

8-2017

## Evaluating the Impact of an All-Vegetable Protein Supplement Containing Fat Emulsifiers and Probiotics on Broiler Performance

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Evaluating the Impact of an All-Vegetable Protein Supplement Containing Fat Emulsifiers and Probiotics on Broiler Performance

A thesis submitted in partial fulfillment  
of the requirements for the degree of  
Master of Science in Agricultural Economics

by

Jordan Weil  
University of Florida  
Bachelor of Science in Animal Science, 2015

August 2017  
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This thesis is approved for recommendation to the Graduate Council.

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## **Abstract**

Currently, several niche markets, such as organic and “all natural”, are marketed within the poultry industry. However, little research has been undertaken to determine the cost-effectiveness of poultry diets used to produce chicken within these markets. Specifically, the economics and resulting profits associated with raising chicken with antibiotic-free, all-vegetable (AV) diets are not evaluated. As national chain restaurants such as Chick-fil-A and Panera Bread begin to mandate poultry suppliers raise chicken without antibiotics and use AV diets, it becomes increasingly important to evaluate the economics for integrators. Given the rapidly changing feeding strategies necessary to respond to consumers’ and wholesalers’ demands for this niche poultry, this study assesses new feeding approaches to compare their economic viability. This study focuses on the effects of feeding a proprietary vegetable protein supplement on broiler growth, feed consumption and thus performance and carcass yields. This study also identifies a feed supplement that contains additional fat sources compared to a conventional diet, as well as including probiotics to increase feed efficiency. An experiment was used to compare various custom and proprietary poultry feeds to an AV control diet by using data generated through fourteen hundred forty male broilers. Diets were formulated to accommodate ever-changing needs related to protein, energy, and nutrient requirements of the broilers; therefore, the study consists of a starter, grower, finisher, and withdrawal diet. The eight treatments within the study were: 1) All-vegetable control; 2) AV diet supplemented with a direct fed microbial (DFM); 3) All-vegetable diet with a fat emulsifier (FE); 4) All-vegetable diet containing both DFM and FE; 5) an all-vegetable proprietary blend (PB); 6) PB supplemented with the DFM; 7) PB and a FE; 8) PB containing both the DFM and FE. At 50 days of age, following an 8-hour feed withdrawal, broilers were processed to determine the economic value of each treatment

through average weights and carcass yields from breasts, wings, and leg quarters. Data was analyzed using SAS. Results showed that body weights were significantly lower for 42 day old birds fed Treatment 8, concluding that a price discount is necessary for this product to remain competitive.

## **Dedication**

For my parents. I am forever grateful for your continuous love, support, and encouragement.

Without your words of wisdom during my frantic 12 AM calls, I could have not made it this far in my academic studies. Thank you for always believing in me.

## **Acknowledgements**

I would like to express my deepest gratitude to Dr. H.L. Goodwin and Dr. Lawton Lanier Nalley for their unceasing support and advice during my graduate studies. I appreciate your constant assistance throughout this process. My academics would not be near as great without the counsel that each one of you provided me during my studies. Moreover, your inputs to my thesis have been invaluable.

I would also like to thank Dr. Karen Christensen for her ability to get this project underway. Also, I would like to express gratitude to Dr. Craig Coon for your willingness to serve on my committee and answer my endless questions. Each of your expertise in your respective fields has allowed me to develop a comprehensive knowledge base that will follow me throughout my career.

Additionally, I would like to thank Kevin Thompson for his statistical advice and Dr. Dawn Koltes for the help she contributed to this project. I also send a heartfelt thank you to the feed company that funded this research project.

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## **Introduction**

On January 1, 2006, the use of antibiotics as growth enhancers in cattle, swine, and poultry became illegal in the European Union (European Commission, 2005; Castanon, 2007). Outlawing antibiotics was primarily driven by the belief that nontherapeutic use of antibiotics in livestock could lead to antibiotic-resistant bacteria in humans through the consumption of meat from treated animals. Although current scientific literature does not support this claim, the issue of antibiotic resistance became a focus of consumers, scientists, and activists globally. Given its controversy in the European Union (EU), the use of antibiotics in poultry production became a talking point in the United States (U.S.), eventually leading to the implementation of a voluntary program for phasing out antibiotics in the livestock industry in 2009. The voluntary phase out, issued by the U.S. Food and Drug Administration (FDA), consisted of two “Guidance for Industry (GFI)” documents released by FDA to curb antibiotic use in the United States livestock industry. Differences in the EU and U.S. antibiotic guidelines can be attributed to the focus on food safety and human health in the respective countries. Although the U.S. did not mandate restrictive antibiotic use in 2009, it is possible that the government understood the public’s perception and respective influence on the meat industry. With time, consumer demand has pushed companies to reduce antibiotic use in their supply chains.

Guidance for Industry (GFI) #209, published by the FDA in 2012, resulted from increased concern over antibiotic abuse in the United States livestock industries. Antibiotic abuse often refers to the overuse of antibiotics, which occurs in both human medicine and livestock industries. Overusing antibiotics does not necessarily refer to intentional overuse of these medications, but often entails integrators or producers providing animals with antibiotics as a preventative measure to avoid disease. As such, GFI #209 details recommendations tailored



towards the judicious use of antibiotics as well as for veterinary oversight when using antibiotics to treat animals, either prophylactically or therapeutically. An additional document, GFI #213, supplements GFI #209 by providing information regarding voluntary changes related to the application of pharmaceutical products for approval within the livestock industries. A specific company may choose to revise label claims regarding antibiotic use if statements related to growth promoting characteristics are present. Both documents are intended for use within the beef, dairy, pork, and poultry industries to reduce the instance of antibiotic abuse and thus antibiotic resistance.

Antibiotic resistance poses a problem to not only the livestock industry, but also to the human medical industry. The inability of medications to effectively treat bacterial infections increases instances of severe illnesses and human fatalities that might otherwise have been prevented through antibiotic use. Failure to control bacterial presence and growth raises the possibility of antibiotic resistance and disease transmission to other humans as well as animals. Despite the fact that government entities such as the Department of Health and Human Services (HHS) and the United States Department of Agriculture (USDA) have increased veterinary oversight on antibiotic use in livestock industries, consumer groups and activists such as the Center for Food Safety and the Natural Resources Defense Council (NRDC) are vigilant about eradicating antibiotics used for growth in the American livestock industry (U.S. Department of Health & Human Services, 2017; United States Department of Agriculture, 2014). Although livestock industries including but not limited to cattle, hogs, and poultry are affected by the change in antibiotic use, it is possible that due to the industry size and respective market saturation, the poultry industry is heavily pressured to change its antibiotic use. Similarly, some American consumers have begun to prefer “safer” food or food created with minimal processed

ingredients or additives due perceived and actual health alerts (Nucci, Cuite, & Hallman, 2009). In the poultry industry, this preference has led to an increase in demand for chicken raised without the use of antibiotics as well as organically produced chicken (Diaz-Sanchez, Moscoso, Solis de los Santos, Andino, & Hanning, 2015; Crandell, et al., 2009; Nutrition Business Journal (NBJ), 2006). Raising chickens without the use of antibiotics is a relatively new concept for the poultry industry. While many consumers believe that the term “antibiotic-free” (ABF) is interchangeable with “raised without antibiotics,” (RWA) the terms do not signify the same grow-out method. All poultry products are antibiotic-free, as federal regulations require drug withdrawals before birds are processed. Integrators may choose to use antibiotics in their flocks to ensure birds are free of disease or suffering, eliminating the ability for that company to use the term “raised without antibiotics” on their product label.

Although chicken raised without antibiotics is one of the more commonly recognized niche markets within the United States, the American poultry industry has several other niche markets all of which encompass RWA. For instance, Tyson Foods labels several of its products as “all natural.” All natural products are those that have little to no processing, artificial ingredients, or preservatives (U.S. Food and Drug Administration, 2017). Other terms used by Tyson Foods include “No antibiotics ever (NAE)” as well as “NAE+,” which signifies the flock is fed a vegetarian diet without antibiotics. Poultry products labeled as NAE+ are more commonly seen in health food markets, but experts expect this market to grow very quickly due to consumer, and thus restaurant, demands.

To further complicate the details regarding RWA flocks, marketing labels on poultry products have various requirements concerning medications and feed supplements. A product labeled as RWA may include ionophores, which are not medically important in human medicine.

Ionophores are medications only used within veterinary medicine. Due to the method in which ionophores readjust how certain ions move across biological membranes, these medications are considered a specific class of antibiotics (Reinhardt, 2016). If medications other than those contained within the ionophore class are used within a flock, processors may no longer use the RWA label on product. Currently, such products can be directed to another supply chain, but it is unknown if this outlet will remain as restaurants and consumers refuse to purchase poultry raised with antibiotics. Therapeutic use of medication used within the human medical industry is prohibited for poultry products marketed as RWA. Although antibiotics cannot be used in RWA products, feed supplements such as organic acids, direct-fed microbials, and fat emulsifiers can improve weight gain and feed conversion ratios while preserving the fact that the flock was raised without antibiotics. The intention of using organic acids and direct-fed microbials (sometimes referred to as probiotics) in poultry feeds is to improve gut health. Additionally, fat emulsifiers are often used to increase fat absorption in broilers (Guerreiro, et al., 2011). Each of these feed supplements are purposefully used as methods of improving bird health without the use of antibiotics. The presence of multiple marketing labels makes it of utmost importance that both consumers and producers can correctly distinguish between the marketing terms used within the poultry industry, considering that the method, production costs, and potential market premiums associated with how the birds were raised differ for each marketing category.

