	0.64	0.69	0.60	0.64	0.69	
dig-Val, %	0.71	0.76	0.81	0.71	0.76	0.81	
dig-Arg, %	0.94	1.02	1.10	0.94	1.02	1.10	
Avail. P, %	0.38	0.38	0.38	0.38	0.38	0.38	
Ca, %	0.80	0.80	0.80	0.80	0.80	0.80	
Na, %	0.17	0.17	0.17	0.17	0.17	0.17	
Analyzed Nutri	ient Content (
Ash	4.8	4.2	4.4	4.1	4.2	4.4	
Crude Fat	4.5	4.5	5.2	6.3	6.9	7.6	
Lysine	1.01	1.07	1.17	0.99	1.07	1.15	
Methionine	0.49	0.51	0.54	0.48	0.52	0.56	
Threonine	0.73	0.75	0.85	0.70	0.78	0.82	
Arginine	1.10	1.14	1.28	1.08	1.17	1.26	
Valine	0.85	0.87	0.95	0.83	0.89	0.96	

Table III-3. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the finisher phase.

¹Corn Distillers dried grains with solubles..

 2 The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin.

³The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus*.

⁴The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

	Withdrawal			Withdrawal			
Ingredient	$Laa \times Len$	$Maa \times Len$	$Haa \times Len$	$Laa \times Hen$	$Maa \times Hen$	Haa imes Hen	
Corn	72.80	70.93	68.44	72.61	69.44	65.98	
Soybean meal	15.49	17.99	20.70	15.51	18.16	21.08	
Meat & Bone	3.00	3.00	3.00	3.00	3.00	3.00	
DDGS ¹	5.00	5.00	5.00	5.00	5.00	5.00	
DL- Met	0.15	0.18	0.21	0.15	0.18	0.21	
L-Lysine	0.21	0.21	0.21	0.21	0.21	0.21	
L-Threonine	0.06	0.07	0.08	0.06	0.07	0.08	
Corn Oil	0.78	0.83	1.08	2.09	2.61	3.17	
Limestone	0.82	0.82	0.82	0.82	0.82	0.82	
Phosphate	0.11	0.07	0.03	0.11	0.07	0.03	
Vitamins ²	0.01	0.01	0.01	0.01	0.01	0.01	
Trace Min ³	0.03	0.03	0.03	0.03	0.03	0.03	
NSPase ⁴	0.03	0.03	0.03	0.03	0.03	0.03	
Calculated Nut	rient Content						
ME, kcal/kg	3118	3118	3118	3227	3227	3227	
CP, %	15.52	16.56	17.65	15.51	16.54	17.64	
Crude Fat, %	4.62	4.66	4.85	5.92	6.36	6.83	
dig-Met, %	0.39	0.43	0.47	0.39	0.43	0.47	
dig-Lys, %	0.82	0.89	0.96	0.82	0.89	0.96	
dig-TSAA, %	0.63	0.68	0.73	0.63	0.68	0.73	
dig-Thr, %	0.55	0.60	0.64	0.55	0.60	0.64	
dig-Val, %	0.66	0.71	0.76	0.66	0.71	0.76	
dig-Arg, %	0.87	0.94	1.02	0.87	0.94	1.02	
Avail. P, %	0.38	0.38	0.38	0.38	0.38	0.38	
Ca, %	0.80	0.80	0.80	0.80	0.80	0.80	
Na, %	0.17	0.17	0.17	0.17	0.17	0.17	
Analyzed Nutri	ient Content (,					
Ash	5.3	4.8	4.3	4.2	4.1	4.3	
Crude Fat	4.9	4.7	4.8	5.6	6.0	6.3	
Lysine	0.97	1.04	1.10	0.98	1.05	1.13	
Methionine	0.42	0.47	0.50	0.45	0.46	0.52	
Threonine	0.69	0.74	0.80	0.69	0.73	0.77	
Arginine	1.03	1.13	1.19	1.03	1.11	1.20	
Valine	0.81	0.88	0.91	0.80	0.85	0.92	

Table III-4. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the withdrawal phase.

¹Corn Distillers dried grains with solubles.

 2 The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin.

³The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus*.

⁴The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

Performance. Main effects for live performance responses as affected by dietary amino acid and AME density are presented in tables 5, 6 and 7. On day 26, an interaction between amino acid and AME was present, where feeding $Laa \times Len$ reduced (P \leq 0.05) BW compared to the other treatments. The main effect of feeding Laa diets reduced ($P \le 0.05$) d 26 BW compared to broilers fed Maa and Haa. Increasing the amino acid density continued to improve BW at d 37 and 49. Broilers fed Haa were 50 and 70 grams heavier than those fed Laa on days 37 and 49, respectively. Increasing the AME content of the diets led to a reduction ($P \le 0.05$) in feed intake (g/bird/d) during the grower phase in broilers fed Haa and Hen diets compared to those fed Laa and Len diets. Mortality adjusted feed conversion was the most responsive parameter to increasing amino acid and AME density, and no interaction between amino acid and AME was observed. In the grower and finisher phases and from days 1-26, 1-37 and 1-49, a stepwise reduction ($P \le 0.05$) in FCR coincided with increasing amino acid density. The stepwise improvement in FCR was most dramatic from days 1-49, where feeding diets containing Maa vs. Laa and Haa vs. Laa led to a 5.4 and 9 point reduction $(P \le 0.05)$, respectively. Increasing AME density improved $(P \le 0.05)$ FCR during the grower and finisher phases and from days 1-26, 1-37 and 1-49. No treatment effects on mortality were observed.

Treatment ¹				Body Weight (kg)					
AA	Energy	Day 0	Day 12	Day 26	Day 37	Day 49			
High	High	0.038	0.353	1.327	2.326	3.497			
High	Low	0.038	0.349	1.311	2.297	3.445			
Low	High	0.037	0.349	1.277	2.257	3.373			
Low	Low	0.037	0.352	1.305	2.271	3.416			
Medium	High	0.038	0.352	1.316	2.293	3.451			
Medium	Low	0.038	0.352	1.306	2.287	3.453			
			I	Amino Acid Densit	у				
High	-	0.038	0.351	1.319 ^b	2.312 ^b	3.471 ^b			
Low	-	0.037	0.351	1.291ª	2.264 ^a	3.395 ^a			
Medium	-	0.038	0.352	1.311 ^b	2.290 ^{ab}	3.452 ^{ab}			
		Energy Density							
-	High	0.038	0.351	1.307	2.292	3.440			
-	Low	0.038	0.351	1.307	2.285	3.438			
				— Probability —					
Treatment									
AA		0.11	0.77	0.01	0.05	0.04			
Energy		0.64	0.88	0.95	0.65	0.93			
AA × Energy		0.60	0.29	0.03	0.54	0.31			
SEM		0.000	0.001	0.004	0.008	0.013			

Table III-5. Body weight (kg) of Cobb 700 × MV mixed-sex broilers fed diets containing varying nutrient density to 49 d.

¹ Treatments within AA column are high (medium +7.5% dLys), medium, and low (medium +7.5% dLys) fed to Cobb × MV from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: EAA across all treatments. Treatments within the energy column are low and high (low +110 kcal/kg).

Treatment ¹				H	Feed Conversion F	Ratio ²		
AA	Energy	Starter	Grower	Finisher	Withdrawal	d 1-26	d 1-36	d 1-49
High	High	1.250	1.492	1.700	2.269	1.432	1.545	1.780
High	Low	1.260	1.520	1.710	2.309	1.455	1.565	1.809
Low	High	1.246	1.583	1.752	2.392	1.497	1.608	1.861
Low	Low	1.261	1.598	1.807	2.498	1.513	1.638	1.888
Medium	High	1.259	1.513	1.723	2.302	1.449	1.566	1.809
Medium	Low	1.256	1.558	1.748	2.294	1.482	1.696	1.832
					Amino Acid Den	sity		
High	-	1.255	1.506 ^a	1.705 ^a	2.289ª	1.444 ^a	1.555ª	1.794 ^a
Low	-	1.253	1.590 ^c	1.779 ^c	2.395 ^b	1.505 ^c	1.623°	1.874 ^c
Medium	-	1.257	1.535 ^b	1.735 ^b	2.298ª	1.466 ^b	1.581 ^b	1.820 ^b
					Energy Densit	у		
-	High	1.252	1.529 ^a	1.725 ^a	2.321	1.459 ^a	1.573 ^a	1.817ª
-	Low	1.259	1.558 ^b	1.755 ^b	2.334	1.483 ^b	1.600 ^b	1.843 ^b
					Probability			
Treatment								
AA		0.87	≤ 0.01					
Energy		0.28	≤ 0.01	≤ 0.01	0.66	≤ 0.01	≤ 0.01	≤ 0.01
AA × Energy		0.50	0.14	0.16	0.78	0.39	0.62	0.94
SEM		0.003	0.006	0.007	0.015	0.004	0.005	0.006

Table III-6. Feed conversion ratio of Cobb $700 \times MV$ mixed-sex broilers fed diets containing varying nutrient density to 49 d.

¹ Treatments within AA column are high (medium +7.5% dLys), medium, and low (medium +7.5% dLys) fed to Cobb × MV from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: EAA across all treatments. Treatments within the energy column are low and high (low +110 kcal/kg). ²Mortality corrected feed conversion ratio.

Treatment ¹			Feed Int	ake (g/bird/	(d)				Mortality (%)	
AA	Energy	Starter	Grower	Finisher	Withdrawal	Star	ter	Grower	Finisher	Withdrawal	Total
High	High	32.4	105.0	188.4	221.5	3.	1	1.3	1.6	1.5	6.9
High	Low	32.5	105.4	187.9	224.0	2.	5	1.2	0.9	2.9	7.0
Low	High	31.8	105.2	190.8	223.1	4.	5	0.3	0.6	1.6	6.7
Low	Low	32.8	109.4	194.5	228.2	2.	2	0.6	0.9	0.6	4.2
Medium	High	32.4	105.0	187.8	224.7	4.	5	1.5	0.6	1.6	7.8
Medium	Low	32.3	106.6	190.9	222.8	4.	4	0.6	1.2	0.3	6.4
					Amir	no Acid	Dens	ity			
High	-	32.4	105.2ª	188.2	222.7	2.	8	1.3	1.3	2.2	7.0
Low	-	32.3	107.3 ^b	192.6	225.6	3.	4	0.5	0.7	1.1	5.4
Medium	-	32.4	105.8 ^{ab}	189.4	223.7	4.	5	1.1	0.9	0.9	7.1
					Er	ergy De	nsity				
-	High	32.2	105.1ª	189.0	223.1	4.	0	1.0	0.9	1.6	7.1
-	Low	32.5	107.1 ^b	191.1	225.0	3.	1	0.8	1.0	1.3	5.8
		Probability									
Treatment											
AA		0.89	0.03	0.06	0.49	0.2	.9	0.33	0.54	0.17	0.40
Energy		0.19	≤ 0.01	0.18	0.33	0.2	8	0.63	0.85	0.60	0.24
$AA \times Energy$		0.16	0.06	0.50	0.36	0.5	6	0.52	0.36	0.15	0.64
SEM		0.10	0.40	0.90	1.10	0.4	-0	0.20	0.20	0.30	0.50

Table III-7. Feed intake (g/bird/d) and mortality (%) of Cobb $700 \times MV$ mixed-sex broilers fed diets containing varying nutrient density to 49 d.

 1 Treatments within AA column are high (medium +7.5% dLys), medium, and low (medium +7.5% dLys) fed to Cobb × MV from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: EAA across all treatments. Treatments within the energy column are low and high (low +110 kcal/kg).

Processing Yield. Main effects of 50 d carcass traits as affected by dietary amino acid and AME density are shown in table 8. Changing the amino acid density or AME content of experimental diets did not lead to changes in BW or carcass yield of broilers selected for processing. An incremental increase ($P \le 0.05$) in boneless-skinless breast meat yield was observed with each increase in amino acid density. Broilers that were fed Maa had a 0.6% and Haa a 1.4% increase ($P \le 0.05$) in breast meat yield compared to those fed Laa. Tenderloin yield in broilers fed Haa was 0.26% higher ($P \le 0.05$) than those that were fed Laa. Tenderloin yield was not different in broilers fed Maa compared to Laa. Increasing AME density negatively impacted breast and tenderloin yield. Broilers that were fed Hen had a 0.5% and 0.11% reduction ($P \le 0.05$) in breast and tender yield, respectively. Changing amino acid and AME density did not change wing or leg yield. An incremental reduction ($P \le 0.05$) in fat pad yield was observed with each increase in amino acid density. Fat pad yield was reduced ($P \le 0.05$) in broilers that were fed Haa by 0.40% and Maa by 0.21% compared to those fed Laa. The presence of woody breast was not changed by modifying the amino acid or AME density and the average score across all treatments was 1.06.

Treatment ²		Meat Quality			Yield (9	%, live)		
AA	Energy	Woody Breast	Carcass	Breast	Tender	Wing	Leg	Fat Pad
High	High	1.23	75.7	23.8	4.82	7.65	23.4	1.74
High	Low	1.16	77.7	24.5	5.04	7.85	24.1	1.68
Low	High	1.14	75.2	22.5	4.63	7.69	23.6	2.13
Low	Low	1.09	75.8	23.0	4.72	7.67	23.7	2.08
Medium	High	1.19	75.9	23.2	4.77	7.67	23.5	2.01
Medium	Low	1.16	76.0	23.6	4.78	7.69	23.5	1.83
				Amino A	Acid Density			
High	-	1.20	76.7	24.2 ^c	4.93 ^b	7.75	23.7	1.71 ^a
Low	-	1.11	75.5	22.8ª	4.67 ^a	7.68	23.6	2.11 ^c
Medium	-	1.17	75.9	23.4 ^b	4.78^{a}	7.68	23.5	1.92 ^b
				Energ	y Density			
-	High	1.19	75.6	23.2ª	4.74 ^a	7.67	23.5	1.96
-	Low	1.14	76.5	23.7 ^b	4.85 ^b	7.74	23.8	1.86
					Probability -			
					-			
Treatment								
AA		0.27	0.27	≤ 0.01	≤ 0.01	0.75	0.70	≤ 0.01
Energy		0.25	0.12	0.03	0.03	0.41	0.25	0.11
$AA \times Energy$		0.94	0.41	0.76	0.21	0.51	0.36	0.60
SEM		0.02	0.00	0.00	0.00	0.00	0.00	0.00

Table III-8. Carcass traits of Cobb $700 \times MV$ mixed-sex broilers fed diets containing varying nutrient density to 49 d.

¹Observed means were calculated from 12 replicate values using the pen as the experimental unit. Each replicated pen contributed 4 male and 4 female carcasses. ²Treatments within AA column are high (medium +7.5% dLys), medium, and low (medium +7.5% dLys) fed to Cobb × MV from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: EAA across all treatments. Treatments within the energy column are low and high (low +110 kcal/kg). *Linear Regression Analysis.* The equations from the linear regression analyses are shown in table 9. The per replicate average dLys intake ranged from 61 g/bird to 76 g/bird from 1-49 days. When FCR was regressed to dLys intake (g/bird), a significant negative linear relationship was observed. This relationship indicates that for each additional gram of dLys intake, there was a 0.9 point reduction in FCR. Similarly, breast meat yield was regressed to dLys intake and a significant positive linear relationship was observed. The linear relationship between dLys intake and breast meat yield indicate that for each gram increase in intake, there was a 0.1% increase in yield. When regression lines were fitted to replicates on the basis of AME treatment, the similar slopes indicate that the response of FCR and breast meat yield associated with AME were independent of lysine intake as the intercept was shifted for both FCR and breast meat yield.

	Linear Regression Equation	R ² Value	P- Value
_	Breast m	eat yield	
Hen	Y = 0.118x + 15.20	0.340	$P \leq 0.01$
Len	Y = 0.140x + 13.96	0.381	$P \leq 0.01$
	FC	CR	
Hen	Y = -0.008x + 2.349	0.469	$P \le 0.01$
Len	Y = -0.009x + 2.432	0.622	$P \leq 0.01$

Table III-9. Linear regression analysis of dLys intake to breast meat yield and FCR in Cobb $700 \times MV$ mixed-sex broilers fed diets containing varying nutrient density to 49 d.

Discussion

Dietary amino acid supplementation is an established method to increase lean tissue accretion, reduce fat deposition and improve feed efficiency (Kerr et al., 1999; Kidd et al., 2004; Kidd et al., 2005; Zhai et al., 2013; Zhai et al., 2016).

In the present experiment, incremental changes in breast and fat pad yield were observed with each increase in amino acid density. Broilers fed the H*aa* diet had a 1.4% and 0.26% increase in breast fillet and tenderloin yield, respectively. Linear regression analysis indicated that for each additional gram of dLys intake there was an increase in breast meat yield by 0.12%. The similarity between regression lines independently applied to replicates from H*en* and L*en* indicate that AME did not impact breast meat yield as dLys intake increased. Several authors have reported increased breast meat yield when heavy broilers were fed diets with higher amino acid density (Kidd et al., 2004; Kidd et al., 2005; Corzo et al., 2006; Dozier III et al., 2007b; Zhai et al., 2013).

Breast muscle contains a high amount of lysine, which contributes to the proportionately greater growth response of the pectoralis major to increased lysine intake (Hamm, 1981; Kerr et al., 1999). Tesseraud et al. (1996a) compared the growth of the pectoralis major, anterior latissimus dorsi and sartorius muscles in broilers and found that the pectoralis major undergoes a higher growth rate when dietary lysine is increased. These findings correspond with the absence of treatment effects on wing or leg yield in the present study. Congruent with previous research, the present data showed a consequential reduction in fat pad yield as amino acid intake increased (Kerr et al., 1999; Kidd et al., 2004; Corzo et al., 2005; Kidd et al., 2005; Dozier III et al., 2007b; Zhai et

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al., 2013; Zhai et al., 2014). Reports have suggested that the efficiency of protein utilization has a linear relationship with energy intake (Gous et al., 1990; Gous et al., 2018). When amino acid intake is deficient and protein deposition is limited, consumption of excess dietary energy leads to increased fat accretion. In the present data, broilers that were fed L*aa* diets were subject to a suboptimal level of amino acid intake, which amplified fat deposition and led to the increase in fat pad yield.

Previous research has shown that increasing dietary amino acid density leads to improved feed efficiency and BW in heavy broilers (Kerr et al., 1999; Kidd et al., 2004; Corzo et al., 2005; Kidd et al., 2005; Corzo et al., 2006; Dozier III et al., 2007b; Zhai et al., 2013; Zhai et al., 2016). Results from the regression analysis in the present experiment show a significant linear relationship where, for each additional gram of dLys intake there was a 0.9 point reduction in FCR. Kidd et al. (2004) fed three levels of amino acid density to 49-day old broilers and found that BW was increased ($P \le 0.05$) by 607 grams and FCR was reduced ($P \le 0.05$) by 42 points in those fed high amino acid density diets. Our findings showed a similar trend to those of Kidd et al. (2004), but the magnitude of response was markedly less. For example, broilers fed the Haa versus Laa diets had a 70 g increase in BW and a 10 point reduction in FCR at d 49. Several differences in the design of the experiment conducted by Kidd et al. (2004) may explain the disparity in performance responses. In the present experiment, dietary treatments did not begin until day 12 and consisted of a 7.5% change in amino acid density. Conversely, the dietary treatments fed by Kidd et al. (2004) consisted of a 15% change in amino acid density and were fed starting at day 0. The appropriate amino acid density

of a diet is crucial for adequate performance and this is especially evident for young broilers.

The first week of post hatch development in broiler chickens provides a nutritional opportunity to enhance the physiological mechanisms that control protein deposition. The morphological changes that occur during muscle development coincide with the availability of dietary amino acids (Vieira and Angel, 2012; Powell et al., 2013). Consequently, the amino acid density of a diet fed during the starter phase provides a substantial impact on the meat yield of a broiler at slaughter (Kemp et al., 2005). This effect is especially apparent in the pectoralis major, where the amount of essential amino acids in the diet affect the rate of protein deposition (Tesseraud et al., 2001; Urdaneta-Rincon and Leeson, 2004). In the present experiment, all broilers were fed a common starter feed which contained 1.18% dLys. Formulating with this level of dLys during the starter feeding phase is a conservative approach compared to breeder recommendations, which advocate feeding 1.22% dLys from 0-10 days of age (Cobb-Vantress Inc., 2012). In future studies, the amino acid treatment should be applied during the starter phase to evaluate the effects on overall performance and processing yield of the Cobb $700 \times MV$.

Amino acids in broiler diets are supplied by feeding costly ingredients such as meat and bone meal, soybean meal and synthetic crystalline sources. Subsequently, the amino acid density of a diet contributes largely to the overall formula cost. To maximize profitability, it is important to develop feeding strategies that align with the cost structure of a producer. The premise of raising heavy broilers is to optimize the

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efficiency of meat production for the cut up and further processed market. Several economic analyses have been conducted to determine if the increased cost of feeding high amino acid density diets is justified by the improvements in meat yield (Kidd et al., 2005; Dozier III et al., 2007b; Zhai et al., 2013; Zhai et al., 2014; Zhai et al., 2016). The continual change in the price of meat and feed ingredients prevent these analyses from being universally applied, but they offer a basis to determine the economic viability of feeding additional amino acids. Similarly, the present data can be utilized by integrators producing the Cobb $700 \times MV$ to ascertain whether feeding high amino acid density diets is profitable.

Response to increased AME is dependent upon several factors, including strain, age and the presence of other limiting nutrients. In the current study, increasing AME improved feed efficiency during the grower and finisher phases and from days 1-26, 1-36, and 1-49. Dozier III et al. (2006b) found similar results after feeding heavy broilers four levels of AME ranging from 3,175 to 3,310 kcals/kg between 30 and 59 days of age. In that study, increasing dietary AME led to a linear reduction ($P \le 0.05$) in feed intake and FCR. The present research shows that feed intake was reduced in the grower phase with no impact on weight gain. Several authors have suggested that modern broilers can adjust feed intake to compensate for changes in dietary AME, which is partly responsible for the improvements in feed efficiency (Leeson et al., 1996; Hidalgo et al., 2004; Dozier III et al., 2007a). The present data also showed a reduction in breast meat and tender yields in response to increased dietary AME. Similar studies have demonstrated that the effect of AME on feed intake leads to a consequential reduction in

amino acid intake, which negatively impacts breast meat yield (Dozier III et al., 2006b; Dozier III et al., 2007a; Dozier III and Gehring, 2014). Fat pad yield was not affected by increased AME in the present study. Increased fat deposition in broilers appears to be more responsive to reduced amino acid intake, rather than increased dietary AME (Dozier III et al., 2006b; Basurco et al., 2015).

The objective of the present study was to evaluate commercial feeding strategies and determine the sensitivity of the Cobb $700 \times MV$ strain to changes in dietary amino acid density and AME intake. Strategies often involve feeding low or high nutrient density diets to either maximize meat yield or minimize cost. Subsequently, the dietary treatments fed in this experiment were formulated to simulate these two scenarios. The present data indicate that the Cobb $700 \times MV$ broiler was responsive to changes in dietary amino acid and AME density. These results coincide with an established trend in broiler nutrition, where increased amino acid and AME density leads to improvements in meat yield, BW and feed efficiency.

CHAPTER IV

INCREASING AMINO ACID DENSITY IMPROVES PERFORMANCE AND PROCESSING YIELD IN THE COBB 700 \times MV MIXED-SEX BROILERS

Introduction

Broiler diets must contain a balanced supply of amino acids to optimize performance and meat yield. This is achieved by feeding costly ingredients, such as soybean meal, meat and bone meal, and crystalline amino acids. Amino acid requirements are influenced by a variety of environmental, genetic, and physiological factors. Modern broilers incorporate dietary amino acids into lean tissue more efficiently and consume less feed per unit BW gain than broilers raised in the past (Edwards III and Baker, 1999; Havenstein et al., 2003). Consequently, strains selected for improved performance and processing yield are more sensitive to changes in amino acid intake. For example, Tesseraud et al. (1999) found that an experimental line of broilers selected for greater breast meat yield and reduced fat deposition more efficiently utilized dietary lysine for increased BW gain and pectoralis major growth when compared to the unselected control line. The genetic effects on amino acid requirements drive the need to feed broilers on strain-specific basis. A comparison between two modern broiler strains found a 5-7 % difference in the dietary lysine required to maximize weight gain (Dozier III and Payne, 2012). Changes in metabolism (Tesseraud et al., 1999), feed intake (Havenstein et al., 2003) and allometric growth patterns (Schmidt et al., 2009) in

response to genetic selection provide the basis for the deviation in amino acid requirements.

The United States poultry industry has continually increased the number of heavy broilers processed above 3.5 kg in order to meet the demand for breast fillet and value added products (Dozier III et al., 2007b). Producers must feed diets containing amino acid dense ingredients in order to maximize breast meat yield and feed efficiency in heavy broilers. Approximately 70% of the cumulative feed intake occurs between 2.0 and 3.6 kg of live weight (Dozier III et al., 2007a). Depending on the cost of ingredients and the value of breast meat, the substantial quantity of feed consumed at heavier body weights can make high amino acid density diets prohibitively expensive. An assessment of broiler performance and meat yield to dietary amino acid density found that increasing density by 10% led to a 5.6% increase in diet cost (Kidd et al., 2005). Nonetheless, an economic evaluation by that study found that the resulting increase in breast meat yield maximized profitability despite the higher diet cost. Therefore, it is essential to identify the amino acid density needed to maximize meat yield in order to determine the economic feasibility of feeding high density diets.

The dietary inclusion of amino acids has a major effect on the growth and development of various carcass components. Many studies have shown that breast meat yield is increased when broilers are fed diets with high amino acid density (Kerr et al., 1999; Kidd et al., 2004; Dozier III et al., 2007b). Furthermore, breast meat yield has been shown to be maximized at a higher digestible lysine (dLys) estimate than growth (Kerr et al., 1999). The sensitivity of the pectoralis muscles to the dLys content of a diet is likely due to the high level of lysine found in the tissue (Hamm, 1981; Tesseraud et al., 1996a). Additionally, increasing dietary amino acid density reduces fat pad yield (Kidd et al., 2004; Dozier III et al., 2007b; Zhai et al., 2013). This effect has been attributed to the metabolic changes that occur in response to increased amino acid intake, which leads to a reduction in lipogenesis (Rosebrough et al., 1999; Rosebrough et al., 2002). Heavy broilers are primarily raised for breast meat, making the effects of amino acid density on carcass yield a substantial impact on profitability.

The Cobb $700 \times MV$ broiler is a strain selected to maximize feed efficiency and breast meat yield when raised to a heavier BW (Cobb-Vantress, 2019). Few studies have been conducted on the responsiveness of the Cobb $700 \times MV$ broiler to feeding diets with increased amino acid density. Philpot (2018) and Johnson (2018) found that increasing amino acid density improved breast meat yield and feed efficiency. Conversely, Maynard et al. (2018); (2019) found that feeding diets with increased amino acid density had no impact on breast meat yield. The primary objective in raising heavy broilers is to maximize the efficiency in which dietary amino acids are deposited as meat yield. Therefore, we evaluated the effects of feeding incremental increases in amino acid density on the performance and processing yield of the Cobb $700 \times MV$ mixed sex broiler.

Materials & Methods

Experimental Animals. On the day of hatch, 1,680 Cobb 700 × MV broilers were obtained from a commercial hatchery, where they were vaccinated for Newcastle and Marek's disease. The straight run study was conducted in a tunnel ventilated broiler barn with 60 floor pens, each containing a tube feeder, nipple drinker line, and shaved pine litter. Feed and water were provided to broilers on an *ad libitum* basis. Barn temperature was recorded daily and adjusted in response to bird comfort. The lighting program followed the standard operating procedure for broilers raised at the research facility. Broilers were raised in accordance with procedures approved by the Texas A&M University Institutional Animal Care and Use Committee.

Dietary Treatments. Dietary treatments were formulated on a least cost basis and were primarily composed of corn, soybean meal, corn DDGS, and pork meat and bone meal. Composite samples of corn, soybean meal, corn DDGS, and pork meat and bone meal were analyzed by near infrared spectroscopy to determine amino acid content. Diets were manufactured at the Texas A&M Poultry Science Department research feed mill in four phases: starter (d 0-12), grower (d 12-26), finisher (d 26-36), and withdrawal (d 36-49). All diets were steam pelleted and, during the starter phase, diets were crumbled using a roller mill. Beginning in the starter phase, broilers were fed one of five dietary treatments. The control (CON), which was based off breeder nutrient recommendations for the Cobb $700 \times MV$ broiler (Cobb-Vantress, 2019), or the CON

plus 5, 10, 15, or 20% increases in digestible essential amino acid content (dEAA). Treatments were formulated to maintain equivalent dEAA to dLys ratios across all diets within a given phase. A composite feed sample was collected for each diet and submitted to a third party laboratory (Carat-Adisseo Analytical Laboratory, Antony, France) for proximate nutrient and amino acid analysis. Increasing the dEAA content of treatment diets was achieved by decreasing the inclusion of corn and increasing the inclusion animal vegetable blended fat, soybean meal, and crystalline amino acids. Meat and bone meal and corn DDGS were included at a constant level across all treatments within a given phase.

Performance Measurements. Bird weight and feed intake were measured by pen at the end of each feeding phase on days 12, 26, 36, and 49. Mortality was recorded daily and used as an adjustment factor when calculating feed conversion. Average body weight (BW), g/bird/d feed intake (FI), percent mortality, and mortality adjusted feed conversion ratio (FCR) were determined by phase and on a cumulative basis. At 49 days of age, male and female broilers were weighed separately in order to determine the average bird weight for both sexes of each pen. Average lysine intake (g/bird) was determined by multiplying average replicate FC (g/bird) by the calculated dietary dLys content for each feeding phase. The average lysine intake (g/bird) of each phase was then summed to determine the total lysine intake on an average per replicate basis (g/bird). *Processing Measurements.* At 50 days of age, 4 males and 4 females of representative weight were selected for processing from each pen. Individual live weight was determined, and birds were electrically stunned, bled, scalded, mechanically plucked, and manually eviscerated. Fat pad and hot carcass weight was measured for each bird. Carcasses were chilled in an immersion tank for 60 min at 1° C and manually agitated at regular intervals. After chilling, carcasses were deboned and the weight of legs, wings, skinless-breast fillet, and tenderloin were determined for each bird. Parts yield was determined for each bird and was calculated as the percent live weight, where the weight of the part was divided by the live weight. The severity of woody breast (WB) was determined for each breast fillet using tactile evaluation as described by Tijare et al. (2016).

Statistical Analysis. Each pen was designated as an experimental unit and 12 pens were assigned to each dietary treatment. Treatments were arranged in a randomized complete block design, with the location of the pen in the barn used as the blocking factor.

All data were analyzed by one-way ANOVA using α =0.05. Significantly different means were separated using Duncan's Multiple Range Test *post-hoc*. Feed conversion ratio and the percentage of total breast meat yield (breast fillet + tenderloin) for each replicate pen was regressed with the average per bird lysine intake using linear regression analysis where α =0.05. The slope of the line represented the improvement in either breast meat yield or feed efficiency for each gram increase in lysine intake. The

coefficient of determination (R^2) was used to establish the goodness of fit for replicate means to the regression line.

Results

Dietary Analysis. Overall, the experimental diets provided the intended nutrient variation between treatments (Tables 1, 2, 3, & 4) The average difference in analyzed lysine values between the CON and the +5%, +10%, +15%, and +20% dEAA treatments across all phases was 5.73, 9.77, 16.80, and 20.92%. Similarly, for analyzed methionine and threonine, the average differences were all within 2% of the intended difference. Increasing the dEAA content of treatment diets led to an increase in the inclusion of animal-vegetable blended fat, and this was confirmed by the crude fat analysis conducted on the treatment feed samples.