One of the largest niche marketing and production categories within the American poultry industry is the organic market. According to the 2012 Census of Agriculture, sales for organic poultry (layers, pullets, and broilers) in the United States increased approximately 4.82% from 2007 to 2012 (National Agricultural Statistics Service, 2012). The demand for organic poultry meat is partially driven from the belief that meat raised without antibiotics is healthier

and “cleaner” with respect to environmental and human health as opposed to conventionally raised (non-organic) products, which are poultry products in which the bird may or may not have been treated with antibiotics to prevent disease (Jolly, Schutz, Diaz-Knauf, & Johal, 1989). Additionally, as consumers become more aware of diets fed to broilers, there is a shifting preference to purchase chickens which were raised on vegetarian diets. These diets are a relatively new concept, as non-vegetarian, diets containing meat by-products, are the conventional standard. The presence of meat proteins within poultry diet does not necessarily pose any risks, but the public perceives this as unnatural to poultry and possibly harmful due to beef recalls stemming from bovine spongiform encephalopathy (BSE) outbreaks. This new consumer awareness places poultry producers in a position to either capture a new market (marketing an all-vegetable diet) or perhaps risk losing customers as they purchase organic poultry products as a substitute for conventionally-produced poultry. Moreover, because this is a new sector of the American poultry industry, the producer must internally weigh new costs associated with producing an alternative product (organic, RWA, all-vegetable diet, etc.) to the potential premium and market share that could exist. Given the relative infancy of specialty markets such as producing poultry with an all-vegetable diet, producers are struggling to estimate new costs associated with these possible benefits.

Although the notion of niche markets for poultry products is relatively new, it is rapidly evolving. The American poultry and egg industry was estimated at \$42.8 billion in 2012 by the Census of Agriculture (United States Department of Agriculture, 2015). That same year, National Chicken Council predicted that 44% of poultry sales were contributed to the foodservice market segment (National Chicken Council, 2012). The public has increased its scrutiny of poultry served in restaurant chains in recent years. In 2014, Chick-fil-A announced its

plan to serve only chicken raised without the use of antibiotics. Since the Chick-fil-A announcement, the fast food industry has followed, making modifications to sourcing its products that have ultimately changed the scope of the United States poultry industry.

Chick-fil-A has great influence over the poultry industry, considering it was ranked #1 in 2015 and 2016 for chain shares of the chicken segment (Figure 1) (Nation's Restaurant News, 2016). In 2015, fast food retailers McDonald's and Subway, two of the five largest restaurants in the world, also declared plans to either lessen or eliminate use of antibiotics in their poultry supply chain (Forbes, 2017; McDonald's, n.d.; Subway, 2015). Figure 2 and Figure 3 illustrate the total poultry market segment sales held by McDonald's and Subway's, respectively, in 2016 (Nation's Restaurant News, 2016). In December of 2015, Papa Johns followed suit as the public began to pressure the food industry to prepare food with "cleaner" ingredients (Table 1).

The demand for chicken raised with all-vegetable (AV) diets is grabbing consumer traction as consumers strive to eliminate all "risks" associated with live production practices. As a result, the increase in demand for these types of poultry products pressures the industry to provide meat that accommodates these demands while ensuring operations remain profitable. If, like poultry produced with no antibiotics, all-vegetable diets become an industry standard and not a niche market, producers will need to know the economics of raising chickens with a vegetarian diet.

Table 1. Restaurant Chains Announcing Proposed Dates to Change Antibiotic Use Within Their Poultry Suppliers

<b>Restaurant</b>	<b>Proposed/Effective Date</b>
Burger King	2017
Chick-fil-A	December 2019
Kentucky Fried Chicken	December 2018
McDonald's USA	2017
Panera Bread	2014*
Papa Johns	Summer 2016
Pizza Hut	March 2017
Sonic	2017
Starbucks	2020
Subway	2016
Taco Bell	Q1 2017
Wendy's	2017

\*Since 2004, Panera has served chicken that was raised without the use of antibiotics; however, in 2016, 86% of chicken served within the chain was raised without antibiotics and with vegetarian diets (Kowitt, 2014; Panera Bread, 2016).

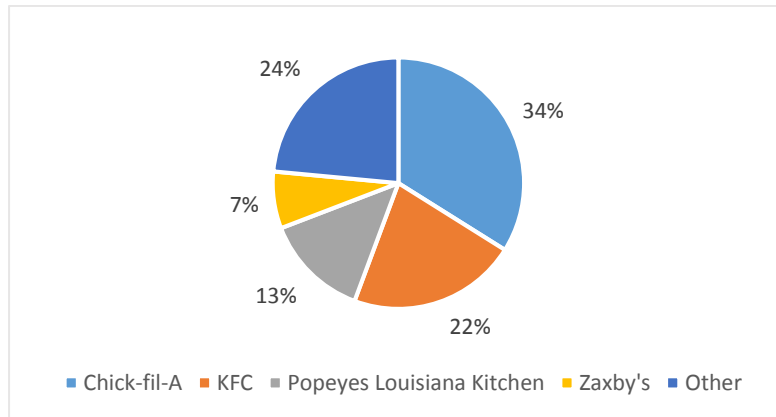


Figure 1. Market Share of Fast Food Chicken Sales

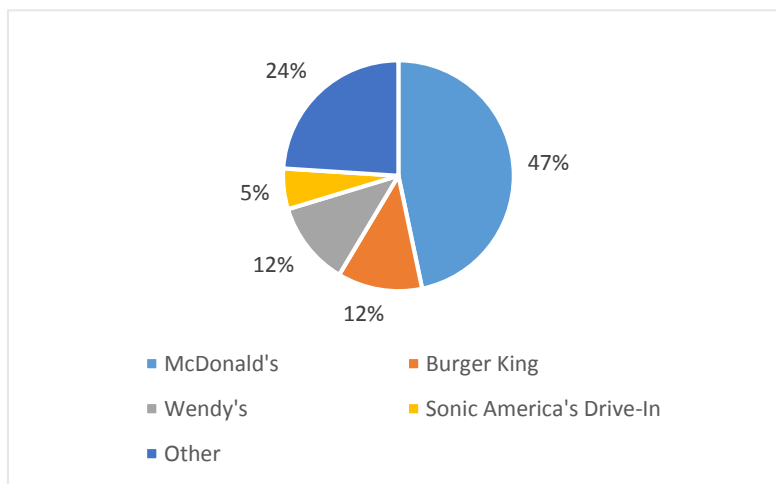


Figure 2. Market Share of Limited Service/Burger Segment

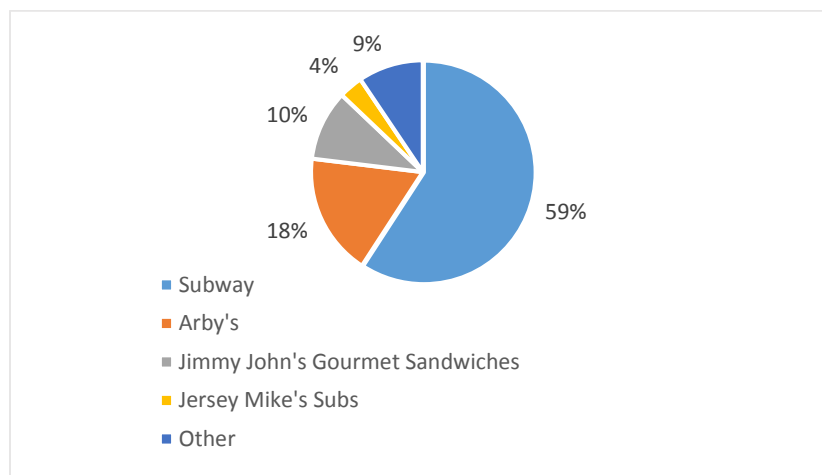


Figure 3. Market Share of Limited Service/Sandwich Segment

Source: Nation's Restaurant News, 2016

### ***Statement of the Problem***

American poultry integrators acknowledge the increase in production costs to meet new consumer demands, such as the use of all-vegetable diets, but are uncertain as to how it will affect their overall profitability. Currently, consumers are willing to pay a premium ranging from 75-100% more for niche products and as such poultry integrators are able to cover increased production costs. However, if attributes like AV diets become the industry norm and not simply a niche market, premiums may dissipate. AV diets are commonly coupled with ABF diets, so the transition from conventional flocks becomes increasingly important for integrators. As such, integrators include alternatives to antibiotics within their broiler diets to improve gut health and feed efficiency but maintain their marketing antibiotic free premium. Alternatives may include, but are not limited to: enzymes, prebiotics, probiotics, organic acids, and nutritional emulsifiers. Given there is not an elixir to solve problems related to gut health without the use of antibiotics, research is necessary to continually find alternatives to antibiotics as well as the optimal dosage rates for these supplements.