Ingredient (%)CON $\pm 5\%$ $\pm 10\%$ $\pm 15\%$ $\pm 20\%$ Corn60.1556.6053.0549.4545.85Soybean meal28.5031.5034.4537.5040.45Meat and Bone Meal4.004.004.004.004.00Corn DDGS ¹ 5.005.005.005.005.00DL-Methionine0.280.300.320.340.36L-Lysine HCL0.290.280.270.250.24L-Threonine0.080.080.080.080.08Fal²0.150.801.402.002.60Limestone0.810.790.760.730.70Phosphate³0.050.030.010.000.00Vitamins40.030.030.030.030.03Nrace Minerals ⁵ 0.080.080.080.080.08NSPase ⁶ 0.030.030.030.030.03Octacluated Nutrient Content ME 2.9824.0625.1626.24dig-Met, %0.590.620.650.680.71dig-Thr, %0.770.810.840.880.92dig-Thr, %0.770.810.840.880.92dig-Thr, %0.770.810.840.880.92dig-Thr, %0.180.180.180.180.18Avail, P, %0.460.460.460.460.47Ca, %0.92 </th <th></th> <th></th> <th></th> <th>Starter</th> <th></th> <th></th>				Starter		
Soybean meal28.5031.5034.4537.5040.45Meat and Bone Meal4.004.004.004.004.00Corn DDCS ¹ 5.005.005.005.00DL- Methionine0.280.300.320.340.36L-Lysine HCL0.290.280.270.250.24L-Threonine0.080.080.080.080.08Fat ² 0.150.801.402.002.60Limestone0.810.790.760.730.70Phosphate ³ 0.050.030.010.000.00Vitamins ⁴ 0.030.030.030.030.03NSPase ⁶ 0.030.030.030.030.03Nicarbazin ⁷ 0.050.050.050.050.05Calculated Nutrient Content T T T T ME, kcal/kg30033003300330033003Grep, %1.181.241.301.361.42dig-TSAA, %0.870.920.961.001.05dig-TsKA, %0.870.920.920.920.92Na0.1251.331.411.491.57Avail, P, %0.460.460.460.460.47Ca, %0.920.920.920.920.92Na0.180.180.180.18Analyzed Nutrient Content (%)TT5.305.60Crud	Ingredient (%)	CON	+5%	+10%	+15%	+20%
Meat and Bone Meal 4.00 4.00 4.00 4.00 4.00 Corn DDGS ¹ 5.00 5.00 5.00 5.00 DL- Methionine 0.28 0.30 0.32 0.34 0.36 L-Lysine HCL 0.29 0.28 0.27 0.25 0.24 L-Threonine 0.08 0.08 0.08 0.08 0.08 Fat ² 0.15 0.80 1.40 2.00 2.60 Limestone 0.81 0.79 0.76 0.73 0.70 Phosphate ³ 0.05 0.03 0.01 0.00 0.00 Vitamins ⁴ 0.03 0.03 0.03 0.03 0.03 NSPase ⁶ 0.03 0.03 0.03 0.03 0.03 Nicarbazin ⁷ 0.05 0.05 0.05 0.05 Calculated Nutrient Content U U 1.18 1.24 ME, kcal/kg 3003 3003 3003 3003 3003 GP, % 21.91 22.98 24.06 25.16 26.24 dig-TbA, % 0.59 0.62 0.65 0.68 0.71 dig-Lys, % 1.18 1.24 1.30 1.36 1.42 dig-TbA, % 0.87 0.92 0.92 0.92 0.92 dig-Thr, % 0.77 0.81 0.84 0.88 0.92 dig-TbA, % 0.89 0.93 0.97 1.02 1.06 dig-TbA, % 0.18 0.18 0.18 0.18 0.18 <td>Corn</td> <td>60.15</td> <td>56.60</td> <td>53.05</td> <td>49.45</td> <td>45.85</td>	Corn	60.15	56.60	53.05	49.45	45.85
Corn DDGS ¹ 5.00 5.00 5.00 5.00 5.00 DL- Methionine 0.28 0.30 0.32 0.34 0.36 L-Lysine HCL 0.29 0.28 0.27 0.25 0.24 L-Threonine 0.08 0.08 0.08 0.08 0.76 L-Threonine 0.81 0.79 0.76 0.73 0.70 Phosphate ³ 0.05 0.03 0.01 0.00 0.00 Vitamins ⁴ 0.03 0.03 0.03 0.03 0.03 Nicarbazin ⁷ 0.05 0.05 0.05 0.05 0.05 Calulated Nutrient Content Vitanis 1.00 1.00 1.05 ME, kcal/kg 3003 3003 3003 3003 3003 3003 CP, % 21.91 22.98 24.06 25.16 26.24 dig-Tkr, % 0.77 0.81 0.84 0.88 0.92 dig-Tkr, % 0.77 0.81 0.84 0.80	Soybean meal	28.50	31.50	34.45	37.50	40.45
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Meat and Bone Meal	4.00	4.00	4.00	4.00	4.00
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Corn DDGS ¹	5.00	5.00	5.00	5.00	5.00
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	DL- Methionine	0.28	0.30	0.32	0.34	0.36
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	L-Lysine HCL	0.29	0.28	0.27	0.25	0.24
Limestone 0.81 0.79 0.76 0.73 0.70 Phosphate ³ 0.05 0.03 0.01 0.00 0.00 Vitamins ⁴ 0.03 0.03 0.03 0.03 0.03 Trace Minerals ⁵ 0.08 0.08 0.08 0.08 0.08 NSPase ⁶ 0.03 0.03 0.03 0.03 0.03 Nicarbazin ⁷ 0.05 0.05 0.05 0.05 Calculated Nutrient ContentME, kcal/kg 3003 3003 3003 3003 CP, % 21.91 22.98 24.06 25.16 26.24 dig-Met, % 0.59 0.62 0.65 0.68 0.71 dig-TSAA, % 0.87 0.92 0.96 1.00 1.05 dig-Thr, % 0.77 0.81 0.84 0.88 0.92 dig-Val, % 0.89 0.93 0.97 1.02 1.06 dig-Arg, % 1.25 1.33 1.41 1.49 1.57 Avail. P, % 0.46 0.46 0.46 0.46 0.46 0.92 0.92 0.92 0.92 0.92 Na, % 0.18 0.18 0.18 0.18 0.18 0.18 0.18 0.18 0.18 0.18 0.18 0.192 0.92 0.92 0.92 0.92 0.92 Na, % 0.18 0.18 0.18 0.18 0.18 0.192 0.96 1.00 1.02 1.09	L-Threonine	0.08	0.08	0.08	0.08	0.08
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Fat ²	0.15	0.80	1.40	2.00	2.60
Vitamins ⁴ 0.03 0.03 0.03 0.03 0.03 Trace Minerals ⁵ 0.08 0.08 0.08 0.08 0.08 NSPase ⁶ 0.03 0.03 0.03 0.03 0.03 Nicarbazin ⁷ 0.05 0.05 0.05 0.05 Calculated Nutrient ContentME, kcal/kg 3003 3003 3003 3003 Colspan="4">Colspan="4"Colspan="4">Colspan="4"Colspan="4"Colspan="4"Colspan="4"Colspan="4"Colspan="4"Colspan	Limestone	0.81	0.79	0.76	0.73	0.70
Trace Minerals 0.08 0.08 0.08 0.08 0.08 NSPase 0.03 0.03 0.03 0.03 0.03 Nicarbazin 0.05 0.05 0.05 0.05 Calculated Nutrient ContentME, kcal/kg 3003 3003 3003 3003 CP, % 21.91 22.98 24.06 25.16 26.24 dig-Met, % 0.59 0.62 0.65 0.68 0.71 dig-Lys, % 1.18 1.24 1.30 1.36 1.42 dig-TSAA, % 0.87 0.92 0.96 1.00 1.05 dig-Thr, % 0.77 0.81 0.84 0.88 0.92 dig-Val, % 0.89 0.93 0.97 1.02 1.06 dig-Arg, % 1.25 1.33 1.41 1.49 1.57 Avail. P, % 0.46 0.46 0.46 0.46 0.47 Ca, % 0.92 0.92 0.92 0.92 0.92 Na, % 0.18 0.18 0.18 0.18 0.18 Crude Protein 22.90 24.10 25.40 26.40 26.80 Crude Fat 4.20 4.30 4.70 5.30 5.60 Lysine 1.38 1.49 1.56 1.71 1.72 Methionine 0.60 0.69 0.68 0.69 0.71 Threonine 0.94 0.96 1.00 1.02 1.09 Arginine 1.38 1.54	Phosphate ³	0.05	0.03	0.01	0.00	0.00
NSPase ⁶ 0.03 0.03 0.03 0.03 0.03 Nicarbazin ⁷ 0.05 0.05 0.05 0.05 0.05 Calculated Nutrient Content 0.05	Vitamins ⁴	0.03	0.03	0.03	0.03	0.03
Nicarbazin ⁷ 0.05 0.05 0.05 0.05 Calculated Nutrient Content ME, kcal/kg 3003 3003 3003 3003 3003 CP, % 21.91 22.98 24.06 25.16 26.24 dig-Met, % 0.59 0.62 0.65 0.68 0.71 dig-Lys, % 1.18 1.24 1.30 1.36 1.42 dig-TSAA, % 0.87 0.92 0.96 1.00 1.05 dig-Thr, % 0.77 0.81 0.84 0.88 0.92 dig-Val, % 0.89 0.93 0.97 1.02 1.06 dig-Arg, % 1.25 1.33 1.41 1.49 1.57 Avail. P, % 0.46 0.46 0.46 0.46 0.46 0.46 0.46 0.46 Analyzed Nutrient Content (%) 0.18 0.18 0.18 0.18 0.18 0.18 0.18 Crude Protein 22.90 24.10 25.40 26.40 26.80	Trace Minerals ⁵	0.08	0.08	0.08	0.08	0.08
$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$	NSPase ⁶	0.03	0.03	0.03	0.03	0.03
ME, kcal/kg 3003 3003 3003 3003 3003 3003 CP, % 21.91 22.98 24.06 25.16 26.24 dig-Met, % 0.59 0.62 0.65 0.68 0.71 dig-Lys, % 1.18 1.24 1.30 1.36 1.42 dig-TSAA, % 0.87 0.92 0.96 1.00 1.05 dig-Thr, % 0.77 0.81 0.84 0.88 0.92 dig-Val, % 0.89 0.93 0.97 1.02 1.06 dig-Arg, % 1.25 1.33 1.41 1.49 1.57 Avail. P, % 0.46 0.46 0.46 0.46 0.47 Ca, % 0.92 0.92 0.92 0.92 0.92 Na, % 0.18 0.18 0.18 0.18 0.18 Crude Protein 22.90 24.10 25.40 26.40 26.80 Crude Fat 4.20 4.30 4.70 5.30 5.60 Lysine 1.38 1.49 1.56 1.71 1.72 Methionine 0.60 0.69 0.68 0.69 0.71 Threonine 0.94 0.96 1.00 1.02 1.09 Arginine 1.38 1.54 1.61 1.65 1.76	Nicarbazin ⁷	0.05	0.05	0.05	0.05	0.05
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Calculated Nutrient	Content				
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	ME, kcal/kg	3003	3003	3003	3003	3003
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	CP, %	21.91	22.98	24.06	25.16	26.24
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	dig-Met, %	0.59	0.62	0.65	0.68	0.71
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	dig-Lys, %	1.18	1.24	1.30	1.36	1.42
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	dig-TSAA, %	0.87	0.92	0.96	1.00	1.05
dig-Arg, %1.251.331.411.491.57Avail. P, %0.460.460.460.460.47Ca, %0.920.920.920.920.92Na, %0.180.180.180.180.18Crude Protein22.9024.1025.4026.4026.80Crude Fat4.204.304.705.305.60Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76	dig-Thr, %	0.77	0.81	0.84	0.88	0.92
Avail. P, % 0.46 0.46 0.46 0.46 0.46 0.47 Ca, % 0.92 0.92 0.92 0.92 0.92 Na, % 0.18 0.18 0.18 0.18 0.18 Analyzed Nutrient Content (%)Crude Protein 22.90 24.10 25.40 26.40 26.80 Crude Fat 4.20 4.30 4.70 5.30 5.60 Lysine 1.38 1.49 1.56 1.71 1.72 Methionine 0.60 0.69 0.68 0.69 0.71 Threonine 0.94 0.96 1.00 1.02 1.09 Arginine 1.38 1.54 1.61 1.65 1.76	dig-Val, %	0.89	0.93	0.97	1.02	1.06
Ca, %0.920.920.920.920.92Na, %0.180.180.180.180.18Analyzed Nutrient Content (%)Crude Protein22.9024.1025.4026.4026.80Crude Fat4.204.304.705.305.60Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76		1.25	1.33	1.41	1.49	1.57
Na, % 0.18 0.18 0.18 0.18 0.18 Analyzed Nutrient Content (%) 2 3 2 2 3 2 2 3 3 2 3	Avail. P, %	0.46	0.46	0.46	0.46	0.47
Analyzed Nutrient Content (%)Crude Protein22.9024.1025.4026.4026.80Crude Fat4.204.304.705.305.60Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76						
Crude Protein22.9024.1025.4026.4026.80Crude Fat4.204.304.705.305.60Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76	Na, %	0.18	0.18	0.18	0.18	0.18
Crude Fat4.204.304.705.305.60Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76		Content (%)				
Lysine1.381.491.561.711.72Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76	Crude Protein	22.90	24.10	25.40	26.40	26.80
Methionine0.600.690.680.690.71Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76	Crude Fat	4.20	4.30	4.70	5.30	5.60
Threonine0.940.961.001.021.09Arginine1.381.541.611.651.76	Lysine		1.49	1.56	1.71	1.72
Arginine1.381.541.611.651.76						
c						
Valine 1.08 1.14 1.20 1.25 1.29	-					
	Valine	1.08	1.14	1.20	1.25	1.29

Table IV-1. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the starter.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp*.

⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

			Grower		
Ingredient (%)	CON	+5%	+10%	+15%	+20%
Corn	65.00	61.70	57.95	54.65	51.40
Soybean meal	23.80	26.60	29.45	32.20	34.90
Meat and Bone Meal	4.00	4.00	4.00	4.00	4.00
Corn DDGS ¹	5.00	5.00	5.00	5.00	5.00
DL- Methionine	0.25	0.27	0.29	0.31	0.32
L-Lysine HCL	0.29	0.28	0.26	0.25	0.24
L-Threonine	0.07	0.07	0.07	0.07	0.07
Fat ²	0.20	0.75	1.45	2.05	2.60
Limestone	0.77	0.74	0.71	0.68	0.65
Phosphate ³	0.00	0.00	0.00	0.00	0.00
Vitamins ⁴	0.02	0.02	0.02	0.02	0.02
Trace Minerals ⁵	0.06	0.06	0.06	0.06	0.06
NSPase ⁶	0.03	0.03	0.03	0.03	0.03
Nicarbazin ⁷	0.05	0.05	0.05	0.05	0.05
Calculated Nutrient (
ME, kcal/kg	3058	3058	3058	3058	3058
CP, %	20.12	21.13	22.14	23.12	24.10
dig-Met, %	0.54	0.57	0.60	0.62	0.65
dig-Lys, %	1.07	1.12	1.18	1.23	1.29
dig-TSAA, %	0.80	0.84	0.88	0.92	0.96
dig-Thr, %	0.70	0.73	0.77	0.80	0.84
dig-Val, %	0.81	0.85	0.90	0.94	0.98
dig-Arg, %	1.13	1.20	1.27	1.35	1.42
Avail. P, %	0.44	0.45	0.45	0.46	0.46
Ca, %	0.88	0.88	0.88	0.88	0.88
Na, %	0.17	0.17	0.17	0.17	0.17
Analyzed Nutrient C					
Crude Protein	20.10	20.90	21.50	23.40	23.80
Crude Fat	4.00	4.50	4.80	5.90	5.90
Lysine	1.27	1.32	1.38	1.43	1.51
Methionine	0.55	0.61	0.63	0.67	0.72
Threonine	0.81	0.81	0.87	0.96	0.95
Arginine	1.22	1.32	1.37	1.49	1.49
Valine	0.95	0.99	1.06	1.11	1.18

Table IV-2. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the grower phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp*.

⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

			Finisher		
Ingredient (%)	CON	+5%	+10%	+15%	+20%
Corn	69.05	66.80	64.30	61.30	58.30
Soybean meal	20.95	22.80	24.85	27.40	29.90
Meat and Bone Meal	3.00	3.00	3.00	3.00	3.00
Corn DDGS ¹	5.00	5.00	5.00	5.00	5.00
DL- Methionine	0.23	0.25	0.27	0.29	0.31
L-Lysine HCL	0.26	0.27	0.27	0.25	0.24
L-Threonine	0.07	0.08	0.08	0.08	0.08
Fat ²	0.20	0.60	1.00	1.50	2.05
Limestone	0.76	0.74	0.72	0.69	0.67
Phosphate ³	0.00	0.00	0.00	0.00	0.00
Vitamins ⁴	0.01	0.01	0.01	0.01	0.01
Trace Minerals ⁵	0.04	0.04	0.04	0.04	0.04
NSPase ⁶	0.03	0.03	0.03	0.03	0.03
Nicarbazin ⁷					
Calculated Nutrient	Content				
ME, kcal/kg	3102	3102	3102	3102	3102
CP, %	18.55	19.23	20.00	20.92	21.81
dig-Met, %	0.50	0.53	0.56	0.58	0.61
dig-Lys, %	0.96	1.01	1.06	1.10	1.15
dig-TSAA, %	0.75	0.79	0.83	0.86	0.90
dig-Thr, %	0.64	0.68	0.71	0.74	0.77
dig-Val, %	0.76	0.78	0.81	0.85	0.89
dig-Arg, %	1.02	1.07	1.12	1.19	1.26
Avail. P, %	0.40	0.40	0.40	0.41	0.41
Ca, %	0.79	0.79	0.79	0.79	0.79
Na, %	0.17	0.17	0.17	0.17	0.17
Analyzed Nutrient C					
Crude Protein	18.60	19.10	20.40	21.00	22.00
Crude Fat	4.20	4.30	4.50	5.00	5.70
Lysine	1.12	1.18	1.24	1.25	1.34
Methionine	0.55	0.60	0.61	0.62	0.69
Threonine	0.72	0.77	0.79	0.88	0.87
Arginine	1.13	1.15	1.19	1.30	1.38
Valine	0.91	0.92	0.95	0.98	0.99

Table IV-3 Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the finisher phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp*.

⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

	Withdrawal					
Ingredient (%)	CON	+5%	+10%	+15%	+20%	
Corn	71.85	70.50	68.35	66.20	63.80	
Soybean meal	18.15	19.90	21.65	23.45	25.45	
Meat and Bone Meal	3.00	3.00	3.00	3.00	3.00	
Corn DDGS ¹	5.00	5.00	5.00	5.00	5.00	
DL- Methionine	0.20	0.22	0.24	0.25	0.28	
L-Lysine HCL	0.25	0.26	0.26	0.27	0.27	
L-Threonine	0.06	0.06	0.07	0.08	0.08	
Fat ²	0.00	0.05	0.40	0.75	1.20	
Limestone	0.61	0.59	0.57	0.55	0.53	
Phosphate ³	0.00	0.00	0.00	0.00	0.00	
Vitamins ⁴	0.01	0.01	0.01	0.01	0.01	
Trace Minerals ⁵	0.03	0.03	0.03	0.03	0.03	
NSPase ⁶	0.03	0.03	0.03	0.03	0.03	
Nicarbazin ⁷						
Calculated Nutrient						
ME, kcal/kg	3113	3113	3113	3113	3113	
CP, %	17.46	18.17	18.82	19.50	20.22	
dig-Met, %	0.45	0.48	0.51	0.53	0.56	
dig-Lys, %	0.89	0.94	0.98	1.03	1.07	
dig-TSAA, %	0.69	0.73	0.76	0.80	0.83	
dig-Thr, %	0.60	0.63	0.66	0.69	0.72	
dig-Val, %	0.71	0.74	0.77	0.79	0.82	
dig-Arg, %	0.94	0.99	1.04	1.09	1.14	
Avail. P, %	0.39	0.40	0.40	0.41	0.40	
Ca, %	0.72	0.72	0.72	0.72	0.72	
Na, %	0.17	0.17	0.17	0.17	0.17	
Analyzed Nutrient C	Content (%)					
Crude Protein	17.40	17.80	19.30	19.90	20.70	
Crude Fat	4.10	4.20	4.30	4.50	4.90	
Lysine	1.06	1.12	1.13	1.26	1.23	
Methionine	0.52	0.50	0.53	0.58	0.59	
Threonine	0.67	0.72	0.75	0.78	0.79	
Arginine	1.03	1.11	1.14	1.20	1.18	
Valine	0.81	0.85	0.83	0.91	0.97	

Table IV-4 Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed to Cobb $700 \times MV$ mixed-sex broilers during the withdrawal phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp*.

⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-Phytase.

Performance. The effect of dEAA on live performance of Cobb 700 × MV broilers is presented in Table 5, 6, & 7. Day 0 BW averaged 0.040 kg and there were no observed treatment differences. At days 12, 26 and 36, BW was increased ($P \le 0.05$) by feeding diets containing increased dEAA. At d 12, BW was increased ($P \le 0.05$) by 2.93% in broilers fed treatments +5% dEAA and above compared to those fed the CON. Treatment separations were furthered at d 26, where broilers that were fed the +5% and +10% dEAA treatments were 3.65% and 5.70% heavier ($P \le 0.05$) than those fed the CON, respectively. By d 36, treatment effects on BW were still evident where broilers that were fed the +10% treatment were 3.17% heavier ($P \le 0.05$) than those fed the CON. Broilers fed the +15% dEAA treatment were 203 g numerically heavier than those fed the CON.

Feed conversion ratio was improved ($P \le 0.05$) by increased dEAA content during the starter, grower, and finisher phases and from days 1-26, 1-36, and 1-49. During the starter phase, broilers that were fed the +5% and +20% dEAA treatments had a 3- and 8-point improvement ($P \le 0.05$) in FCR. A stepwise reduction in FCR was observed during the grower phase, where birds that were fed the +5, +10, and +15% dEAA treatments had a 3 -, 6-, and 10-point reduction ($P \le 0.05$) in FCR compared to those fed the control, respectively. During the finisher phase, broilers fed +10% and +15% dEAA treatments had an 8- and 4-point reduction ($P \le 0.05$) in FCR compared to the CON. No significant differences in FCR were observed during the withdrawal phase. A stepwise reduction in FCR was evident from 1-26 days, where broilers that were fed the +5%, +10%, and +15% dEAA treatments compared to the CON had a 3-, 6-, and 9point improvement (P \leq 0.05), respectively. This effect continued from 1-36 days and broilers that were fed the +5% and +10% dEAA treatments had a 3- and 7-point reduction (P \leq 0.05) in FCR compared to those fed the CON. Broilers that were fed the +15% dEAA treatment had the greatest improvement in cumulative FCR that amounted to an 8-point improvement (P \leq 0.05) compared to the control. Feed intake was reduced (P \leq 0.05) during the starter and grower phase and from 1-26 days in broilers that were fed the increasing dEAA treatment. No treatment effects were observed on mortality and the average cumulative mortality was 4.48% (not shown).

	Body Weight (kg)					Body Weight Gain (kg)				
Treatment ¹	Day 0	Day 12	Day 26	Day 36	Day 49		Starter	Grower	Finisher	Withdrawal
CON	0.039	0.307 ^b	1.317°	2.369 ^b	3.285		0.268	1.010 ^c	1.052	0.909
+5%	0.040	0.316 ^a	1.365 ^b	2.417 ^{ab}	3.396		0.276	1.049 ^b	1.053	0.950
+10%	0.040	0.315 ^a	1.364 ^b	2.444 ^a	3.382		0.275	1.050 ^b	1.079	0.929
+15%	0.040	0.316 ^a	1.392 ^a	2.456 ^a	3.488		0.276	1.076 ^a	1.064	1.006
+20%	0.040	0.317 ^a	1.387 ^{ab}	2.416 ^{ab}	3.462		0.278	1.070^{ab}	1.029	1.025
	Probability									
Treatment	0.28	≤ 0.01	≤ 0.01	0.06	0.32		0.08	≤ 0.01	0.42	0.67
SEM	0.000	0.000	0.006	0.011	0.036		0.001	0.005	0.009	0.028

Table IV-5 Body weight (kg) and body weight gain (kg) of Cobb 700 × MV mixed sex broilers fed diets containing varying nutrient density to 49 d of age.

¹Treatments were control (CON.), CON +5% dEAA, CON. +10% dEAA, CON +15% dEAA, or CON +20% dEAA fed to Cobb 700 \times MV mixed sex broilers from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

	Feed Conversion Ratio ²							
Treatment ¹	Starter	Grower	Finisher	Withdrawal	1-26	1-36	1-49	
CON	1.204ª	1.404 ^a	1.733 ^a	2.694	1.361 ^a	1.524 ^a	1.813 ^a	
+5%	1.167 ^b	1.370 ^b	1.703 ^{ab}	2.685	1.327 ^b	1.491 ^b	1.783 ^{ab}	
+10%	1.156 ^b	1.336 ^c	1.647°	2.587	1.298 ^c	1.451°	1.752 ^{bc}	
+15%	1.147 ^b	1.296 ^d	1.688 ^b	2.568	1.265 ^d	1.445 ^c	1.732 ^c	
+20%	1.117°	1.302 ^d	1.694 ^{ab}	2.413	1.264 ^d	1.448 ^c	1.716 ^c	
				Probability				
Treatment	≤ 0.01	≤ 0.01	≤ 0.01	0.56	≤ 0.01	≤ 0.01	\leq 0.01	
SEM	0.005	0.007	0.008	0.061	0.006	0.005	0.010	

Table IV-6 Feed conversion ratio of Cobb $700 \times MV$ mixed sex broilers fed diets containing varying nutrient density to 49 d of age.

¹Treatments were control (CON.), CON +5% dEAA, CON. +10% dEAA, CON +15% dEAA, or CON +20% dEAA fed to Cobb 700 × MV mixed sex broilers from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

²Mortality corrected feed conversion ratio.

	Feed Intake (g/bird/d)							
Treatment ¹	Starter	Grower	Finisher	Withdrawal	1-26	1-36	1-49	
CON	28.9ª	99.9 ^{ab}	159.3	175.8	68.2 ^{ab}	94.5	116.0	
+5%	29.1ª	101.8 ^a	161.0	181.8	69.4 ^a	96.2	119.2	
+10%	28.6 ^{ab}	99.2 ^b	158.1	179.3	67.9 ^b	94.1	117.7	
+15%	28.7 ^{ab}	98.2 ^b	159.0	184.2	67.2 ^b	93.4	117.3	
+20%	28.2 ^b	98.8 ^b	157.1	182.7	67.6 ^b	94.2	118.0	
				Probability -				
Treatment	0.06	0.03	0.32	0.82	0.03	0.18	0.81	
SEM	0.100	0.400	0.700	2.100	0.200	0.400	0.800	

Table IV-7 Feed intake (g/bird/d) of Cobb $700 \times MV$ mixed sex broilers fed diets containing varying nutrient density to 49 d of age.

¹Treatments were control (CON.), CON +5% dEAA, CON. +10% dEAA, CON +15% dEAA, or CON +20% dEAA fed to Cobb 700 × MV mixed sex broilers from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

Processing Yield. The effects of dEAA treatment on 50 d carcass traits are expressed as the percentage of live weight in table 8. Skinless breast fillet yield was increased ($P \le 0.05$) by 5.75% in broilers that were fed the +20% dEAA treatment compared to the CON. Additionally, tenderloin yield was increased ($P \le 0.05$) by 4.59% in broilers that were fed the +10% dEAA treatment and above compared to the control. Lastly, fat pad yield was reduced ($P \le 0.05$) by 28.84% in broilers that were fed the +20% dEAA treatment compared to the CON. Feeding increased dEAA density did not lead to improvements in carcass, wing or leg yields in the Cobb 700 × MV mixed sex broiler. No treatment effects were observed on the severity of woody breast.

		Meat Quality						
Treatment ²	Wing	Tenderloin	Breast Fillet	Leg	Fat Pad	Woody Breast Score ³		
CON	8.15	4.57 ^b	22.6 ^c	24.4	1.56 ^a	0.87		
+5%	8.15	4.63 ^b	23.1 ^{bc}	24.2	1.46 ^a	1.02		
+10%	8.32	4.78^{a}	23.3 ^{abc}	24.2	1.21 ^b	1.08		
+15%	8.10	4.78^{a}	23.4 ^{ab}	24.1	1.23 ^b	1.08		
+20%	8.06	4.79 ^a	23.9 ^a	23.9	1.11 ^b	1.16		
	Probability							
Treatment	0.57	0.01	0.03	0.49	≤ 0.01	0.06		
SEM	0.05	0.03	0.10	0.10	0.03	0.04		

Table IV-8. Processing attributes of Cobb 700 \times MV mixed –sex broilers fed diets containing varying nutrient density to 49 d¹.

¹Observed means were calculated from 12 replicate values using the pen as the experimental unit. Each replicated pen contributed 4 male and 4 female carcasses.

 2 Treatments were control (CON.), CON +5% dEAA, CON. +10% dEAA, CON +15% dEAA, or CON +20% dEAA fed to Cobb 700 × MV mixed sex broilers from 0 to 12, 12 to 26, 26 to 36, and 36 to 49 d of age. Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

³Woody breast scoring method as described by Tijare et al. (2016).

Linear Regression Analysis. A linear regression analysis was conducted to determine the effect of increasing dLys intake on total breast meat yield and FCR (Figures 1 & 2). The per replicate 1-49 d average dLys intake ranged from 49 g/bird to 73 g/bird. When total breast meat yield was regressed to dLys intake (g/bird), a significant positive linear relationship was observed. This relationship indicates that for each additional gram of dLys intake, there was a 0.13% increase in breast meat yield. Similarly, a significant negative linear relationship was observed between FCR and dLys intake (Figure 2). The linear relationship indicates that for each additional gram increase in dLys intake there was a 1.5 point reduction in FCR.

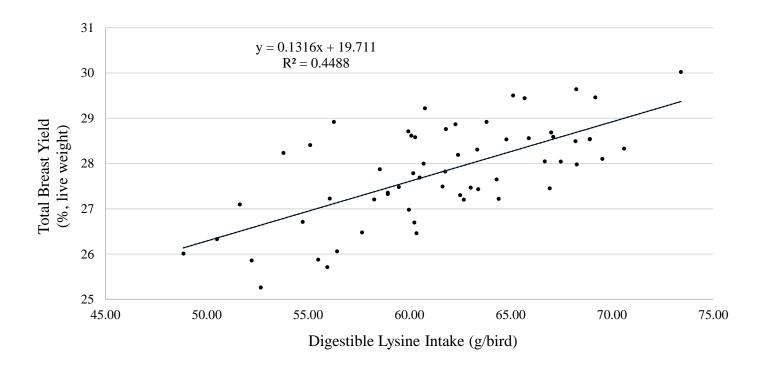


Figure IV-1 Linear regression (p \leq 0.05) of total breast meat yield to digestible lysine intake from 0-49 d in Cobb 700 \times MV straight-run broilers.

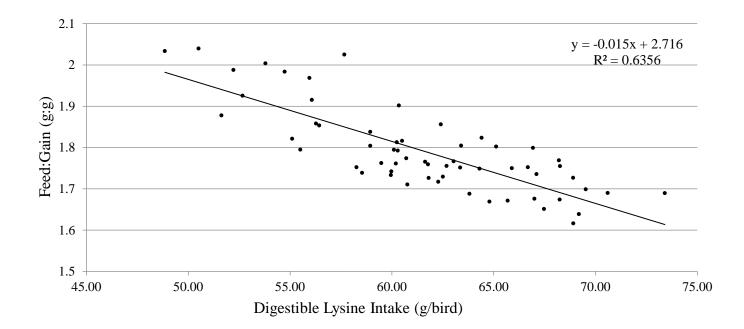


Figure IV-2 Linear regression ($p \le 0.05$) feed:gain to digestible lysine intake from 0-49 d in Cobb 700 × MV straight-run broilers.

Discussion

The goal of diet manufacturing with respect to amino acid density was to increase the dEAA content of the CON by 5, 10, 15 or 20%. The amino acid analysis conducted on the experimental feed samples confirm that the intended nutrient variation was achieved, indicating that diets were properly mixed and manufactured. The performance and processing yield responses follow a classic trend to increasing amino acid density, furthering the notion that treatment diets were adequately formulated. When the amino acid density of the treatment diets increased, there was a corresponding rise in the inclusion of soybean meal and animal-vegetable blended fat and decrease in the inclusion of corn. Proximate analysis of the treatment feed samples correspond with this assertion and show that crude fat increases with increased dEAA treatment.

Feeding diets with high amino acid density is an established method to improve broiler live performance throughout the production cycle (Kidd et al., 1998; Zhai et al., 2013; Zhai et al., 2016). For example, protein translation is upregulated in the pectoralis major and feed efficiency is improved in chicks fed diets with increased amino acid density for five days post hatch (Kidd et al., 2005; Everaert et al., 2010). Conversely, feeding suboptimal levels of amino acids to chicks can impair muscle development and negatively impact growth (Tesseraud et al., 1996b; Everaert et al., 2010; Powell et al., 2013). Kidd et al. (1998) found that feeding broilers high lysine diets from 1-18 d improved final BW and breast meat yield irrespective of the lysine level fed from 19-50 d. Data from the present experiment show that broilers fed the increased dEAA treatments had significant improvements in BW and feed efficiency at d 12. A study conducted by Willemsen et al. (2008) observed that BW at d 7 is the best predictor for BW at harvest. Additionally, reports documented an association between BW and breast meat yield and indicate that this allometric relationship has intensified with modern genetic selection (Zuidhof, 2005; Schmidt et al., 2009; Zuidhof et al., 2014). These studies, combined with the present data, advance the notion that feeding high amino acid density diets during the starter phase may affect BW and breast meat yield at harvest. Further research should be conducted with the Cobb $700 \times MV$ broiler to ascertain the impact of starter phase nutrition on harvest weight, performance, and processing yield.

Body weight was continually improved by increased dEAA treatment at d 26 and 36. No significant difference in d 49 BW was observed, although broilers that were fed the +15% dEAA treatment were 6% heavier than those fed the control. Similarly, Dozier III et al. (2006a) noted improved BW in broilers fed high amino acid density diets at 35 days, but this improvement did not continue to 46 days. In the present study, a high level of non-treatment variability was observed with final BW. Factors including the use of a mixed-sex flock and elevated environmental temperatures as rearing took place during the summer could have increased variability. Feed efficiency was the most responsive performance parameter, where stepwise improvements were observed with increasing amino acid density. On a cumulative basis, this amounted to a 6- and 8-point reduction in FCR for broilers that were fed the +10% and +15% dEAA treatments, respectively. Results from the regression analysis confirm a significant linear relationship, where for each additional gram of dLys intake there was a 1.5-point reduction in FCR. In a review of 16 data sets, the dLys estimate for optimal Several authors have found that the

optimal dLys estimate varies depending on the selection criteria in question (Vazquez and Pesti, 1997; Kerr et al., 1999). For example, a review of 16 independent research reports showed that the 0-21 d lysine (total) requirement for optimal feed efficiency and BW gain was 1.32% and 1.21%, respectively (Vazquez and Pesti, 1997). The findings in the present data do not confirmation of the aforementioned study, as final BW and cumulative FCR were both optimized at the +15% dEAA treatment.