Eliminating antibiotic use in production increases flock mortality as broilers must overcome conditions such as coccidiosis and necrotic enteritis, two diseases commonly prevented in conventional operations through use of antibiotics (Greenwood, 2011). To prevent illness, producers often increase downtime between flocks and decrease bird stocking density, both of which decrease economic viability. Coupling antibiotic-free production with AV diets leads to additional obstacles, further increasing production costs. For instance, broilers do not easily digest non-starch polysaccharides commonly used in plant proteins. The inability to efficiently digest these ingredients leads to greater feed consumption, thus increasing feed costs. Feed accounts for 50-80% of live production costs (depending on flock location) in conventional



settings; therefore, operations raising broilers using vegetarian diets may increase these production expenses (Schnepf, 2011; Chiba, 2014).

### ***Objectives***

Given that experts predict AV, RWA poultry operate under higher production costs, the focus of this paper is to evaluate the performance metrics associated with feeding fat emulsifiers (FE) and direct-fed microbials (DFM) as well as the costs for an all-vegetable protein supplement. Specific objectives include the following:

- 1) Identify a direct-fed microbial that yields better weight gain and thus greater profit per bird when compared to traditional vegetable diets containing ionophores, but do not contain a DFM, protein supplement or antibiotics;
- 2) Recommend a fat emulsifier that yields better weight gain and thus greater profit per bird when compared to traditional vegetable diets containing ionophores, but do not contain a FE, supplemental protein, or antibiotics;
- 3) Determine the premium per kilogram necessary when feeding an all-vegetable feeding regimen to help maintain profits under increasing costs; and
- 4) Determine if significant differences ( $p < 0.05$ ) in weight gain, adjusted feed conversion ratios, and processing yields are present when comparing a traditional vegetable diet to a proprietary all-vegetable protein supplement, both with and without fat emulsifiers and direct-fed microbials.

## Literature Review

### *Antibiotic Use in the Poultry Industry*

Vertical integration of the American poultry industry began in the 1940s and allowed poultry companies to become more efficient by lessening production costs associated with feed mills, hatcheries, and processing. This assimilation resulted from companies purchasing various production stages, in turn allowing the company to perfect its product quality and consistency from inputs to final products. That efficiency aside, feed costs still account for up to 80% of live poultry production costs today (National Chicken Council, 2012; Schnepf, 2011). As such, the poultry industry has continually strived to reduce these costs by improving feed to live weight conversions. Feed conversions, or the amount of feed required to produce one unit of mass gain, are commonly used in the poultry industry to measure diet efficiency. On average, American poultry producers in 2015 needed 1.89 pounds of feed to produce one pound of poultry meat compared to 4.7 pounds of feed in 1925 (National Chicken Council, 2016). Although feed conversions have improved in the past 90 years, integrators continually seek to improve the feed conversion of rations while reducing feed costs.

Feed efficiency in broilers, or the ratio of weight gain (outputs) to feed consumed (inputs), is affected by a litany of production practices and by poultry physiology including, but not limited to: barn lighting, ingredient quality, and overall bird health (Mavromichalis, 2016). Gaining knowledge about the influence of these practices on broiler performance has led the industry to adopt standards regarding feed, stocking densities, and environmental conditions that have proven effective for raising cost-efficient birds. Economic viability, driven by cost-effectiveness in the industry, begins with the health of individual birds and therefore relates to overall flock health, vaccination rates, and the digestion of feed rations. Particularly,

conventional settings often include meat products, such as bone or blood meal, in broiler rations, as these diets provide the necessary nutrients required for growing broilers. A deficiency in nutrients, metabolizable energy, or digestible amino acids can lead to an increase in feed consumption as broilers search to eliminate deficiencies (Leeson, n.d.). The increase in feed intake in turn raises poultry production costs. Realizing so, respondents in a survey conducted by Capps, et al. (2015) reported that amino acid availability was of highest importance when considering feed attributes. Another technique used to lessen production costs in conventional poultry flocks is the inclusion of antibiotics in feed rations (Gustafson & Bowen, 1997). The addition of antibiotics to animal feeds is driven by the cost effectiveness and benefits of including such medications. Antibiotics can reduce feed required in live production settings as well as control diseases within the flock (Gustafson & Bowen, 1997) Depending on the company, antibiotics may or may not have not been included in the feed in sub-therapeutic levels before the FDA released its GFIs even if the poultry was conventionally raised.

The inclusion of antibiotics, first chronicled in the mid-1940s, was commonly used to prevent disease outbreaks (Moore, et al., 1946; Gustafson & Bowen, 1997). For producers, disease prevention is critical to their economic success as most avian diseases, such as necrotic enteritis and Pullorum disease, affect entire flocks as opposed to individual birds (Gustafson & Bowen, 1997; Mississippi State University, n.d.). It is also unlikely that producers can identify or treat a single bird that may experience illness or infection due to the housing methods used in the poultry industry. Commonly, in the poultry industry, thousands of birds are contained in one barn. To further complicate matters, there are instances where birds do not show clinical signs of infection, but can be infected with disease (Timbermont, Haesebrouck, Ducatelle, & Immerseel, 2011). Thus, treating all birds in a flock will ensure the ill birds not showing symptoms are

treated as well. Poultry producers also commonly experience problems associated with parasites known as coccidia, which are prevented through antibiotics (Gerhold, Jr., 2016). These parasites lead to intestinal damage within 4-7 days of infection, characterized by animal suffering, and sometimes death, resulting in financial losses for producers. In 2000, it was estimated that necrotic enteritis led to damages of \$2 billion USD to the global poultry industry (Van der Sluis, 2000).

Several countries including, but not limited to, Australia, the United Kingdom, Canada, France, and the United States have experienced problems with enteric diseases such as clostridial necrotic enteritis (Nairn & Bamford, 1967; Parish, 1961; Helmboldt & Bryant, 1971; Casewell, Friis, Marco, McMullin, & Phillips, 2003; Li, et al., 2010). Yoni Segal, a veterinary consultant for the Food and Agriculture Organization of the United Nations (FAO), estimated a cost of \$0.054/kg of poultry meat to overcome disease as opposed to \$0.016/kg for prevention measures (Segal, 2011). Thus, preventing a disease is approximately 337% more cost effective than treating it once an outbreak has occurred. A secondary benefit of including antibiotics in broiler rations is the ability for the medications to enhance feed efficiencies and, consequently, growth (Gustafson & Bowen, 1997). Since 1965, the rearing period for broilers has decreased on average by 70 days with the feed conversion ratio improving from 4.7 to 1.8 (Diaz-Sanchez, Moscoso, Solis de los Santos, Andino, & Hanning, 2015). This improvement in feed conversion is partially due to the use of antibiotics, according to Diaz-Sanchez, et al. (2015).

The specific reason for improvement in feed efficiency and growth that results from the inclusion of antibiotics in livestock production is currently debated in the literature (Lorenzoni, 2010). However, some research has shown that broad spectrum antibiotics reduce the bacterial load within the gut of various livestock species such as pigs, cattle, and poultry (Rettedal, et al.,

2009; Cromwell, 2002). As a result of the reduction in bacteria count, animals can digest more nutrients from rations improving the feed conversion ratio, in turn decreasing feed costs for producers (Rettedal, et al., 2009; Gaskins, Collier, & Anderson, 2002). Antibiotics may also be used as growth promotants and can reduce the thickness of the intestinal wall, allowing the digestive system to further absorb the included feed nutrients which is another economic benefit (Gaskins, Collier, & Anderson, 2002). The increased digestion of nutrients improves feed conversions, thus coining the term “antibiotic growth promoters” (AGP). AGP have become a controversial topic throughout food industries in many high-income countries, considering the overuse of antibiotics could lead to antibiotic resistance in the human medical industry. This has caused countries such as Sweden and Denmark to ban the inclusion of sub-therapeutic level of antibiotics within livestock feeds, which were originally intended to prevent disease (Cogliani, Goossens, & Greko, 2011).

Since Sweden banned the use of antimicrobials as growth promoters in 1986, other European countries have followed suit, prompting the European Union (EU) to enact an EU-wide ban in January 2006 (Wierup, 2001; Singer & Hofacre, 2006; European Commission, 2005). Changes in regulation enforced by the EU were partially due to the occurrence of antibiotic resistant bacteria in the medical industry, which were believed to have initiated from the use of antibiotics in the cattle, hog, and broiler livestock industries (Aarestrup, 1995; European Commission, 2005). Some researchers have since refuted this claim (Cromwell, 2002; Chang, Wang, Regev-Yochay, Lipsitch, & Hanage, 2015); however, the opinion of much of the population has caused livestock industries around the world to minimize or eliminate antibiotic use in production. A study conducted by Hwang et al. (2005) showed American consumers were intermediately concerned with antibiotics in the summer of 2002, considering that in those

surveys, it was understood there were positive effects for animals as well as negative attributes regarding antibiotic resistance. Antibiotic resistant concerns have grown in the United States, changing the landscape of the poultry industry and prompting key producers such as Tyson and Perdue to diversify their production lines to provide “no antibiotics ever” (NAE) chicken (Tyson Foods, Inc., 2017; Perdue Farms, Inc., 2016).