Increasing dietary amino acid density leads to reduced fat deposition and increased lean tissue accretion (Kerr et al., 1999; Kidd et al., 2004; Kidd et al., 2005; Zhai et al., 2013; Zhai et al., 2016). In the present experiment, incremental changes in breast, tender, and fat pad yield were observed in birds fed diets with increasing dEAA. Broilers fed the +20% dEAA treatment had a 5.75% increase in breast meat yield compared to those fed the control. Additionally, tender yield was increased by 4.59% in those fed the +10% dEAA treatment and above. Linear regression analysis indicated that for each additional gram of dLys intake, breast meat yield was increased by 0.13%. Congruent with previous studies, feeding increased amino acid density diets did not lead to changes in wing or leg yield. Tesseraud et al. (1996a) found that the supply of dietary lysine had greater effect on the rate of protein synthesis and deposition in the pectoralis major compared to muscles in the legs or wings. The relative sensitivity of breast muscle to lysine intake may be due to the type of muscle fibers and the high level of lysine found in the tissue (Hamm, 1981; Tesseraud et al., 1996a). Breast meat yield is maximized at a higher lysine estimate than growth or feed efficiency (Kidd et al., 1998; Kerr et al., 1999). The present findings are in agreement and show that feed efficiency

and growth were optimized in broilers fed the +15% dEAA treatment, whereas breast meat yield was highest in those fed the +20% dEAA treatment.

Fat pad yield was also affected by amino acid density, where broilers fed the +20% dEAA had 28.9% reduction in fat pad yield compared to those fed the CON. Increasing amino acid intake has been repeatedly shown to reduce fat deposition (Kerr et al., 1999; Kidd et al., 2004; Kidd et al., 2005; Zhai et al., 2013). Dietary amino acid intake is a regulator of *de novo* lipid metabolism in chickens and can affect gene expression, hormone production and the formation of metabolic substrates (Yeh and Leveille, 1969; Rosebrough and Steele, 1985; Rosebrough et al., 1999; Rosebrough et al., 2002). For example, feeding increased amino acid density diets to 7-28 d old chickens increased IGF-1 and T₄ and decreased lipogenesis, malic enzyme activity, and T₃ (Rosebrough et al., 1999). The metabolic changes that occur in response to increased amino acid intake provide a likely explanation for the reduced fat pad yield in broilers fed the increased dEAA treatments.

As dietary amino acid density is increased, there is a corresponding reduction in the efficiency of protein utilization (e_p) (Jackson et al., 1982; Deschepper and DeGroote, 1995; Bregendahl et al., 2002; Swennen et al., 2004). The inverse relationship between amino acid intake and e_p suggests that a balance between dietary amino acid density and energy is required for optimal lean tissue deposition. This balance represents an energy dependent phase, where e_p in animals with high amino acid intake is dependent upon the availability of dietary energy (Kyriazakis and Emmans, 1992; Liu et al., 2016; Gous et al., 2018). In the present experiment, isoenergetic dietary treatments were fed with incremental increases in amino acid density and CP:AME ratios. For example, the grower phase CP:AME ratios ranged from 15.7 in the CON to 18.8 in the +20% dEAA treatment. If a balance is required between the level of energy and amino acid density, then consideration should be given to the CP:AME ratio during diet formulation to prevent a reduction in e_p. Gous et al. (2018) fed 7-21 day old broilers six dietary treatments with CP:AME ratios ranging from 13.9 to 24.6. The efficiency of protein utilization followed a linear-plateau function, where utilization was reduced in broilers fed diets with CP:AME ratios of 21.1 and above. Similarly, Liu et al. (2016) fed 7-28 day old broilers five diets with CP:AME ratios that ranged from 10.4 to 25.7. A quadratic relationship between CP:AME and N retention was observed. Broilers fed diets with a CP:AME ratio of 25.7 had a 10% reduction in N retention compared to those fed a CP:AME ratio of 16.9. Considering the aforementioned studies and the improvements in meat yield and performance in the present experiment, it appears that the Cobb $700 \times MV$ can efficiently utilize amino acids supplied in a diet with a CP:AME ratio of 18.8. Previous reports have suggested that the selection for improved meat yield, efficiency, and growth rate, leads to a proportionately greater increase in the demand for amino acids compared to energy (Morris and Njuru, 1990; Kidd et al., 1998; Gous, 2010). Therefore, continual research should be conducted on newly developed broiler strains to determine the dietary amino acid density needed to optimize performance and processing yield while maintaining maximum e_p .

The objective of the present study was to evaluate commercial feeding strategies and determine the responsiveness of the Cobb $700 \times MV$ broiler to increases in dietary amino acid density. The results herein coincide with an established trend in broiler nutrition, where incremental improvements in FCR and breast meat yield were observed with stepwise increases in density. Amounting to a linear relationship between dLys intake, where each additional gram of intake improved FCR and total breast meat yield by 1.5 points and 0.13%, respectively. Ingredients high in amino acids contribute largely to the overall cost of a broiler diet. Therefore, it is important for a nutritionist to consider the responsiveness of an individual strain when making diet formulation decisions. The data from this study can be used by producers as a basis for determining the aptness of feeding increased amino acid density diets to the Cobb $700 \times MV$ broiler.

CHAPTER V

FEEDING INCREASED AMINO ACID DENSITY AT VARIOUS AGES IMPROVES PERFORMANCE AND PROCESSING YIELD IN COBB 700 × MV MIXED-SEX BROILERS

Introduction

The average live weight of broilers processed in the United States has increased by 29% over the last 20 years (USDA, 2019). This trend has coincided with the demand for breast fillets and value added products, leading to an increase in the number of heavy broilers marketed above 3.5 kg. The high value of breast meat make maximizing yield the primary goal in raising heavy broilers. To achieve this goal, it is essential to feed diets containing adequate levels of digestible essential amino acids (dEAA). The amino acid density of a feed contributes largely to the overall cost of production. Depending on ingredient prices, increasing amino acid density by 10% can lead to a 5.6% increase in diet cost (Kidd et al., 2005). To sustain the economic viability of a feeding program, it is paramount for a producer to continually appraise the performance and meat yield responses of feeding higher levels of dietary dEAA.

Several assessments have been conducted to evaluate the effects of feeding increasing amino acid density to broilers at various ages (Kidd et al., 1998; Kidd et al., 2004; Corzo et al., 2005; Kidd et al., 2005; Dozier III et al., 2006a; Dozier III et al., 2007b; Coneglian et al., 2010; Corzo et al., 2010; Taschetto et al., 2012; Vieira et al., 2012; Zhai et al., 2013). These studies aim to capitalize on the prevailing believe that the starter phase is the optimal period for nutritional intervention. The intake of dEAA during this period influences the physiological mechanisms that control muscle development. For example, feeding increased amino acid density diets for 5 d post-hatch improves the rate of protein synthesis in the pectoralis major (Everaert et al., 2010). The physiological changes that occur in response to dEAA intake during the starter phase translate to improved performance and meat yield at harvest (Kidd et al., 1998). Furthermore, feed consumption from hatch to 14 d of age accounts for only 6% of the cumulative consumption in a 55 d old. Thus, increasing amino acid density during the starter phase imparts a low monetary cost while providing an opportunity to improve breast meat production.

The Cobb 700 × MV broiler is a strain selected to maximize feed efficiency and breast meat yield when raised to a heavier body weight (Cobb-Vantress, 2019). As broilers are selected for improved yield and weight gain, there is a consequential increase in the level of dietary amino acids needed to sustain growth (Gous, 2010). Furthermore, variation in selection criteria between strains has introduced the need to formulate on a strain specific basis. The limited number of studies which have investigated the responses of the Cobb 700 × MV broiler to feeding increased amino acid density have produced conflicting results. Maynard et al. (2018) found that feeding increased density to Cobb 700 × MV broilers improved feed conversion, but did not affect final body weight or breast meat yield. Conversely, abstracts presented by Philpot (2018) and Johnson (2018) showed that increasing density improved breast meat yield and feed efficiency at harvest. Further research is warranted to determine the level of dEAA needed to optimize meat yield and performance in the Cobb $700 \times MV$.

Therefore, we conducted two experiments to define the responses of feeding increased amino acid density at various ages on the performance and processing yield of mixed sex Cobb $700 \times MV$ broilers.

Materials & Methods

Two identically designed experiments were conducted to evaluate the response of Cobb $700 \times MV$ mixed sex broilers to feeding diets with increasing amino acid density at varying ages. Experiment 1 (EXP 1) was conducted from January to February 2019 and, in the same experimental facility, experiment 2 (EXP 2) was conducted from May to July 2019. The following section describes the experimental procedure for EXP 1 and EXP 2.

Experimental Animals & Measurements. Mixed sex Cobb $700 \times MV$ chicks (n=2,380) were obtained from a commercial hatchery on the day of hatch, where they were vaccinated for Newcastle and Marek's disease. Chicks were feather sexed prior to randomization, weighing, and placement into the experiment. Each pen contained 17 male and 17 female broilers. The barn temperature and lighting program followed the standard operating procedures for broilers raised at the research facility. All experimental procedures were performed in compliance with protocols approved by the Texas A&M University Institutional Animal Care and Use Committee.

Broilers and feed were weighed at d 12, 26, 36, and 49 in order to determine growth and feed disappearance. Average body weight (BW), g/bird/d feed intake (FI), mortality adjusted feed conversion ratio (FCR), and BW gain were determined by period and cumulatively for each pen. Average digestible lysine (dLys) intake (g/bird) was calculated by multiplying the replicate phase FC (g/bird) by the dLys content of each feeding phase. Mortality was recorded daily and the weight of the deceased bird was used as an adjustment factor for FCR. At 49 days of age, male and female broilers were weighed separately in order to determine the average bird weight for both sexes of each pen.

At 50 days of age, 3 males and 3 females of representative weight were selected for processing from each pen. Individual live weight was determined, and birds were electrically stunned, bled, scalded, mechanically plucked and manually eviscerated. Fat pad and hot carcass weight was measured for each bird. Hot carcasses were deboned and the weight of legs, wings, skinless-boneless breast fillet, and tenderloin were determined. Parts yield was calculated as the percentage of live weight, where the weight of the part was divided by the live weight. The severity of woody breast (WB) was determined for each breast fillet using tactile evaluation as described by Tijare, et al. [20].

Experimental Design and Diets. Dietary treatments were formulated on a leastcost basis using corn, soybean meal, corn DDGS, and pork meat and bone meal. Composite samples of corn, soybean meal, corn DDGS and pork meat and bone meal were analyzed by near infrared spectroscopy to determine amino acid content. The total amino acid content of each ingredient was then multiplied by a digestibility coefficient to determine the digestible amino acid content of each ingredient. The digestible amino acid value for each ingredient was then used to formulate all dietary treatments. Diets were manufactured at the Texas A&M Poultry Science Department research feed mill using a double-ribbon horizontal mixer and a steam pellet mill. During the starter phase, all dietary treatments were pelleted and then crumbled using a roller mill. In all feeding phases, triplicate feed samples were collected from each dietary treatment. A composite sample for each treatment was then submitted to a third party laboratory [21] for proximate nutrient and amino acid analysis.

Ten pens were assigned to each dietary treatment. Dietary treatments were manufactured in four phases: starter (d 0-12), grower (d 12-26), finisher (d 26-36) and withdrawal (d 36-49). Broilers were fed either a low (L), medium (M), or high (H) dEAA density during the starter phase and either a L, M, H or High+ (H+) dEAA density during the grower, finisher, and withdrawal phases. In combination, a total of seven treatments were fed which consisted of LLLL, MLLL, MMMM, MHHH HMMM, HHHH and HH+H+H+. Treatments were formulated to maintain equivalent dEAA to dLys ratios across all diets within a given phase. Therefore, for each increase in dLys there was an equivalent increase in all other dEAA. Treatment nutrient differences were achieved by changing the inclusion of corn, soybean meal, animal-vegetable blended fat, and crystalline amino acids. Meat and bone meal and corn DDGS were included at a constant level across all treatments within a given phase. Diets were conditioned before pelleting and pellets were 4.4 mm in diameter.

Statistical Analysis. Data from EXP 1 and EXP 2 were analyzed independently using one-way ANOVA where α =0.05 and replicate pens were defined as the experimental unit. Significantly different means were separated post-hoc using Duncan's Multiple Range Test. Data from EXP 1 and EXP 2 were combined for regression analysis. The average FCR and percentage of breast fillet yield for each replicate pen

was regressed with the average per bird lysine intake using linear and quadratic regression analysis where α =0.05. The slope of the line represented the improvement in either breast meat yield or feed efficiency for each gram increase in lysine intake. The coefficient of determination (R2) was used to establish the goodness of fit for replicate means to the regression line.

Results

Dietary Analyses. The proximate analyses (Tables 1, 2, 3, 4, 5, 6, 7, & 8) show that the experimental diets generally provided the desired nutrient variation between treatments. Stepwise increases in the level of crude protein coincided with increasing calculated dEAA density in EXP 1 and EXP 2. This was evident in all treatments except in the L starter phase treatment of EXP 2, where crude protein was in excess of the calculated inclusion level. Additionally, crude protein was below the calculated inclusion level in the H+ treatment of the grower phase in EXP 2.

		Starter	
Ingredient (%)	Low	Medium	High
Corn	57.45	53.52	46.14
Soybean meal	29.38	32.62	38.68
Meat and Bone Meal	4.00	4.00	4.00
Corn DDGS ¹	5.00	5.00	5.00
DL- Methionine	0.34	0.38	0.46
L-Lysine HCL	0.24	0.24	0.25
L-Threonine	0.11	0.12	0.14
Fat ²	1.96	2.66	3.97
Limestone	0.70	0.68	0.64
Phosphate ³	0.13	0.11	0.07
Salt	0.36	0.36	0.36
Vitamins ⁴	0.03	0.03	0.03
Trace Minerals ⁵	0.08	0.08	0.08
Choline	0.09	0.08	0.05
Copper Sulfate	0.05	0.05	0.05
NSPase ⁶	0.03	0.03	0.03
Zoalene ⁷	0.05	0.05	0.05
Calculated Nutrient Co			
ME, kcal/kg	3029	3029	3029
CP, %	22.33	23.52	25.77
dig-Met, %	0.66	0.71	0.81
dig-Lys, %	1.22	1.30	1.45
dig-TSAA, %	0.95	1.01	1.13
dig-Thr, %	0.79	0.85	0.94
dig-Val, %	0.95	1.00	1.10
dig-Arg, %	1.28	1.37	1.52
Avail. P, %	0.46	0.46	0.46
Ca, %	0.97	0.97	0.97
Na, %	0.18	0.18	0.18
Analyzed Nutrient Con	tent (%)		
Crude Protein	22.45	24.50	27.00
Crude Fat	3.83	4.88	6.17
Lysine	1.36	1.49	1.58
Methionine	0.56	0.64	0.69
Threonine	0.89	0.94	0.99
Arginine	1.31	1.45	1.59
Valine	1.08	1.19	1.18

Table V-1. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 1 to Cobb 700 × MV mixed-sex broilers during the starter phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg

⁶The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.* ⁷Zoalene 25% was used for prevention of coccidiosis.
 ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

Phytase.

	Grower						
Ingredient (%)	Low	Medium	High	High +			
Corn	61.96	58.52	55.57	52.14			
Soybean meal	24.69	27.52	29.95	32.78			
Meat and Bone Meal	4.00	4.00	4.00	4.00			
Corn DDGS ¹	5.00	5.00	5.00	5.00			
DL- Methionine	0.25	0.29	0.31	0.35			
L-Lysine HCL	0.24	0.24	0.24	0.24			
L-Threonine	0.09	0.10	0.11	0.12			
Fat ²	2.50	3.11	3.63	4.24			
Limestone	0.57	0.55	0.54	0.52			
Phosphate ³	0.06	0.04	0.02	0.00			
Salt	0.37	0.37	0.37	0.37			
Vitamins ⁴	0.02	0.02	0.02	0.02			
Trace Minerals ⁵	0.06	0.06	0.06	0.06			
Choline	0.08	0.06	0.05	0.04			
Copper Sulfate	0.05	0.05	0.05	0.05			
NSPase ⁶	0.03	0.03	0.03	0.03			
Zoalene ⁷	0.05	0.05	0.05	0.05			
Calculated Nutrient	Content						
ME, kcal/kg	3117	3117	3117	3117			
CP, %	20.48	21.52	22.42	23.46			
dig-Met, %	0.55	0.59	0.63	0.67			
dig-Lys, %	1.10	1.17	1.23	1.30			
dig-TSAA, %	0.83	0.88	0.92	0.98			
dig-Thr, %	0.72	0.76	0.80	0.85			
dig-Val, %	0.87	0.92	0.96	1.00			
dig-Arg, %	1.16	1.23	1.29	1.37			
Avail. P, %	0.44	0.44	0.44	0.44			
Ca, %	0.88	0.88	0.88	0.88			
Na, %	0.18	0.18	0.18	0.18			
Analyzed Nutrient C							
Crude Protein	20.53	21.83	22.54	23.65			
Crude Fat	4.80	5.46	5.67	6.54			
Lysine	1.18	1.34	1.35	1.40			
Methionine	0.51	0.49	0.61	0.65			
Threonine	0.76	0.81	0.89	1.03			
Arginine	1.13	1.23	1.28	1.40			
Valine	0.95	1.01 blandad fat ³ PioEcc 16% col	1.06	1.09			

Table V-2. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 1 to Cobb 700 × MV mixed-sex broilers during the grower phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B12, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.*⁻⁷Zoalene 25% was used for prevention of coccidiosis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

Phytase.

	Finisher						
Ingredient (%)	Low	Medium	High	High +			
Corn	69.05	65.12	61.67	58.22			
Soybean meal	18.66	21.90	24.73	27.56			
Meat and Bone Meal	4.00	4.00	4.00	4.00			
Corn DDGS ¹	5.00	5.00	5.00	5.00			
DL- Methionine	0.19	0.23	0.27	0.30			
L-Lysine HCL	0.23	0.23	0.24	0.24			
L-Threonine	0.08	0.10	0.11	0.12			
Fat ²	1.56	2.26	2.87	3.49			
Limestone	0.71	0.69	0.67	0.66			
Phosphate ³	0.06	0.04	0.02	0.00			
Salt	0.34	0.34	0.34	0.34			
Vitamins ⁴	0.01	0.01	0.01	0.01			
Trace Minerals ⁵	0.04	0.04	0.04	0.04			
Choline	0.04	0.02	0.01	0.00			
Copper Sulfate	0.00	0.00	0.00	0.00			
NSPase ⁶	0.03	0.03	0.03	0.03			
Zoalene ⁷	0.00	0.00	0.00	0.00			
Calculated Nutrient	Content						
ME, kcal/kg	3139	3139	3139	3139			
СР, %	18.25	19.45	20.49	21.54			
dig-Met, %	0.47	0.52	0.56	0.61			
dig-Lys, %	0.95	1.03	1.10	1.17			
dig-TSAA, %	0.73	0.79	0.84	0.89			
dig-Thr, %	0.64	0.69	0.74	0.78			
dig-Val, %	0.77	0.82	0.87	0.92			
dig-Arg, %	1.00	1.08	1.16	1.23			
Avail. P, %	0.43	0.43	0.43	0.43			
Ca, %	0.91	0.91	0.91	0.91			
Na, %	0.17	0.17	0.17	0.17			
Analyzed Nutrient C							
Crude Protein	19.90	19.70	20.96	21.84			
Crude Fat	4.37	4.83	5.60	6.15			
Lysine	1.13	1.12	1.13	1.31			
Methionine	0.51	0.47	0.50	0.62			
Threonine	0.72	0.75	0.92	0.97			
Arginine	1.09	1.16	1.29	1.39			
Valine	0.89	0.59	1.10	1.07			

Table V-3. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 1 to Cobb 700 × MV mixed-sex broilers during the finisher phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B12, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.*⁷Zoalene 25% was used for prevention of coccide, si. Ing of total selenium, 0.013 g of *bacillus spp.*⁷Zoalene 25% was used for prevention of coccide, sis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

	Withdrawal						
Ingredient (%)	Low	Medium	High	High +			
Corn	71.45	68.97	66.50	64.02			
Soybean meal	16.65	18.67	20.70	22.72			
Meat and Bone Meal	4.00	4.00	4.00	4.00			
Corn DDGS ¹	5.00	5.00	5.00	5.00			
DL- Methionine	0.17	0.19	0.22	0.24			
L-Lysine HCL	0.23	0.23	0.23	0.23			
L-Threonine	0.07	0.08	0.09	0.10			
Fat ²	1.28	1.72	2.17	2.61			
Limestone	0.71	0.70	0.68	0.67			
Phosphate ³	0.04	0.03	0.02	0.00			
Salt	0.34	0.34	0.34	0.34			
Vitamins ⁴	0.01	0.01	0.01	0.01			
Trace Minerals ⁵	0.03	0.03	0.03	0.03			
Choline	0.00	0.00	0.00	0.00			
Copper Sulfate	0.00	0.00	0.00	0.00			
NSPase ⁶	0.03	0.03	0.03	0.03			
Zoalene ⁷	0.00	0.00	0.00	0.00			
Calculated Nutrient	Content						
ME, kcal/kg	3150	3150	3150	3150			
СР, %	17.50	18.24	18.99	19.74			
dig-Met, %	0.43	0.46	0.50	0.53			
dig-Lys, %	0.90	0.95	1.00	1.05			
dig-TSAA, %	0.69	0.72	0.76	0.80			
dig-Thr, %	0.60	0.64	0.67	0.70			
dig-Val, %	0.74	0.77	0.80	0.84			
dig-Arg, %	0.95	1.00	1.05	1.10			
Avail. P, %	0.43	0.43	0.43	0.43			
Ca, %	0.90	0.90	0.90	0.90			
Na, %	0.17	0.17	0.17	0.17			
Analyzed Nutrient C							
Crude Protein	17.57	18.60	19.79	20.18			
Crude Fat	4.46	4.73	5.01	5.36			
Lysine	1.10	1.11	1.20	1.24			
Methionine	0.47	0.43	0.51	0.50			
Threonine	0.69	0.77	0.81	0.86			
Arginine	1.00	1.10	1.15	1.20			
Valine	0.83	0.90	0.95	0.99			

Table V-4. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 1 to Cobb $700 \times MV$ mixed-sex broilers during the withdrawal phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B12, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.*⁻⁷Zoalene 25% was used for prevention of coccidiosis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

		Starter	
Ingredient (%)	Low	Medium	High
Corn	56.46	52.31	43.85
Soybean meal	29.95	33.37	40.39
Meat and Bone Meal	4.00	4.00	4.00
Corn DDGS ¹	5.00	5.00	5.00
DL- Methionine	0.35	0.38	0.45
L-Lysine HCL	0.23	0.23	0.21
L-Threonine	0.10	0.11	0.12
Fat ²	2.40	3.15	4.65
Limestone	0.70	0.68	0.63
Phosphate ³	0.13	0.11	0.06
Salt	0.36	0.36	0.36
Vitamins ⁴	0.03	0.03	0.03
Trace Minerals ⁵	0.08	0.08	0.08
Choline	0.09	0.08	0.05
Copper Sulfate	0.05	0.05	0.05
NSPase ⁶	0.03	0.03	0.03
Zoalene ⁷	0.05	0.05	0.05
Calculated Nutrient Co	ntent		
ME, kcal/kg	3028	3028	3028
CP, %	21.82	23.07	25.61
dig-Met, %	0.66	0.71	0.80
dig-Lys, %	1.22	1.3	1.45
dig-TSAA, %	0.95	1.01	1.13
dig-Thr, %	0.79	0.85	0.94
dig-Val, %	0.93	0.99	1.10
dig-Arg, %	1.28	1.37	1.55
Avail. P, %	0.46	0.46	0.46
Ca, %	0.97	0.97	0.97
Na, %	0.18	0.18	0.18
Analyzed Nutrient Con	tent (%)		
Crude Protein	19.67	25.96	24.11
Crude Fat	4.75	6.77	5.95
Lysine	1.10	1.69	1.55
Methionine	0.58	0.77	0.67
Threonine	0.76	1.06	0.98
Arginine	1.12	1.60	1.48
Valine	0.83	1.14	1.10

Table V-5. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 2 to Cobb 700 × MV mixed-sex broilers during the starter phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg

⁶The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.* ⁷Zoalene 25% was used for prevention of coccidiosis.
 ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

	Grower						
Ingredient (%)	Low	Medium	High	High +			
Corn	60.96	57.38	53.96	49.95			
Soybean meal	25.28	28.23	31.08	34.40			
Meat and Bone Meal	4.00	4.00	4.00	4.00			
Corn DDGS ¹	5.00	5.00	5.00	5.00			
DL- Methionine	0.26	0.29	0.31	0.34			
L-Lysine HCL	0.23	0.23	0.22	0.20			
L-Threonine	0.08	0.09	0.10	0.10			
Fat ²	2.93	3.57	4.17	4.89			
Limestone	0.56	0.55	0.53	0.51			
Phosphate ³	0.06	0.04	0.02	0.00			
Salt	0.37	0.37	0.37	0.37			
Vitamins ⁴	0.02	0.02	0.02	0.02			
Trace Minerals ⁵	0.06	0.06	0.06	0.06			
Choline	0.07	0.06	0.05	0.03			
Copper Sulfate	0.05	0.05	0.05	0.05			
NSPase ⁶	0.03	0.03	0.03	0.03			
Zoalene ⁷	0.05	0.05	0.05	0.05			
Calculated Nutrient	Content						
ME, kcal/kg	3117	3117	3117	3117			
CP, %	19.99	21.07	22.10	23.30			
dig-Met, %	0.55	0.59	0.63	0.67			
dig-Lys, %	1.1	1.17	1.23	1.3			
dig-TSAA, %	0.83	0.88	0.92	0.98			
dig-Thr, %	0.72	0.76	0.80	0.85			
dig-Val, %	0.85	0.90	0.95	1.00			
dig-Arg, %	1.16	1.23	1.31	1.39			
Avail. P, %	0.44	0.44	0.44	0.44			
Ca, %	0.88	0.88	0.88	0.88			
Na, %	0.18	0.18	0.18	0.18			
Analyzed Nutrient C	Content (%)						
Crude Protein	21.12	21.99	22.48	22.24			
Crude Fat	5.69	6.27	5.84	7.12			
Lysine	1.24	1.20	1.27	1.37			
Methionine	0.57	0.51	0.58	0.61			
Threonine	0.82	0.84	0.85	0.89			
Arginine	1.22	1.24	1.33	1.33			
Valine	0.91	0.95	0.96	0.98			

Table V-6. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 2 to Cobb $700 \times MV$ mixed-sex broilers during the grower phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B12, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.*⁻⁷Zoalene 25% was used for prevention of coccidiosis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

	Finisher								
Ingredient (%)	Low	Medium	High	High +					
Corn	68.03	64.11	60.33	56.25					
Soybean meal	19.29	22.50	25.63	29.00					
Meat and Bone Meal	4.00	4.00	4.00	4.00					
Corn DDGS ¹	5.00	5.00	5.00	5.00					
DL- Methionine	0.20	0.24	0.27	0.30					
L-Lysine HCL	0.22	0.23	0.22	0.20					
L-Threonine	0.08	0.10	0.10	0.11					
Fat ²	1.97	2.68	3.35	4.08					
Limestone	0.70	0.69	0.67	0.65					
Phosphate ³	0.06	0.04	0.02	0.00					
Salt	0.34	0.34	0.34	0.34					
Vitamins ⁴	0.01	0.01	0.01	0.01					
Trace Minerals ⁵	0.04	0.04	0.04	0.04					
Choline	0.03	0.02	0.01	0.00					
NSPase ⁶	0.03	0.03	0.03	0.03					
Calculated Nutrient	Content								
ME, kcal/kg	3139	3139	3139	3139					
CP, %	17.79	18.97	20.11	21.32					
dig-Met, %	0.47	0.52	0.56	0.60					
dig-Lys, %	0.95	1.03	1.10	1.17					
dig-TSAA, %	0.73	0.79	0.84	0.89					
dig-Thr, %	0.64	0.69	0.74	0.78					
dig-Val, %	0.76	0.81	0.86	0.91					
dig-Arg, %	1.00	1.08	1.16	1.25					
Avail. P, %	0.43	0.43	0.43	0.43					
Ca, %	0.91	0.91	0.91	0.91					
Na, %	0.17	0.17	0.17	0.17					
Analyzed Nutrient C									
Crude Protein	18.46	19.34	20.52	21.59					
Crude Fat	5.58	5.55	6.69	6.42					
Lysine	1.12	1.09	1.33	1.28					
Methionine	0.49	0.55	0.59	0.58					
Threonine	0.74	0.74	0.84	0.87					
Arginine	1.13	1.09	1.28	1.37					
Valine	0.84	0.79	0.91	0.99					

Table V-7. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 2 to Cobb 700 × MV mixed-sex broilers during the finisher phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp*. ⁷Zoalene 25% was used for prevention of coccidiosis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

	Withdrawal							
Ingredient (%)	Low	Medium	High	High +				
Corn	70.42	67.95	65.33	62.37				
Soybean meal	17.28	19.30	21.45	23.88				
Meat and Bone Meal	4.00	4.00	4.00	4.00				
Corn DDGS ¹	5.00	5.00	5.00	5.00				
DL- Methionine	0.17	0.20	0.22	0.24				
L-Lysine HCL	0.22	0.22	0.22	0.21				
L-Threonine	0.07	0.08	0.09	0.09				
Fat ²	1.68	2.13	2.61	3.14				
Limestone	0.70	0.69	0.68	0.67				
Phosphate ³	0.04	0.03	0.01	0.00				
Salt	0.34	0.34	0.34	0.34				
Vitamins ⁴	0.01	0.01	0.01	0.01				
Trace Minerals ⁵	0.03	0.03	0.03	0.03				
Choline	0.00	0.00	0.00	0.00				
NSPase ⁶	0.03	0.03	0.03	0.03				
Calculated Nutrient	Content							
ME, kcal/kg	3150	3150	3150	3150				
CP, %	17.05	17.79	18.57	19.45				
dig-Met, %	0.43	0.47	0.50	0.53				
dig-Lys, %	0.90	0.95	1.00	1.05				
dig-TSAA, %	0.69	0.72	0.76	0.80				
dig-Thr, %	0.60	0.64	0.67	0.70				
dig-Val, %	0.72	0.76	0.79	0.83				
dig-Arg, %	0.95	1.00	1.05	1.12				
Avail. P, %	0.43	0.43	0.43	0.43				
Ca, %	0.90	0.90	0.90	0.90				
Na, %	0.17	0.17	0.17	0.17				
Analyzed Nutrient C	Content (%)							
Crude Protein	17.42	18.79	19.48	19.68				
Crude Fat	5.41	6.44	6.25	6.07				
Lysine	1.00	1.05	1.14	1.17				
Methionine	0.51	0.52	0.56	0.53				
Threonine	0.75	0.68	0.80	0.81				
Arginine	1.00	1.17	1.22	1.18				
Valine	0.76	0.83	0.89	0.88				

Table V-8. Dietary formulations, calculated nutrient content and analyzed nutrient content of diets fed in experiment 2 to Cobb $700 \times MV$ mixed-sex broilers during the withdrawal phase.

⁴The vitamin premix supplied per kilogram of diet: 7700 IU vitamin A, 5500 ICU vitamin D₃, 55 IU vitamin E, 1.5 mg vitamin K-3, 0.01 mg B₁₂, 6.6 mg riboflavin, 38.5 mg niacin, 9.9 mg d-pantothenic acid, 0.88 mg folic acid, 2.75 mg pyridoxine, 1.54 mg thiamine, 0.08 mg biotin. ⁵The mineral premix supplied per kilogram of diet: 180 mg of manganese, 108 mg of total zinc, 5.1 mg of copper, 3.51 mg of iodine, 0.3 mg of total selenium, 0.013 g of *bacillus spp.* ⁷Zoalene 25% was used for prevention of coccidiosis. ⁶The NSPase supplied per kilogram of diet: 88.3 U Alpha-amylase, 1765 U Subtilisin, 882.6 U Endo-1, 4,-Beta-Xylanase, and 1198 FTU 6-

Phytase.

Experiment 1 Performance. Stepwise changes in FCR, BW, BW gain, and FI were observed with increasing dEAA density throughout EXP 1 (Tables 9, 10, & 11). At d 12, broilers that were fed the M and H diets had a 3.8- and 9.5-point improvement ($p \le 0.05$) in FCR, respectively, compared to those fed the L. This trend continued during the grower phase, where feeding the MM and HH+ diets led to a 5.3- and 10.7-point improvement ($p \le 0.05$) in FCR, respectively, compared to those fed the ML diets. No differences were observed in finisher and withdrawal phase FCR. From 1-26 d, feeding the MM, MH, HM, or, HH and feeding HH+ reduced ($p \le 0.05$) FCR by 4.9- and 9.2-points, respectively, compared to those fed the LL or ML treatments. A similar response was observed from 1-36 d, amounting to 3.7- and 7.4-point improvement ($p \le 0.05$) in FCR in broilers fed the LLL and MLL diets. Cumulative FCR was reduced ($p \le 0.05$) by 4.3-, and 7.7-points in broilers fed MMMM and HH+H+H diets, respectively, compared to those fed the MLLL treatment.