### ***Biosecurity Measures***

For integrators, eliminating the use of antibiotics in live production increases the frequency and severity of potential disease outbreaks (Casewell, Friis, Marco, McMullin, & Phillips, 2003). This is due in part to the lack of preventative measures that can stop or mitigate outbreaks resulting from overpopulation of bacteria within the poultry gut. Antibiotics are shown to prevent several avian diseases including, but not limited to, necrotic enteritis, coccidiosis, salmonellosis, and colibacillosis (Calnek, Barnes, Beard, McDougald, & Saif, 1997; Samad, 2000). Thus, flocks raised without antibiotics experience increased illness and mortality rates, ultimately raising production costs for producers and potential end-product prices to consumers. To prevent disease occurrence within flocks, poultry producers have chosen to implement disease prevention methods in order to reduce mortality in lieu of the use of antibiotics. Biosecurity protocols for broiler operations, regardless of flock type, have traditionally been at the center of disease reduction and have become of even greater importance with the elimination of antibiotic use. Biosecurity measures include vehicle and foot washing, limited traffic on farms, and ‘all-in, all-out’ (A IAO) systems (Clark, 2001). AIAO systems eliminate the need to transfer birds to different farms throughout their lifespan, in turn minimizing possibility of disease infection. Additionally, broilers are placed in the barn at the same time in AIAO systems. A key difference in biosecurity measures for conventional flocks and flocks raised without

antibiotics is the increased instance of vaccinations and longer downtime between flocks for RWA operations. An informal survey conducted by senior technical services veterinarian at Zoetis, Dr. Tim Cummings, showed that a broiler company committed to raising RWA flocks has made an effort to allocate at least 14 days of downtime between flocks (Thornton, 2014). Barns that house conventional flocks are recommended in the Cobb Broiler Management Guide to have a downtime of at least 12 days (Cobb-Vantress, 2013). Increasing the amount of time between placing new flocks in barns has consequences for poultry producers and result in decreased supply and increased opportunity costs, thus raising the final product price and reducing total revenue for growers.

Along with increased downtime, integrators may choose to use ionophores as a preventative mechanism against disease (Reinhardt, 2016). Although the definition of an ionophore differs between the US and EU, regulators for both have agreed that ionophores are not critically important to human medicine and thus lessen the concern about their usage in the poultry industry. These medications are derivative of antibiotics and are not used within the human medical industry, reducing the concern for the possibility of antibiotic resistance in humans (Boothe, 2016; Reinhardt, 2016). In 2005, the World Health Organization (WHO) published a document titled “Critically Important Antimicrobials for Human Medicine,” which the American poultry industry acknowledges and follows to ensure the safety of both humans and animals (World Health Organization, 2011). In the poultry industry, ionophores are commonly used as antiparasitics to prevent necrotic enteritis.

### ***Alternatives to Antibiotics***

Feed additives included in rations as a method to replace antibiotics may include, but are not limited to, prebiotics, probiotics, enzymes, and organic acids (Allen, Levine, Looft,

Bandrick, & Casey, 2013). Direct-fed microbials, a term defined by the U.S. Food and Drug Administration (FDA) as products that “contain live (viable) microorganisms (bacteria and/or yeast),” are used within the poultry industry to improve gut health (U.S. Food and Drug Administration, 1995; Jin, Ho, Abdullah, & Jalaludin, 1998; Kalavathy, Abdullah, Jalaludin, & Ho, 2003). These products are often referred to as probiotics (U.S. Food and Drug Administration, 1995). By altering the microbial population in the broiler gut, direct-fed microbials can prevent pathogenic bacteria colonization of *Clostridium* and *Campylobacter* (Yang, Iji, & Choct, 2009). Several studies have been conducted to determine if direct-fed microbials reduce both mortality rates and performance metrics (Jin, Ho, Abdullah, & Jalaludin, 1998; Jahromi & Altaher, 2016). In a study conducted by Timmerman et. al. (2006), mortality rates decreased when using probiotics whereas Zulkifli, et al. (2000) found an increase in mortality. Jin, Ho, and Abdullah (1998) found improvements in body weight gain and feed conversion ratios when including a probiotic; Murry et al. (2006) did not. Research for probiotics has been inconsistent, perhaps due to environmental conditions of experiments (Yang, Iji, & Choct, 2009). Factors such as bacterial strain, inclusion levels, method of delivery, and nutrition regimens also play a role in the efficacy of probiotics, contributing to the perplexity of probiotics and their effects on broilers (Yang, Iji, & Choct, 2009; Edens, 2003).

In recent years, producers have begun to explore feed supplements in addition to antibiotic alternatives. Feed supplements are often used within the poultry industry in both conventional and specialty flocks to increase feed digestibility and nutrient absorption. Specialty flocks refers to those flocks other than those raised in conventional settings. For example, flocks that are raised to meet certain marketing strategies such as organic or cage-free are in specialty or niche markets. Although feed supplements are used in both conventional and niche market



settings, there is an increase of supplements used in RWA flocks, considering nutrient absorption is lower. Fat emulsifiers, a form of feed supplements, are sometimes fed to increase fat absorption in broilers (Guerreiro, et al., 2011). Although increased absorption and digestibility is desired throughout the lifetime of a broiler, it is especially important in younger birds, considering these animals do not absorb fat efficiently. Jozefiak et al. (2014) showed the source of fat affects feed intake and thus body weight gain as animal fats were shown to increase body weight gain more than fats from vegetable sources.

The transition to antibiotic alternatives and additional supplements is not seamless. The intricacies of the broiler digestive systems and the current lack of research on alternatives and supplements compound this situation. Currently, little is known about how probiotics may affect livestock or their effect on body weight gain due to conflicting experimental results (Yang, Iji, & Choct, 2009). For instance, experimental results from Liu et al. showed that these alternatives can improve feed efficiency, while Murry and fellow researchers showed otherwise (Liu, Lai, & Yu, 2007; Murry, Hinton, & Buhr, 2006). Although exact efficacies of specific bacterial strains of probiotics are not known, several researchers have identified supplements that improve food ration digestibility. These supplements have economic benefit if final flock market weights are shown to increase due to better feed conversions or reduced mortality.

Given the competitive nature of the poultry industry, large poultry producers each have their own proprietary alternatives that combat disease occurrence and/or improve feed efficiency; therefore, most supplements used within the industry are not publicly known. With the transition of the poultry industry towards the elimination of antibiotics, the ever present question regarding the use of antibiotic alternatives revolves around resistance. If bacteria can evolve to become resistant to engineered medications and supplements, could pathogenic bacteria resist the

underlying properties natural products? Although alternatives such as prebiotics and probiotics do not play a critical role in human health like antibiotics, the subject does pose an issue as additional flocks may need antibiotic treatment to prevent animal welfare issues or flock mortality.

### ***Types of Flocks***

The American poultry industry has developed several niche markets in response to recent changes in consumer demand (Table 2). Each poultry product varies slightly due to environmental conditions, products used in live production, or diet. Li and Hooker (2009) found that the term “antibiotic free” increases value in the meat industry, generating a price premium of 19.3 or 25.7 cents per ounce (estimated price premium is dependent on type of model). The possibility to increase the price of poultry products due to their label presents a unique opportunity to producers hoping to capitalize on specialized products within the commodity base. Although some poultry companies market these products to capture a premium, federal entities do not approve or regulate certain claims (USDA Food Safety and Inspection Service, 2013).

Table 2. Marketing Strategies Commonly Used in the Poultry Industry

Label	Description	Federal Certification Required for Label
All-vegetable	Meat proteins were not included in feed rations	No
Cage Free	A term typically used in the egg layer industry; birds are not housed in cages	No
Conventional	A standard method of raising broilers. Typically raised indoors. These flocks may be fed diets that include meat proteins	No
Free Range	Broilers have access to the outdoors	Yes
GMO-free	Animals were fed diets using ingredients that are free of genetically modified organisms (GMO)	No
Natural	No artificial ingredients or added colors used within the product. Product is only slightly processed	No
Nature Raised	Birds were raised in cage-free settings, without the use of antibiotics. Some producers choose to feed these animals vegetarian diets	No
No Antibiotics Ever	Animals were raised without antibiotics	Yes
No Antibiotics Ever Plus	Animals were raised without antibiotics. Birds were also fed all-vegetable diets	No
No Hormones	Hormones are not allowed in poultry production. The use of this statement must be followed by “Federal regulations prohibit the use of hormones.”	No
Organic	Birds have access to the outdoors. Must be fed a certified organic feed. No antibiotics may be used	Yes
Pastured	Animals are solely raised outdoors	No
Raised Without Antibiotics	Similar to “No Antibiotics Ever.” Animals were raised without the use of antibiotics.	No

### *Diets in the Poultry Industry*

All sectors in the poultry industry have specific diets composed of multiple ingredients and supplements. The diets are continually adjusted for the appropriate feeding schedule to accommodate the varying needs of growing birds. For example, integrators provide rations to growers for starter, grower, and finisher phases, traditionally fed days 0-14, days 14-28, days 28-42, respectively. This feeding schedule may change depending on grow-out age and other factors such as end usage. Additionally, integrators may choose to provide a withdrawal diet for days 42-49. The use of a withdrawal feed depends on the economic feasibility of the diet as well as the age in which the birds are to be processed. Albuquerque et al. (2003) determined that the amount of metabolizable energy within the withdrawal feed can increase carcass yields for birds processed at 49 days. Each feeding phase alters the energy, nutrients, and supplements provided to the birds as a means to accommodate the different life stages associated with growth and production.