Increasing BW coincided with dEAA density at d 12, 26, and 36, but no differences were observed at d 49. Broilers that were fed the M and H diets were 13.64 and 16.78% heavier ($p \le 0.05$) at d 12, respectively, compared to those fed the L diets. Feeding either the M or H diets during the starter phase increased ($p \le 0.05$) d 26 BW irrespective of dEAA density fed from d 12-26. This effect continued at d 36, where broilers that were fed the M or H diets during the starter phase were heavier ($p \le 0.05$) than those fed the L diet irrespective of dEAA density fed from d 12-36. During the starter phase, BW gain was increased ($p \le 0.05$) by 13.64 and 16.78% in broilers fed the M and H diets compared to the L. Significant dietary effects on BW gain were evident during the grower and finisher phases, but these effects did not follow a consistent trend. Withdrawal phase BW gain was highest ($p \le 0.05$) in broilers fed the LLLL treatment, which may be due to the effects of compensatory gain.

Increasing (≤ 0.05) FI during the starter and grower phases and from d 1-26 and 1-36 tended to coincide with increasing BW. Significant dietary effects on FI were also observed during the grower phase and from d 1-49, but these effects did not follow a consistent trend. During the finisher phase, FI was reduced (p ≤ 0.05) as dietary amino acid density increased. Significant increases in dLys intake coincided with increasing dEAA density.

	<u> </u>	Boc	ly Weight	(kg)		Body Weight Gain (kg)			
Treatment ¹	Day 0	Day 12	Day 26	Day 36	Day 49	Starter	Grower	Finisher	Withdrawal
LLLL	40.1 ^b	0.286 ^c	1.227 ^d	2.187 ^b	3.587	0.246 ^c	0.941°	0.960 ^c	1.400^{a}
MLLL			1.335 ^{ab}	2.298 ^a	3.548		1.008 ^{ab}	0.963°	1.273 ^b
MMMM	40.4 ^a	0.325 ^b	1.344 ^{ab}	2.334 ^a	3.606	0.285 ^b	1.003 ^a	0.990 ^{abc}	1.272 ^b
MHHH			1.278 ^c	2.303 ^a	3.560		0.954 ^c	1.025 ^a	1.258 ^b
HMMM			1.360 ^a	2.336 ^a	3.573		1.025 ^a	0.976 ^{bc}	1.238 ^b
НННН	40.1 ^b	0.334 ^a	1.322 ^b	2.353 ^a	3.660	0.294 ^a	0.938 ^b	1.031 ^a	1.308 ^{ab}
HH+H+H+			1.335 ^{ab}	2.345 ^a	3.590		1.002 ^{ab}	1.010 ^{ab}	1.245 ^b
	Probability								
Treatment	≤ 0.01	\leq 0.01	≤ 0.01	≤ 0.01	0.53	≤ 0.01	≤ 0.01	≤ 0.01	0.03
SEM	0.000	0.003	0.007	0.009	0.015	0.003	0.005	0.009	0.014

Table V-9. Body weight and body weight gain of Cobb $700 \times MV$ mixed sex broilers in experiment 1 fed diets containing varying nutrient density to 49 d of age.

			Fee	d Conversion Rat	io ²		
	Starter	Grower	Finisher	Withdrawal	1-26	1-36	1-49
LLLL	1.269 ^a	1.403 ^{ab}	1.821	2.000	1.373 ^a	1.565 ^a	1.718 ^{ab}
MLLL		1.417 ^a	1.835	2.040	1.374 ^a	1.563 ^a	1.733 ^a
MMMM	1.231 ^b	1.364 ^{bc}	1.808	1.996	1.335 ^b	1.532 ^b	1.693 ^b
MHHH		1.351 ^{cd}	1.792	2.034	1.325 ^b	1.530 ^b	1.712 ^{ab}
HMMM		1.373 ^{abc}	1.804	2.042	1.328 ^b	1.528 ^b	1.717 ^{ab}
НННН	1.174 ^c	1.337 ^{cd}	1.823	2.023	1.296 ^{bc}	1.518 ^{bc}	1.690 ^{bc}
HH+H+H+		1.310 ^d	1.773	2.006	1.282°	1.491°	1.656 ^c
				Probability -			
Treatment	≤ 0.01	≤ 0.01	0.60	0.40	≤ 0.01	≤ 0.01	≤ 0.01
SEM	0.007	0.007	0.009	0.019	0.007	0.005	0.005

Table V-10. Feed conversion ratio of Cobb $700 \times MV$ mixed sex broilers in experiment 1 fed diets containing varying nutrient density to 49 d of age.

			Feed	Intake (g/bird	l/d)			dLys Intake (g/bird)		
	Starter	Grower	Finisher	Withdrawal	1-26 d	1-36 d	1-49 d	1-49 d		
LLLL	27.6 ^b	96.2 ^b	167.2 ^e	204.8 ^a	64.8 ^c	93.2°	121.7 ^{cd}	60.0 ^g		
MLLL		103.1ª	170.4 ^{de}	202.1 ^{ab}	71.2ª	99.1ª	126.1 ^a	61.9 ^f		
MMMM	31.4 ^a	101.1 ^a	172.2 ^{cd}	191.6 ^{de}	69.3 ^b	98.2 ^{bc}	122.5 ^{cd}	64.2 ^e		
MHHH		93.0 ^b	177.4 ^{ab}	196.9°	65.2°	96.7 ^b	122.7 ^{bcd}	67.6°		
HMMM		102.1ª	174.9 ^{bc}	195.0 ^{cd}	69.9 ^{ab}	99.6 ^a	124.8 ^{ab}	65.5 ^d		
HHHH	30.9 ^a	96.9 ^b	178.8ª	198.1 ^{bc}	66.3°	97.6 ^{ab}	123.3 ^{bc}	69.0 ^b		
HH+H+H+		94.8 ^b	173.9 ^{bcd}	189.9 ^e	66.2°	96.4 ^b	120.5 ^d	70.5 ^a		
	Probability									
Treatment	≤ 0.01	≤ 0.01	\leq 0.01	≤ 0.01	≤ 0.01	≤ 0.01	≤ 0.01	≤ 0.01		
SEM	0.210	0.648	0.656	0.891	0.364	0.323	0.355	0.007		

Table V-11. Feed intake and digestible lysine (dLys) intake of Cobb $700 \times MV$ mixed sex broilers in experiment 1 fed diets containing varying nutrient density to 49 d of age.

Experiment 1 Processing. Stepwise changes in breast, tender, and fat pad yield were observed with increasing dEAA density in Exp 1 (Table 12). Breast meat yield was increased ($p \le 0.05$) in broilers fed the MMMM and HHHH by 4.55% compared to those fed the LLLL. Breast meat yield was maximized in broilers fed the HH+H+H+ diets, leading to an 8.18% increase ($p \le 0.05$) compared to those fed the LLLL. Tender yield was increased ($p \le 0.05$) by 4.43% in birds fed the HH+H+H+ treatment compared to all other treatments. Fat pad yield was reduced ($p \le 0.05$) by 9.20, 14.94, and 28.74% in broilers fed the MHHH, HHHH, HH+H+ treatments compared to MLLL. No dietary treatment effects on the severity of woody breast were observed.

		L	ive Weight Yield (%)		Meat Quality
Treatment ²	Wing	Wing Tenderloin Brea		east Fillet Leg		Woody Breast Score ³
LLLL	8.14	4.71 ^b	22.0 ^c	23.6	1.73 ^{ab}	0.85
MLLL	8.09	4.71 ^b	22.7 ^{bc}	23.5	1.74 ^a	0.86
MMMM	8.03	4.75 ^b	22.9 ^b	23.7	1.68 ^{ab}	0.90
MHHH	8.10	4.78 ^b	22.7 ^{bc}	23.9	1.58 ^{bc}	0.81
HMMM	8.10	4.73 ^b	23.2 ^{ab}	23.4	1.60 ^{abc}	0.97
HHHH	8.10	4.74 ^b	23.0 ^b	23.9	1.48 ^c	0.79
HH+H+H+	7.98	4.95 ^a	23.8ª	23.4	1.24 ^d	0.93
			Pr	obability ——		
Treatment	0.84	0.05	≤ 0.01	0.31	≤ 0.01	0.58
SEM	0.031	0.025	0.099	0.075	0.028	0.032

Table V-12. Processing attributes (d 50) of Cobb 700 \times MV mixed sex broilers in experiment 1 fed diets containing varying nutrient density to 49 d of age¹.

¹Observed means were calculated from 12 replicate values using the pen as the experimental unit. Each replicated pen contributed 3 male and 3 female carcasses.

 2 Treatments from 0-11 d are L (1.22% dLys), M (1.30% dLys) and H (1.45% dLys); from 11-25 d are L (1.10% dLys), M (1.17% dLys), H (1.23% dLys) and H+ (1.30% dLys); from 25-36 d are L (0.95% dLys), M (1.03% dLys), H (1.10% dLys) and H+ (1.17% dLys); from 36-49 d are L (0.90% dLys), M (0.95% dLys), H (1.00% dLys) and H+ (1.05% dLys). Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

³Woody breast scoring method as described by Tijare et al. (2016).

Experiment 2 Performance. Increasing dEAA density led to significant changes in BW, BW gain, FCR, and FI throughout EXP 2 (Table 13, 14, & 15). Feeding M or H diets during the starter phase led to a 10% increase ($p \le 0.05$) in d 12 BW compared to those fed the L. Feeding HH or HH+ increased ($p \le 0.05$) d 26 BW by 9.76% compared to broilers fed LL. Additionally, feeding M or H diets during the starter phase increased ($p \le 0.05$) d 26 BW irrespective of the density fed from d 12-26. This effect was also observed at d 36, where broilers that were fed M or H diets during the starter had a 6.02% increase ($p \le 0.05$) in BW irrespective of the density fed from d 12-36. No differences were observed in d 49 BW. Starter phase BW gain was increased ($p \le 0.05$) by 11.93% in broilers fed the M and H diets compared to the L. Increasing dEAA density during the grower phase led to increased ($p \le 0.05$) BW gain, with the highest increase observed in broilers fed HH versus LL diets. No differences in BW gain were observed during the finisher and withdrawal phases.

Stepwise improvements in FCR were evident during the starter and grower phases and from d 1-26, 1-36, and 1-49. Feed conversion was reduced ($p \le 0.05$) by 9.6 and 12.7 points in broilers fed the M and H starter diets compared to the L. Significant differences in grower phase FCR were observed, but these responses did not follow a consistent trend. From d 1-26, FCR was reduced ($p \le 0.05$) by 3.2 and 6.2 points in broilers fed HM and HH diets compared to those fed the LL. This effect continued from d 1-36, where feeding HM and HH diets reduced ($p \le 0.05$) FCR by 4.0 and 6.7 points compared to LL. Cumulative FCR was reduced ($p \le 0.05$) by 8.7 points in broilers fed HHHH diets compared to those fed LLLL. Increasing FI during the starter and grower phases and from d 1-26 coincided with increasing BW. Significant differences in FI were observed during the finisher phase and from d 1-36 and 1-49, but these differences did not coincide with BW or dEAA density. No differences were observed in withdrawal phase FI. Stepwise increases $(p \le 0.05)$ in dLys intake were observed with increasing dEAA density.

		Boo	ly Weight	(kg)			Body Weight Gain (kg)				
Treatment ¹	Day 0	Day 12	Day 26	Day 36	Day 49	-	Starter	Grower	Finisher	Withdrawal	
LLLL	0.040	0.325 ^b	1.322 ^d	2.276 ^b	3.271		0.285 ^b	.997°	0.954	0.995	
MLLL			1.388 ^c	2.357ª	3.306			1.031 ^d	0.970	0.996	
MMMM	0.040	0.356 ^a	1.407^{bc}	2.389 ^a	3.299		0.317 ^a	1.053 ^{bcd}	0.982	0.910	
MHHH			1.425 ^{ab}	2.385 ^a	3.288			1.077^{abc}	0.934	0.922	
HMMM			1.409 ^{bc}	2.399 ^a	3.369			1.048 ^{cd}	0.990	0.970	
HHHH	0.040	0.358ª	1.451 ^a	2.412 ^a	3.410		0.319 ^a	1.093 ^a	0.961	0.998	
HH+H+H+			1.435 ^{ab}	2.413 ^a	3.438			1.080 ^{ab}	0.971	1.057	
					Prob	abi	lity				
							-				
Treatment	0.75	≤ 0.01	≤ 0.01	≤ 0.01	0.36		≤ 0.01	0.01	0.28	0.38	
SEM	0.000	0.002	0.007	0.011	0.028		0.002	0.006	0.007	0.024	

Table V-13. Body weight (kg) and body weight gain (kg) of Cobb $700 \times MV$ mixed sex broilers in experiment 2 fed diets containing varying nutrient density to 49 d of age.

			Fee	d Conversion Rat	io ²		
	Starter	Grower	Finisher	Withdrawal	1-26	1-36	1-49
LLLL	1.282ª	1.374 ^b	1.712	2.367	1.353 ^a	1.504 ^a	1.748^{ab}
MLLL		1.410^{a}	1.737	2.472	1.356 ^a	1.513ª	1.764 ^a
MMMM	1.186 ^b	1.353 ^{cd}	1.680	2.504	1.313 ^{bc}	1.465 ^{bc}	1.727 ^{abc}
MHHH		1.337 ^{de}	1.685	2.419	1.306 ^{cd}	1.453 ^{bc}	1.706 ^{bcd}
HMMM		1.372 ^b	1.683	2.371	1.324 ^b	1.473 ^b	1.717 ^{bcd}
НННН	1.155°	1.334 ^e	1.674	2.265	1.294 ^d	1.446 ^c	1.677 ^d
HH+H+H+		1.357 ^{bc}	1.668	2.181	1.314 ^{bc}	1.456 ^{bc}	1.684 ^{cd}
				Probability -			
Treatment	≤ 0.01	≤ 0.01	0.07	0.23	≤ 0.01	≤ 0.01	≤ 0.01
SEM	0.007	0.004	0.008	0.041	0.004	0.004	0.007

Table V-14. Feed conversion ratio of Cobb $700 \times MV$ mixed sex broilers in experiment 2 fed diets containing varying nutrient density to 49 d of age.

¹Treatments from 0-11 d are L (1.22% dLys), M (1.30% dLys) and H (1.45% dLys); from 11-25 d are L (1.10% dLys), M (1.17% dLys), H (1.23% dLys) and H+ (1.30% dLys); from 25-36 d are L (0.95% dLys), M (1.03% dLys), H (1.10% dLys) and H+ (1.17% dLys); from 36-49 d are L (0.90% dLys), M (0.95% dLys), H (1.00% dLys) and H+ (1.17% dLys); from 36-49 d are L (0.90% dLys), M (0.95% dLys), H (1.00% dLys) and H+ (1.17% dLys) and H+ (1.17% dLys); from 36-49 d are L (0.90% dLys), M (0.95% dLys), H (1.00% dLys) and H+ (1.05% dLys) and H+ (1.05% dLys). Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

²Mortality corrected feed conversion ratio

		dLys Intake (g/bird)							
	Starter	Grower	Finisher	Withdrawal	1-26 d	1-36 d	1-49 d	0-49 d	
LLLL	31.1 ^b	97.3°	163.3 ^{abc}	165.2	65.1°	90.6 ^{ab}	110.7 ^a	55.3 ^e	
MLLL		103.7ª	168.3ª	158.8	67.8 ^{ab}	93.2ª	108.4 ^{ab}	56.4 ^e	
MMMM	32.2 ^c	100.9 ^b	164.8 ^{abc}	159.9	67.1 ^b	92.1 ^{ab}	109.7 ^{ab}	58.9 ^d	
MHHH		102.7 ^{ab}	158.7°	157.8	66.5 ^{bc}	88.8 ^b	105.6 ^b	61.2 ^{bc}	
HMMM		103.2ª	167.6 ^{ab}	163.3	68.3 ^{ab}	94.0 ^a	112.6 ^a	60.4 ^{cd}	
HHHH	31.5 ^{ab}	103.7 ^a	161.7 ^{abc}	163.3	68.2 ^{ab}	91.6 ^{ab}	109.6 ^{ab}	62.7 ^b	
HH+H+H+		104.6 ^a	160.9 ^{bc}	167.1	69.2ª	93.3ª	113.1ª	66.5ª	
	Probability								
Treatment	0.02	≤ 0.01	0.03	0.64	≤ 0.01	0.02	0.03	≤ 0.01	
SEM	0.158	0.514	0.994	1.760	0.336	0.481	0.763	0.009	

Table V-15. Feed intake and digestible lysine (dLys) intake of Cobb $700 \times MV$ mixed sex broilers in experiment 2 fed diets containing varying nutrient density to 49 d of age.