Since the 1950s, integrators have continuously improved rations that provide nutrients necessary for optimal performance by improving feed conversions and ensuring high market weight while also preserving meat quality. In conventional settings, these diets include meat by-products such as meat and bone meal as well as grains (barley and corn) and legumes (soybeans) to provide a balanced diet in a cost effective manner. Ingredient inclusion rates have changed over the years as demand for chicken raised with AV diets has increased. In the EU, demand for AV poultry meat stems from the Bovine Spongiform Encephalopathy (BSE) outbreak first identified in the United Kingdom in 1986 (Vieira & Lima, 2005; Bradley, 1991). Banning the inclusion of animal by-products were preventative measures to reduce possibility of transmission of BSE to humans. An experiment with experimental results published in 2011 showed that

chickens are not susceptible to BSE, alleviating the concern that feeding animal products to chickens could induce the spread of BSE (Moore, et al., 2011). In the United States, however, the recent demand for poultry products raised with AV diets stem from the consumer belief that these ingredients are “more natural” (Greenwood, 2011; Vieira & Lima, 2005).

Feeding an AV diet presents additional challenges for poultry producers regarding amount of feed consumed, digestibility, and litter quality. Vegetable protein sources, such as soybean meal, contain oligosaccharides, which are not easily digested by the poultry gut. These carbohydrates increase the moisture contained in excreta, ultimately causing a wetter litter. The quality and moisture of poultry litter is important considering the wet litter is accompanied with foot pad dermatitis, which occurs when paws develop abscesses or ulcerations (Eichner, et al., 2007). These lesions can cause animal welfare problems, increased infection rates, while also decreasing bird weight and thus profitability (Mississippi State University, 2013). The existing problems such as decreased digestibility and increased foot pad dermatitis are then exacerbated by the fact that oftentimes vegetarian diets have deficiencies in essential amino acids such as methionine (Burley, Anderson, Patterson, & Tillman, 2016). Deficiencies in protein, energy, and amino acids can lead to increased feed consumption (Leeson, n.d.). Even though these problems are overcome with the use of supplements, the USDA limits the amount of synthetic supplements that are allowed within poultry feed if the product is to be labeled as “organic” (Baier, 2015). The maximum amount of synthetic amino acids that can be added to the poultry feed depends on the limiting amino acid. Methionine is one such amino acid that is often limited in organic settings to two pounds per ton (Baier, 2015). A deficiency in methionine leads to decreased body weight gain, egg production, and feather growth, all of which cause decreased efficiencies,

increased costs and ultimately higher prices (Jacob, 2013). In turn, this can result in an increase in feather pecking between broilers as well as diminished bird health and/or lifespan.

Although AV feeding regimens lead to several problems, consumers continue to demand changes such as AV diets to be made in the poultry industry. The existing literature is limited in solutions as to why consumers prefer meat raised without the use of animal proteins. Considering chickens are naturally omnivores, most poultry experts agree that many of the consumers demanding change in the industry are those not familiar with animal behavior. An organic inspector, Tracy Favre, mentioned that the increase in vegetarian chicken is prompted “by the fact that most Americans are so far removed from their food supply” (Whoriskey, 2015). This opinion resonates with many professionals, considering a change to the diet fed to poultry flocks does not necessarily mean it is any healthier. In fact, it could mean that the product imposes additional health threats to those eating the products as well as the flocks themselves.

For flocks raised with AV diets, feed ingredients and their possible contamination from mycotoxins or other fungi becomes of increasing importance to ensure the public’s health is not at risk. In vegetarian formulations, integrators often increase the amount of soybean meal used to create a diet with higher amounts of protein. This vegetable protein is also of good quality, further supporting integrators’ desire to include the ingredient in feeds. Although commonly used in vegetarian diets, including soybean meal does not necessarily designate whether the integrator used conventional or organically produced soybeans. Additionally, an increase in soy protein source contained within the diet formulation can lead to an increased instance of soy isoflavones within the consumed animal tissues. For vegetarian raised birds, this results in elevated levels of soy isoflavones in egg yolks or tissues such as the liver, kidney, heart, and muscles (Galdos, Giusti, Litchfield, & Min, 2009). Considering soy is a major allergen,

additional levels of soy isoflavones in animal tissues could pose problems for consumers with allergies to soybeans.

Even though consumer preferences have led companies within the poultry industry to reconsider the method in which broilers are raised, many poultry experts are still attempting to adopt changes that are economically feasible while ensuring the animal's welfare remains unscathed. Poultry integrators have little to no choice but to accommodate these demands, otherwise, consumers may decide to purchase other livestock commodities that choose to adapt to consumer demands more quickly. Due to the expectation that production costs will increase for those producers raising poultry with all-vegetable diets and without the use of antibiotics, integrators must develop economically efficient poultry feeds to retain their profit margins. Even though it is ideal to simply raise the cost of the final product, the premium is expected to dissipate with time. As such, this study sets out to find a superior vegetable protein supplement when compared to a vegetarian diet that does not contain additional protein. The study also intends to compare two feed supplements, a direct-fed microbial and fat emulsifier, to vegetable diets that do not contain supplements.

## **Materials and Methods**

All protocols used within this study were approved by the University of Arkansas Institutional Animal Care and Use Committee (IACUC). Approval number is 15067.

### ***Experimental Design***

The experiment follows a 2 x 2 x 2 factorial arrangement of treatments, using two basal diets and four combinations of dietary supplements, as shown in Table 3. This results in a total of 8 treatments, containing one of two possible basal diets. The two basal diets studied within this experiment were a traditional AV diet, lacking supplemental protein, and an AV proprietary blend (PB). Additionally, two supplements, a direct fed microbial and a fat emulsifier, were examined along with the basal diets. The four supplement levels that were applied to the basal diets consisted of either no supplement (control), an added direct fed microbial (DFM), a fat emulsifier (FE), or both the DFM and FE. Both basal diets were formulated to contain the same nutrient levels as recommended by the National Research Council (NRC). Diet formulations were provided by the national feed company that sponsored the trial. These eight comparisons were done to test if the proprietary protein supplement as well as the direct-fed microbial and fat emulsifier provided superior results with respect to feed conversion ratios and final parts when compared to the basic vegetarian diet.

Treatments were replicated a total of 18 times within 144 pens located at University of Arkansas System's Division of Agriculture Poultry Science Department Research Farm located in Fayetteville, Arkansas. The trial lasted 49 days, spanning from October to December 2016. A total of 10 birds, each selected from Cobb 500 by-products, were placed within each pen. To control variability from environmental factors that could occur throughout the house, the



treatments were assigned to pens at random following a randomized complete block design (RCBD). Pens were randomly assigned to each of the eight treatments.

Table 3. Experiment Design

<b>Treatment</b>	<b>Description<sup>1</sup></b>	<b>Total No. of Pens/Trt</b>	<b>No. of Birds/Pen</b>
1	AV – Control	18	10
2	AV + DFM	18	10
3	AV + FE	18	10
4	AV + DFM + FE	18	10
5	PB – Control	18	10
6	PB + DFM	18	10
7	PB + FE	18	10
8	PB + DFM + FE	18	10

<sup>1</sup>Acronyms used within the description column signify all-vegetable (AV), direct-fed microbial (DFM), fat emulsifier (FE), and proprietary blend (PB).

### ***Bird Husbandry***

Fourteen hundred forty day-old chicks were placed in a floor pen facility. All chicks were obtained from a local hatchery and directly transferred to the university research farm for placement. Chicks were then randomly assigned across 144 floor pens (10 chicks/pen, 1 ft<sup>2</sup>/chick). Only birds that appeared to be healthy were placed in the experiment. Each floor pen was treated as an experimental unit.

At least twice daily, birds were checked to examine overall health, behavior, and accessibility to feed and water. Birds with obvious signs of infection or suffering were removed from pens and culled. The weights of culled birds along with the reason for culling was recorded. Dead birds were also removed from pens. Weights of these birds were recorded. During the course of the trial, birds were not replaced for mortality.

## ***Management***

Birds were raised in an environmentally-controlled barn. Environmental conditions such as ventilation, lighting, and temperature settings were set similar to those used in commercial broiler production. On placement day, the barn temperature was set at 90 °F using digital thermostats on heaters. This temperature was reduced by 1 °F each day until the barn reached 65 °F. Both the set and the actual temperature of the barn were recorded daily. Lighting was set at 20 hours of light for the first seven days, with a total of six hours of darkness set afterwards.

Floor pens contained used pine shavings, approximately 3-4 inches deep. This bedding was used by three flocks before the trial was conducted. All caked material was removed prior to the beginning of the trial. To achieve desired depth, new shavings were added to pens if necessary.

Each pen contained one tube feeder and a standard flow nipple drinker line. The line was equipped with a regulator and four nipple drinkers to ensure access to water. Birds had *ad libitum* access to water and experimental diets throughout the trial.

## ***Dietary Treatments***

Treatment 1 was formulated as an AV control. Treatment 2 contained the AV diet plus a direct fed microbial (DFM). Treatment 3 was formulated to contain the AV diet and a fat emulsifier (FE), while treatment 4 contained the AV basal diet, DFM, and FE. Treatment 5 contained the proprietary blend (PB) as a control. Treatment 6 was formulated to contain PB and a DFM and treatment 7 contained PB and a FE. Lastly, the final treatment, treatment 8, contained PB, a DFM, and a FE.

Each treatment was fed continuously throughout the trial. Broilers were given diets from four feeding phases: starter (d 0-14), grower (d 14-28), finisher (d 28-42), and withdrawal (d 42-

49). Each diet phase was formulated to accommodate the changing needs in nutritional requirements (Tables 4 through 7). Proximate analysis results are located in Tables 8 through 11. The starter diet was crumbled to ensure digestibility of ingredients. All other feeding phases were pelleted.

Table 4. Dietary Formulations for the Starter Phase (days 0-14)

Ingredient	All-vegetable (lbs.)	Proprietary Blend (lbs.)
Corn	1180.870	1207.190
Soybean Meal	705.000	590.000
Proprietary Protein Supplement	0.000	100.000
Soy Oil	32.000	23.000
Dicalcium Phosphate	30.000	30.000
Limestone	30.000	30.000
Salt	8.130	7.810
DL-Methionine	6.000	5.000
Lysine	3.000	2.000
Trace Mineral	2.000	2.000
L-Threonine	1.000	1.000
Vitamin Premix	1.000	1.000
Biocox	1.000	1.000
Calculated Nutrient Content (%)		
Protein	22.002	21.977
Calcium	0.917	0.970
Available Phosphorus	0.440	0.4543
Sodium	0.180	0.180
Fiber	2.674	3.206
Dig. Lysine	1.190	1.191
Dig. Methionine	0.609	0.624
Dig. Met + Cys	0.907	0.921
Dig. Tryptophan	0.217	0.213
Dig. Threonine	0.778	0.791
Dig. Arginine	1.357	1.297
Calculated Nutrient Content (kcal/kg)		
ME Poultry	3001.107	2995.373

Table 5. Dietary Formulations for the Grower Phase (days 14-28)

Ingredient	All-vegetable (lbs.)	Proprietary Blend (lbs.)
Corn	1230.860	1262.290
Soybean Meal	640.000	525.000
Proprietary Protein Supplement	0.000	100.000
Soy Oil	49.000	40.000
Dicalcium Phosphate	30.000	25.000
Limestone	30.000	30.000
Salt	8.140	6.710
DL-Methionine	5.000	3.000
Lysine	3.000	2.000
Trace Mineral	2.000	2.000
Sodium Bicarbonate	0.000	2.000
L-Threonine	0.000	0.000
Vitamin Premix	1.000	1.000
Biocox	1.000	1.000
Calculated Nutrient Content (%)		
Protein	20.550	20.514
Calcium	0.909	0.922
Available Phosphorus	0.435	0.397
Sodium	0.180	0.186
Fiber	2.602	3.140
Dig. Lysine	1.107	1.108
Dig. Methionine	0.543	0.510
Dig. Met + Cys	0.826	0.792
Dig. Tryptophan	0.201	0.197
Dig. Threonine	0.682	0.695
Dig. Arginine	1.260	1.200
Calculated Nutrient Content (kcal/kg)		
ME Poultry	3079.041	3080.205

Table 6. Dietary Formulations for the Finisher Phase (days 28-42)

Ingredient	All-vegetable (lbs.)	Proprietary Blend (lbs.)
Corn	1331.720	1359.030
Soybean Meal	540.000	425.000
Proprietary Protein Supplement	0.000	100.000
Soy Oil	51.000	43.000
Limestone	30.000	30.000
Dicalcium Phosphate	25.000	25.000
Salt	5.280	3.970
DL-Methionine	4.000	3.000
Lysine	4.000	2.000
Sodium Bicarbonate	4.000	5.000
Trace Mineral	2.000	2.000
Vitamin Premix	1.000	1.000
Biocox	1.000	1.000
L-Threonine	1.000	0.000
Calculated Nutrient Content (%)		
Protein	18.546	18.447
Calcium	0.856	0.910
Available Phosphorus	0.375	0.390
Sodium	0.180	0.174
Fiber	2.518	3.052
Dig. Lysine	1.021	0.983
Dig. Methionine	0.471	0.487
Dig. Met + Cys	0.733	0.747
Dig. Tryptophan	0.177	0.173
Dig. Threonine	0.662	0.625
Dig. Arginine	1.114	1.054
Calculated Nutrient Content (kcal/kg)		
ME Poultry	3137.313	3133.803

Table 7. Dietary Formulations for the Withdrawal Phase (days 42-49)

Ingredient	All-vegetable (lbs.)	Proprietary Blend (lbs.)
Corn	1384.430	1415.760
Soybean Meal	495.000	380.000
Proprietary Protein Supplement	0.000	100.000
Soy Oil	47.000	38.000
Limestone	30.000	25.000
Dicalcium Phosphate	25.000	25.000
Sodium Bicarbonate	5.000	6.000
Salt	4.57.000	3.240
DL-Methionine	3.000	2.000
Trace Mineral	2.000	2.000
Lysine	2.000	1.000
Vitamin Premix	1.000	1.000
Biocox	1.000	1.000
Calculated Nutrient Content (%)		
Protein	17.497	17.490
Calcium	0.851	0.809
Available Phosphorus	0.372	0.387
Sodium	0.180	0.174
Fiber	2.488	3.026
Dig. Lysine	0.887	0.888
Dig. Methionine	0.412	0.429
Dig. Met + Cys	0.664	0.679
Dig. Tryptophan	0.166	0.162
Dig. Threonine	0.582	0.595
Dig. Arginine	1.049	0.990
Calculated Nutrient Content (kcal/kg)		
ME Poultry (kcal/kg)	3144.731	3147.383

Table 8. Proximate Analysis of the Starter diet (%)

	Treatment							
	1	2	3	4	5	6	7	8
Protein	21.57	22.60	22.53	23.01	21.60	21.64	22.19	22.16
Moisture	12.51	12.83	12.77	12.96	12.65	12.53	12.52	12.44
Fat	4.57	4.17	4.04	4.24	3.77	4.16	3.88	4.06

Table 9. Proximate Analysis of the Grower Diet (%)

	Treatment							
	1	2	3	4	5	6	7	8
Protein	21.14	21.16	21.37	20.33	20.23	20.31	20.38	21.04
Moisture	11.28	10.88	10.73	11.09	10.87	10.71	10.68	10.44
Fat	4.61	4.17	4.71	4.57	4.46	4.29	4.16	4.52
Fiber	1.69	2.02	2.12	1.86	2.00	1.98	1.89	1.94

Table 10. Proximate Analysis of the Finisher Diet (%)

	Treatment							
	1	2	3	4	5	6	7	8
Protein	19.03	18.77	19.37	18.97	19.44	19.07	18.51	18.38
Moisture	11.56	11.27	11.43	11.05	11.90	11.72	12.18	11.78
Fat	4.52	4.58	4.59	5.05	4.16	4.45	3.88	4.57
Fiber	1.75	1.84	1.88	1.96	1.65	1.96	1.84	2.25

Table 11. Proximate Analysis of the Withdrawal Diet (%)

	Treatment							
	1	2	3	4	5	6	7	8
Protein	17.56	17.66	18.19	16.61	16.75	17.29	17.81	17.07
Moisture	12.68	12.56	11.88	12.27	12.57	12.53	12.62	12.39
Fat	4.14	4.42	4.62	4.66	4.10	3.96	3.98	4.34
Fiber	1.83	1.76	1.89	2.05	1.94	1.98	1.92	2.05



### ***Data Collection***

Live performance was assessed through weekly pen and feed weights. Once the feed intake was recorded, the feed conversion ratio (FCR) for each pen was calculated. This ratio was also adjusted for mortality (Table 12). At 49 days of age, four birds from each pen were randomly selected for processing. The birds selected did not show obvious signs of illness or defects. Once selected, the birds were double-wing banded for processing. On day 50, selected birds were transported to the processing plant using clean coops. Each bird was individually weighed prior to processing (Table 13). Birds were then shackled, stunned, slaughtered via exsanguination, and then processed. Once feathers and viscera were removed, the hot carcass and fat pad was weighed and placed in an ice bath for two hours. Afterwards, the whole carcass was once again re-weighed and deboned. Breasts, tenders, wings, leg quarters, and skin were removed from chilled carcasses and individually weighed.