Experiment 2 Processing. Feeding diets with higher dEAA density led to significant changes in breast, tender, leg, and fat pad yield in Exp 2 (Table 16). Broilers that were fed the MHHH, HMMM, HHHH, HH+H+H had a 5.29% increase ($p \le 0.05$) in tender yield compared to those fed the LLLL treatment. Feeding MLLL and HH+H+H diets led to a 3.94 and 7.92% increase ($p \le 0.05$) in breast meat yield compared to those fed LLLL diets. Conversely, leg yield was increased ($p \le 0.05$) by 2.77% in broilers fed the LLLL and MMMM treatments compared to those fed the HMMM and HH+H+H diets. Broilers that were fed MMMM and HH+H+H had a 14.02 and 24.39% decrease ($p \le 0.05$) in fat pad yield compared to those fed LLLL. No dietary treatment effects on the severity of woody breast were observed.

		Meat Quality							
Treatment ²	Wing	Tenderloin	Breast Fillet	Leg	Fat Pad	Woody Breast Score ³			
LLLL	7.83	4.35 ^b	21.08 ^c	23.36ª	1.64 ^a	0.92			
MLLL	7.82	4.44^{ab}	21.91 ^b	23.16 ^{ab}	1.58^{a}	0.87			
MMMM	7.88	4.48^{ab}	22.24 ^{ab}	23.25ª	1.41 ^b	0.95			
MHHH	7.87	4.58 ^a	22.49 ^{ab}	22.90 ^{ab}	1.36 ^{bc}	0.98			
HMMM	7.86	4.54 ^a	22.19 ^{ab}	22.73 ^b	1.38 ^{bc}	1.00			
HHHH	7.92	4.51 ^a	22.39 ^{ab}	22.88 ^{ab}	1.29 ^{bc}	1.00			
HH+H+H+	7.96	4.57 ^a	22.75 ^a	22.77 ^b	1.24 ^c	1.01			
	Probability								
Treatment	0.39	0.03	≤ 0.01	0.04	≤ 0.01	0.84			
SEM	0.025	0.023	0.126	0.076	0.027	0.031			

Table V-16. Processing attributes (d 50) of Cobb 700 \times MV mixed sex broilers in experiment 2 fed diets containing varying nutrient density to 49 d of age¹.

¹Observed means were calculated from 12 replicate values using the pen as the experimental unit. Each replicated pen contributed 3 male and 3 female carcasses.

 2 Treatments from 0-11 d are L (1.22% dLys), M (1.30% dLys) and H (1.45% dLys); from 11-25 d are L (1.10% dLys), M (1.17% dLys), H (1.23% dLys) and H+ (1.30% dLys); from 25-36 d are L (0.95% dLys), M (1.03% dLys), H (1.10% dLys) and H+ (1.17% dLys); from 36-49 d are L (0.90% dLys), M (0.95% dLys), H (1.00% dLys) and H+ (1.05% dLys). Diets were formulated to maintain similar dLys: dEAA across all treatments within a given phase.

³Woody breast scoring method as described by Tijare et al. (2016).

Linear Regression Analysis. The equations for the linear regression analyses are shown in figures 1 & 2. The per replicate average dLys intake ranged from 51.9 g/bird to 72.4 g/bird from 1-49 days. When FCR was regressed to dLys intake (g/bird), a significant ($p \le 0.05$) negative linear relationship was observed. This relationship indicates that for each additional gram of dLys intake, there was a 0.5 point reduction in FCR. Similarly, a significant ($p \le 0.05$) positive linear relationship was observed between breast meat yield and dLys intake. The linear relationship indicates that for each gram increase in dLys intake there was a 0.13% increase in breast meat yield.

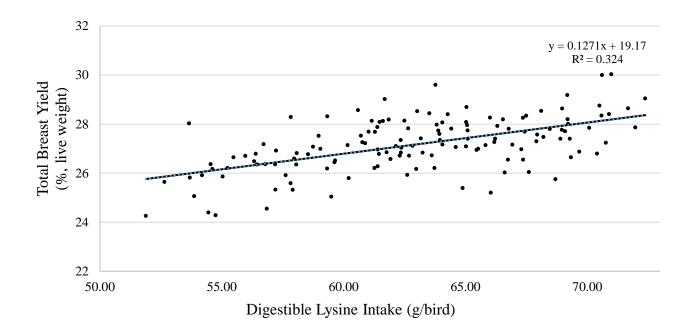


Figure V-1 Linear regression (P \leq 0.05) of total breast meat yield to digestible lysine intake in experiment 1 & 2 (n=138) from 0-49 d in Cobb 700 × MV mixed sex broilers.

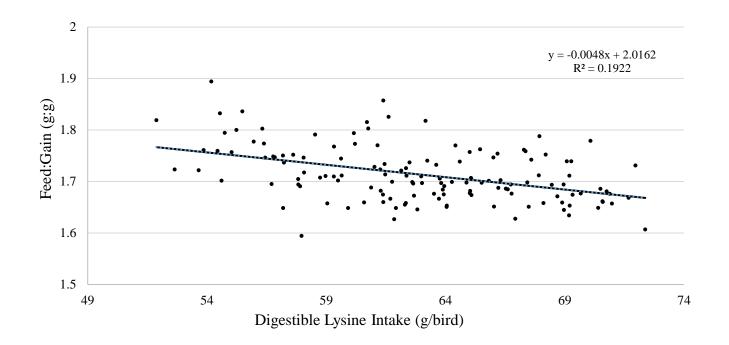


Figure V-2. Linear regression (P \leq 0.05) of mortality corrected feed conversion to digestible lysine intake in experiment 1 & 2 (n=137) from 0-49 d in Cobb 700 × MV mixed sex broilers.

Discussion

The economic viability of a feeding program is dependent upon the cost of feed ingredients and the market demand for poultry products. In heavy broilers, like the Cobb $700 \times MV$, the primary objective is to maximize the efficiency in which dietary amino acids are accreted as muscle. When heavy broilers are fed diets with increased amino acid density, meat yield and feed efficiency is improved. Studies are commonly conducted on a strain specific basis and the few reports pertaining to the Cobb $700 \times$ MV are inconclusive (Maynard et al., 2018; Maynard et al., 2019). The purpose of this research was to determine if the age in which amino acid density is increased affects the performance and meat yield at harvest in the Cobb $700 \times MV$ mixed-sex broiler. In Exp 1 and Exp 2, feeding stepwise increases in density through the duration of the study improved breast and tender yield and reduced FCR and fat pad yield. Furthermore, increasing the starter density alone was found to improve breast meat yield at harvest.

As the amino acid density of a diet increases, there is a corresponding increase in the cost of production. Economic analyses have shown that increasing amino acid density by 10% can lead to a 5.6% increase in diet cost (Kidd et al., 2005). Subsequently, the starter phase is an ideal time to increase density because only 6% of the cumulative feed intake occurs from 0-14 d in a 55 d old broiler (Cobb Vantress Inc., 2012). Furthermore, the intake of dEAA during the first week of post-hatch development affects the physiological mechanisms that control protein deposition and yield at harvest (Tesseraud et al., 1996b; Everaert et al., 2010). In Exp 1, feeding HMMM diets reduced dLys intake by 3.21% compared to MHHH diets. Despite the reduction in dLys intake, breast meat yield was numerically increased by 2.20% in broilers fed the HMMM diet compared to the MHHH diet. A similar response was observed in Exp 2, where broilers fed the MLLL diet had a 3.94% increase in breast meat yield, but consumed the same level of dLys as broilers fed the LLLL diet. These results indicate that breast meat yield at harvest is increased by feeding higher amino acid density during the starter phase. Similar findings were reported by Kidd et al. (1998), where feeding increased amino acid density to Avian $34 \times$ Avian broilers from 1-18 d improved final BW and breast meat yield irrespective of density fed from 19-50 d. Starter phase amino acid nutrition is an important factor in the control of protein deposition and muscle development, and this effect is especially apparent in the pectoralis major. For example, increasing amino acid density in diets fed to chicks for the first five days post-hatch increases protein translation in the pectoralis major (Everaert et al., 2010). Conversely, feeding lysine deficient diets to 0-14 d old broilers reduces the rate and efficiency of protein deposition in the pectoralis major by 50 and 75%, respectively (Tesseraud et al., 1996b).

Feeding diets with high amino acid density is an established method to enhance broiler live performance through the entire grow-out period. Broiler diets are commonly formulated using a dLys: dEAA ratio, where increasing dLys leads to a proportional increase in all other dEAA. Thus, evaluating dLys intake is a satisfactory approach to assess overall amino acid intake. In the present study, feeding the HH+H+H+ diets led to a 17.5 and 20.3% increase in dLys intake in Exp 1 and Exp 2, respectively. Furthermore, cumulative feed conversion was reduced by 7.7- and 8.7-points in broilers fed the HH+H+H+ diet in Exp 1 and the HHHH diet in Exp 2, respectively. Linear regression analysis showed that for each gram increase in lysine intake there was a 0.5-point improvement in feed conversion. Cumulative FCR was not affect by starter phase amino acid density and was only improved when broilers were fed increased density through the duration of the study. Similarly, Maynard et al. (2018) found that increasing total lysine intake by 5.51% improved cumulative FCR by 5.0-points in 46 d old Cobb 700 × MV mixed-sex broilers. Furthermore, that study found that the amino acid density fed in the starter phase did not affect cumulative FCR.

Increasing amino acid density improved BW at d 12, 26, and 36 in Exp 1 and Exp 2, but no differences were present in either at d 49. Similarly, Dozier III et al. (2006a) found that 35 d BW was increased in broilers fed increased amino acid density, but this improvement did not continue to 46 days. Sexual dimorphism can lead to a divergence in BW by 21 d of age in male and female broilers (Fernandes et al., 2013). Thus, the use of a mixed-sex flock in the present study may have suppressed treatment differences in final BW. Increasing dietary amino acid density improved BW gain except during the withdrawal phase of Exp 2, where broilers that were fed the LLLL diet had the highest BW gain. This response is likely an example of compensatory growth, where reducing the dietary amino acid density has a greater effect on the growth rate of young broilers. As the bird matures, they are more able to increase feed intake in a compensatory attempt to obtain the limiting amino acids required for optimal growth rate. Final BW may be similar in broilers fed reduced amino acid density diets, but fat deposition tends to be markedly higher (Moran Jr, 1979; Deschepper and DeGroote,

1995; Eits et al., 2003). The present data supports this assertion, as broilers fed the LLLL diets had the highest fat pad yield.

Few studies have been conducted to determine the effects of dietary amino acid density on the processing yield of the Cobb $700 \times MV$. In EXP 1 and EXP 2, feeding the highest amino acid density diet increased breast fillet yield by 8.2 and 7.9% and reduced fat pad yield by 28.3 and 24.4%, respectively. Linear regression analysis showed that for each gram increase in dLys intake, there was a 0.13% increase in total breast yield. In a similar experiment with Cobb $700 \times MV$ broilers, Maynard et al. (2018) found that increasing dietary amino acid density reduced fat pad yield, but had no effect on breast meat yield. The authors suggested that an energy deficit may have limited the effects of increasing amino acid density, but in an additional study they observed no interactive effects of AME and amino acid content on breast meat yield (Maynard et al., 2019). Furthermore, broilers in the present study had significant improvements in yield while consuming diets with lower AME and higher amino acid density than in the aforementioned studies. In an assessment of lysine and energy trends in broiler diet formulation from 2001-2017, Cerrate and Corzo (2019) observed that AME was reduced by 5 kcals per year, while lysine was increased by 0.009% per year. Additionally, the selection of strains for improved meat yield has led to a 4% increase in whole body protein, a 6% reduction in whole body fat, and an improvement in the efficiency of utilization for lysine and dietary energy. Based off the present study and the assessment conducted by Cerrate and Corzo (2019), it seems unlikely that the Cobb $700 \times MV$ is

less responsive to amino acid density or has a higher AME requirement than other high yielding broilers.

Increasing dietary amino acid density is an effective strategy to improve feed efficiency, reduce fat deposition, and increase lean tissue accretion. The objective of the present study was to evaluate commercial feeding strategies and determine if the age in which amino acid density is increased can affect performance and yield at harvest. The results show that increasing dietary amino acid density in all phases can reduce fat pad yield and FCR and increase breast and tender yield. Moreover, increasing density during from 0-12 d of age led to improved breast meat yield at harvest irrespective of the density fed from 12-49 d. The data from this study can be used by producers as a basis for determining the suitability of feeding increased amino acid density diets to the Cobb $700 \times MV$ broiler.

CHAPTER VI CONCLUSION

The newly developed Cobb 700 \times MV broiler is selected for increased feed efficiency and breast meat yield when raised to a heavier body weight. Selection for these traits in modern strains has improved the efficiency of amino acid utilization, reduced feed consumption per unit body weight gain, and increased sensitivity to changes in dietary amino acid density. As the amino acid density of a feed increases there is a corresponding increase in the cost of production. Thus, the economic viability of a feeding program is dependent upon defining the responses of a particular broiler strain to an array of dietary amino acid densities. Research pertaining to the effects of amino acid density on the performance and processing yield of the Cobb 700 \times MV is limited. Therefore, we sought to define the effects of feeding stepwise increases in density and to determine if the age in which density was increased modified these responses.

In general, increasing amino acid density improved breast and tender yield and reduced feed conversion and fat pad yield (Chapters 3, 4 & 5). Increasing amino acid and energy density independently improved feed conversion, but no interactive effects were observed. Contingent with previous research, the limited response to dietary energy supports the assertion that selection for increased meat yield leads to a proportionately greater increase in the requirement for amino acids as opposed to energy. The effects of energy density on feed intake followed a classic response, where intake was reduced

with increasing density. Reducing feed intake also reduced amino acid intake, leading to decreased breast meat yield. Fat pad yield was not affected by dietary energy level, but was reduced with each stepwise increase in amino acid density. In general, breast meat yield was maximized at a higher amino acid density than feed efficiency or weight gain. This effect supports the assertion that increased meat yield is achieved with a higher dLys estimate than performance. Feeding the highest amino acid density in all three studies led to the greatest increase in breast meat yield, therefore further research is needed to establish the density in which a breakpoint in response is observed. Contingent with previous studies, starter phase amino acid density was an important factor for the growth and development of the pectoralis major. Feeding increased amino acid density from 0-12 d improved breast meat yield at harvest irrespective of the density fed from 13-49 days. Furthermore, dLys intake was linearly associated with feed conversion and breast meat yield. The performance and processing benefits associated with increasing amino acid density were intrinsic in all five experiments and this data can be used to more appropriately feed the Cobb $700 \times MV$ broiler.

The research presented herein demonstrates the definitive yield and performance benefits when Cobb $700 \times MV$ broilers are fed increased amino acid density, but does not imply that this feeding program is profitable. The cost of feed ingredients and the market demand for poultry products dictate the economic viability of feeding increased density. Therefore, it is necessary to employ models that incorporate diet cost and yield improvements to establish the marginal return of each incremental increase in dietary amino acid density.

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Results from chapter four were used in a model that incorporated the diet costs from a producer currently feeding Cobb $700 \times MV$ mixed sex broilers. The economic model evaluated the effect of feeding experimental diets at a broiler operation processing 1.1 million birds per week at an average live weight of 3.34 kg and breast meat prices at \$2.64 per kg. Diet scenarios were conducted by inputting the ingredient matrix values, ingredient costs, and dietary constraints used by the producer at a specific operation into a least cost formulator. The dEAA content was increased by +0%, +10%, +15%, or +20% in the starter, grower, finisher, and withdrawal phases and diets were reformulated to determine the percentage change in diet cost with each increase in amino acid density. The improvements in breast meat yield and feed efficiency that were observed in chapter four were inputted into the model.

Feeding +10%, +15%, and +20% increases in amino acid density increased diet cost by 6.8, 10.5, and 14.5% compared to +0%. The economic model indicated that feeding diets with +10%, +15%, and +20% increases in amino acid density increased net profit by 1.3, 0.8, and 2.4 million \$/year/operation. Profitability was primarily affected by the market price of breast meat and the magnitude of improvement in breast meat yield due to increased amino acid density. For example, feeding +10% versus +0% amino acid density increased diet cost by 6.8%, breast meat yield by 3.1%, and net profit by 1.3 million \$/year/operation. Conversely, feeding +15% versus +0% increased diet cost by 10.5%, but only increased breast meat yield by 3.5%. Thus, net profit was reduced to 0.8 million \$/year/operation because the magnitude of breast meat yield

increase was less even though diet cost was higher. Improvements in feed conversion increased profitability, but not enough to justify the increased diet cost alone.

The results of the current experiments demonstrate the positive performance and yield effects of feeding increased amino acid density to Cobb $700 \times MV$ broilers. These improvements are in agreement with published research regarding other heavy strains. Furthermore, feeding increased density during the starter phase can be a cost effective way to improve breast meat yield at harvest. Producers must consider the demand for poultry products and the cost of feed ingredients to maintain the economic viability of feeding increased density. Economic models can be used as a tool to develop feeding strategies that maximize profitability.

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