Table 12. Lifespan Adjusted Feed Conversion Ratio by Treatment

<b>Treatment</b>		<b>Mean</b>	<b>Std. Dev</b>	<b>Min</b>	<b>Max</b>	<b>Median</b>	<b>N</b>	
All-vegetable	Control	1	1.659	0.029	1.592	1.714	1.663	18
	DFM	2	1.650	0.023	1.607	1.702	1.648	18
	FE	3	1.648	0.029	1.592	1.683	1.662	17*
	DFM + FE	4	1.652	0.021	1.606	1.697	1.655	18
Proprietary Blend	Control	5	1.667	0.028	1.613	1.720	1.673	18
	DFM	6	1.674	0.025	1.643	1.740	1.670	16*
	FE	7	1.670	0.037	1.614	1.752	1.673	18
	DFM + FE	8	1.662	0.018	1.618	1.690	1.668	18

\*Deviations from 18 result from outliers

Table 13. Day 49 Average Bird Weight (kg) by Treatment

<b>Treatment</b>		<b>Mean</b>	<b>Std. Dev</b>	<b>Min</b>	<b>Max</b>	<b>Median</b>	<b>N</b>	
All-vegetable	Control	1	4.176	0.131	3.878	4.370	4.175	18
	DFM	2	4.066	0.122	3.790	4.217	4.103	18
	FE	3	4.127	0.082	3.956	4.290	4.110	18
	DFM + FE	4	4.133	0.126	3.805	4.322	4.150	17*
Proprietary Blend	Control	5	4.148	0.124	3.940	4.521	4.133	18
	DFM	6	4.092	0.129	3.805	4.265	4.099	16*
	FE	7	4.087	0.092	3.860	4.210	4.113	18
	DFM + FE	8	4.093	0.073	3.925	4.200	4.097	18

\*Deviations from 18 result from outliers

### *Statistical Analysis*

Statistical tests were performed using GLM procedures of SAS (SAS Institute Inc., Cary, NC) to identify treatment means for collected data. Using Tukey's HSD test, probability values were considered significantly different at the 5% significance level. To determine if linear, quadratic, and cubic effects were present in dietary treatments, contrast statements were used to make comparisons among the treatment means. Evaluations between treatments were completed using the following null and alternative hypotheses:

$$H_0: \mu_1 = \mu_2 = \mu_3 = \mu_4 = \mu_5 = \mu_6 = \mu_7 = \mu_8$$

$$H_A: \mu_1 \neq \mu_2 \neq \mu_3 \neq \mu_4 \neq \mu_5 \neq \mu_6 \neq \mu_7 \neq \mu_8$$

Where  $\mu_1$  through  $\mu_8$  denotes the means for treatments 1 through 8.

Due to the method in which processed birds were chosen, the live performance and subsequent carcass data were analyzed separately. The live performance data was evaluated as a  $2^3$  factorial treatment design with blocks set as random effects. The least squares regression model used to evaluate the adjusted feed conversion ratio (FCR) is as follows:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{jk} + (\alpha\beta\gamma)_{ijk} + d_l + \varepsilon_{ijkl}$$

where  $Y_{ijk}$  is the adjusted feed conversion ratio in the  $i^{\text{th}}$  level of basal diet,  $j^{\text{th}}$  direct fed microbial level, and  $k^{\text{th}}$  level of fat emulsifier in the  $l^{\text{th}}$  block,  $\mu$  is the overall mean or population mean of the response,  $\alpha_i$  is the effect of the  $i^{\text{th}}$  level of basal diet,  $\beta_j$  is the effect of the  $j^{\text{th}}$  level of direct fed microbial,  $\gamma_k$  is the  $k^{\text{th}}$  level of fat emulsifier,  $(\alpha\beta)_{ij}$  is the effect of the interaction between the  $i^{\text{th}}$  level of basal diet and the  $j^{\text{th}}$  direct fed microbial level,  $(\alpha\gamma)_{ik}$  is the interaction effect between the basal diet and the fat emulsifier,  $(\alpha\beta\gamma)_{ijk}$  is the three-factor interaction effect of the basal diet, direct fed microbial, and fat emulsifier,  $d_l$  is the random effect of the blocking factor, and  $\varepsilon_{ijkl}$  is the effect of random error. Adjusted feed conversion ratios were analyzed for

each dietary phase (d 0-14, d 14-28, d 28-42, and d 42-49) as well as for the duration of the trial (d 0-49). The average bird weight was also examined to determine if statistical differences were present either weekly, throughout dietary phases, or for the length of the trial. For this data exploration, several assumptions regarding analysis of variance (ANOVA) models were followed, such as the independence and normal distribution of errors as well as the assumption that the variances for treatments were equal. Data points greater than three standard deviations away from the grand mean were considered outliers. In total, the results of three pens were outliers.

Because the broilers processed were chosen through subsampling, 72 subsamples per treatment were analyzed to test the effects of basal diets and supplements on live weight, hot WOG, fat pad, chilled WOG, breast, wings, tenders, leg and thighs, skin, and rack weights. The processing yields were later calculated as a percentage of chilled WOG weight.

## Results and Discussion

### *Live Production*

The live production results from the trial, excluding outliers greater than 3 standard deviations from the grand mean, are reported in Tables 14 to 18. Three pens (pens 89, 107, and 133) were removed in the final data analysis, as these pens were considered outliers. Pens 89, 107, and 133 were fed diets consisting of treatments 3, 6, and 6, respectively. Analyses for live performance results including the outliers are reported in the Appendix. Results excluding outliers were robust with marginal differences among treatments. Evaluations of live performance metrics showed the blocking factor is not statistically significant.

Although initial weights recorded on day 0 did not prove to have significant differences ( $p = 0.7909$ ), differences between body weights were statistically significant on days 21 ( $p < 0.0001$ ), 28 ( $p < 0.0001$ ), 35 ( $p < 0.0001$ ), and 42 ( $p < 0.0001$ ) (Tables 14 and 15). When observing the average bird weight, the AV control diet (Treatment 1) is significantly different ( $p < 0.0001$ ) when compared to the PB containing the DFM and FE (Treatment 8) at the 0.05 significance level on day 21. A closer examination at the effect of the two basal diets showed that, on average, feeding the conventional all-vegetable blend (Treatment 1) led to a 58 gram greater bird weight when compared to birds fed the proprietary blend. On day 35, differences between Treatments 1 and 8 were once again observable, with higher weights recorded for broilers fed the all-vegetable control diet. These results demonstrate that live performance weights are negatively impacted by the addition of dietary supplements to the basal diet.

On day 42, the all-vegetable control (Treatment 1) was significantly different than the PB that includes both the direct fed microbial and the fat emulsifier (Treatment 8). These two diets led to observable differences of 140 grams/bird on average. Body weight results on day 42 show

that the inclusion of both supplements (Treatment 8) led to a significantly lower body weight by 97 grams when compared to the PB control (Treatment 5). In a large scale production setting, such differences can have substantial economic consequences, as this is a difference of approximately 0.214 pounds per bird.

To calculate the adjusted feed conversions, the amount of feed consumed (Table 16) for the respective period was divided by the total pen weight. The pen weight used within the feed conversion calculation also included the weights of birds lost to mortality, resulting in an “adjusted feed conversion,” which is a feed conversion adjusted for mortality weights. Statistical analysis of adjusted feed conversions showed that there were not statistically significant treatment differences for the duration of the trial (days 0-49); however, the period from days 0-42 showed significant differences ( $p = 0.0419$ ) between treatments (Table 17). Comparisons of feed conversions observed throughout each dietary phase were insignificant. A statistical analysis of feed conversions observed weekly (Table 18) did not show significant differences between treatments either.

Table 14. Body Weight of Male Broilers, Reported Weekly

Trt	Day 0 (g)	Day 7 (kg)	Day 14 (kg)	Day 21 (kg)	Day 28 (kg)	Day 35 (kg)	Day 42 (kg)	Day 49 (kg)
1	37.374	0.155	0.468	0.978 <sup>a</sup>	1.741 <sup>a</sup>	2.528 <sup>a</sup>	3.383 <sup>a</sup>	4.176
2	37.349	0.150	0.457	0.949 <sup>ab</sup>	1.702 <sup>abc</sup>	2.447 <sup>bc</sup>	3.300 <sup>abcd</sup>	4.066
3	37.482	0.151	0.451	0.924 <sup>c</sup>	1.689 <sup>abc</sup>	2.446 <sup>bc</sup>	3.313 <sup>abcd</sup>	4.127
4	37.719	0.155	0.465	0.967 <sup>ab</sup>	1.713 <sup>ab</sup>	2.469 <sup>ab</sup>	3.337 <sup>abc</sup>	4.132
5	37.539	0.149	0.465	0.941 <sup>abc</sup>	1.688 <sup>abc</sup>	2.437 <sup>bc</sup>	3.340 <sup>ab</sup>	4.148
6	37.842	0.148	0.457	0.917 <sup>c</sup>	1.671 <sup>bc</sup>	2.394 <sup>bc</sup>	3.263 <sup>bcd</sup>	4.092
7	37.699	0.152	0.455	0.928 <sup>bc</sup>	1.683 <sup>bc</sup>	2.394 <sup>bc</sup>	3.255 <sup>cd</sup>	4.087
8	37.487	0.151	0.458	0.920 <sup>c</sup>	1.652 <sup>c</sup>	2.383 <sup>c</sup>	3.243 <sup>d</sup>	4.093
P-value	0.7909	0.0838	0.5262	<0.0001	<0.0001	<0.0001	<0.0001	0.0708

<sup>a-d</sup> Treatment means not connected by the same subscript are significantly different at  $p < 0.05$ .

Table 15. Body Weight of Male Broilers on Day of Diet Change

Treatment	Day 0 (g)	Day 14 (kg)	Day 28 (kg)	Day 42 (kg)	Day 49 (kg)
1	37.374	0.468	1.741 <sup>a</sup>	3.383 <sup>a</sup>	4.176
2	37.349	0.457	1.702 <sup>abc</sup>	3.300 <sup>abcd</sup>	4.066
3	37.482	0.451	1.689 <sup>abc</sup>	3.313 <sup>abcd</sup>	4.127
4	37.719	0.465	1.713 <sup>ab</sup>	3.337 <sup>abc</sup>	4.132
5	37.539	0.465	1.688 <sup>abc</sup>	3.340 <sup>ab</sup>	4.148
6	37.842	0.457	1.671 <sup>bc</sup>	3.263 <sup>bcd</sup>	4.092
7	37.699	0.455	1.683 <sup>bc</sup>	3.255 <sup>cd</sup>	4.087
8	37.487	0.458	1.652 <sup>c</sup>	3.243 <sup>d</sup>	4.093
p-value	0.7909	0.5262	<0.0001	<0.0001	0.0708

Table 16. Amount of Feed Consumed (kg) Per Treatment

Treatment	Day 0-14	Day 14-28	Day 28-42	Day 42-49	Day 0-49
1	0.5290	1.8590	2.9030	0.3900	5.6810
2	0.5140	1.8260	2.8160	0.3890	5.5440
3	0.5150	1.8170	2.8410	0.3950	5.5680
4	0.5230	1.8330	2.8540	0.3990	5.6080
5	0.5070	1.8240	2.9130	0.4030	5.6470
6	0.5030	1.8030	2.8650	0.4100	5.5810
7	0.5120	1.8090	2.8440	0.4060	5.5710
8	0.5060	1.7890	2.8420	0.4090	5.5460

Table 17. Adjusted Feed Conversion Ratios<sup>1</sup> of Male Broilers Grown to 49 Days of Age

Trt	Feed:Gain D0-14	Feed:Gain D14-28	Feed:Gain D28-42	Feed:Gain D42-49	Feed:Gain D0-42	Feed:Gain D0-49
1	1.131 (0.016)	1.464 (0.013)	1.773 (0.015)	2.077 (0.036)	1.565 (0.006)	1.659 (0.006)
2	1.124 (0.016)	1.471 (0.013)	1.769 (0.015)	2.025 (0.036)	1.564 (0.006)	1.650 (0.006)
3	1.147 (0.016)	1.467 (0.013)	1.746 (0.016)	2.018 (0.037)	1.561 (0.006)	1.648 (0.007)
4	1.127 (0.016)	1.463 (0.013)	1.770 (0.015)	2.025 (0.036)	1.564 (0.006)	1.652 (0.006)
5	1.094 (0.016)	1.497 (0.013)	1.783 (0.015)	2.033 (0.036)	1.580 (0.006)	1.667 (0.006)
6	1.101 (0.017)	1.489 (0.014)	1.808 (0.016)	2.032 (0.038)	1.587 (0.007)	1.673 (0.007)
7	1.121 (0.016)	1.473 (0.013)	1.808 (0.015)	2.027 (0.036)	1.585 (0.006)	1.670 (0.006)
8	1.107 (0.016)	1.499 (0.013)	1.794 (0.015)	1.974 (0.036)	1.584 (0.006)	1.662 (0.006)
<i>p</i> -value	0.9854	0.8465	0.6335	0.9185	0.0419	0.1197

<sup>1</sup> Feed:Gain ratio was adjusted for mortality was calculated by dividing the feed consumed by the pen weight including mortality weights



Table 18. Weekly Adjusted Feed Conversion Ratios<sup>1</sup> of Male Broilers Grown to 49 Days

Trt	Feed:Gain D0-7	Feed:Gain D7-14	Feed:Gain D14-21	Feed:Gain D21-28	Feed:Gain D28-35	Feed:Gain D35-42	Feed:Gain D42-49
1	1.040 (0.024)	1.181 (0.025)	1.525 (0.029)	1.430 (0.017)	1.786 (0.026)	1.772 (0.027)	2.077 (0.036)
2	1.057 (0.024)	1.168 (0.025)	1.543 (0.029)	1.429 (0.017)	1.803 (0.026)	1.743 (0.027)	2.025 (0.036)
3	1.056 (0.025)	1.198 (0.026)	1.563 (0.030)	1.413 (0.018)	1.796 (0.027)	1.729 (0.028)	2.018 (0.037)
4	1.018 (0.024)	1.184 (0.025)	1.523 (0.029)	1.431 (0.017)	1.825 (0.026)	1.740 (0.027)	2.025 (0.036)
5	1.007 (0.024)	1.137 (0.025)	1.592 (0.029)	1.443 (0.017)	1.862 (0.026)	1.732 (0.027)	2.033 (0.036)
6	1.016 (0.025)	1.139 (0.027)	1.632 (0.031)	1.407 (0.018)	1.896 (0.028)	1.742 (0.029)	2.032 (0.038)
7	1.008 (0.024)	1.180 (0.025)	1.577 (0.029)	1.412 (0.017)	1.887 (0.026)	1.761 (0.027)	2.027 (0.036)
8	1.012 (0.024)	1.155 (0.025)	1.594 (0.029)	1.445 (0.017)	1.862 (0.026)	1.743 (0.027)	1.974 (0.036)
<i>p</i> - value	0.8306	0.9994	0.8278	0.8075	0.1927	0.9449	0.6107

<sup>1</sup> Feed:Gain ratio was adjusted for mortality was calculated by dividing the feed consumed by the pen weight including mortality weights.

### ***Processing***

The live weights recorded before processing on day 50 did not show significant differences between treatments at the 0.05 significance level; nonetheless, the treatments approached significance with a *p*-value of 0.1147 (Table 19). An increase in live weight was observed when adding supplements (Treatment 8) to the basal diet containing proprietary blend (Treatment 5). Supplementing the diet with the direct-fed microbial led (Treatment 6) to a weight gain of 32.708 grams per bird. Statistically significant differences at the 0.05 level were observable for both pre- and post- chill WOG weights (Table 20). The WOG weight, or carcass weight without giblets, showed significant differences of 104 and 105 grams per bird respectively between treatments 1 (highest) and 3 (lowest) for the pre-chill ( $p = 0.0157$ ) and post-chill ( $p = 0.0111$ ) measurements. Differences between treatments consisting of the

proprietary protein supplement were not statistically significant. However, birds in treatment 6 had the highest WOG weight out of this basal diet, with the post-chill WOG weight 31.706 grams higher than those fed the PB control diet (Treatment 5).

No differences in fat pad ( $p = 0.9203$ ) or skin ( $p = 0.3821$ ) weights were observed between the traditional all-vegetable protein sources and the proprietary blend (Tables 20 and 21). The diets did not produce significant differences in rack, breast, wing, or leg quarter weights upon observing the treatment means (Table 22). Tender weights were significantly different ( $p = 0.0081$ ) between Treatments 1 and 2. The broilers fed diets containing the all-vegetable control had higher weights ( $p < 0.05$ ) than those fed the all-vegetable diet that contained the direct-fed microbial; mean tenders weights differed approximately 8.4 grams between these two treatments (Treatments 1 and 2). A comparison of white meat and dark meat processing yields did not show significant differences (Tables 23 and 24). Examination of breasts did not reveal significant differences of white striping ( $p = 0.8285$ ) or woody breast ( $p = 0.4603$ ) between treatments (Table 25).

Table 19. Slaughter and Carcass Yield Weights of 50 day Old Male Broilers

Treatment	Live Slaughter Weight	Pre-Chill WOG Weight <sup>1</sup>	Post-Chill WOG Weight <sup>2</sup>
1	4159.708	3235.889 <sup>a</sup>	3219.472 <sup>a</sup>
2	4067.292	3150.194 <sup>ab</sup>	3130.583 <sup>ab</sup>
3	4046.083	3132.486 <sup>b</sup>	3114.500 <sup>b</sup>
4	4115.889	3201.167 <sup>ab</sup>	3182.806 <sup>ab</sup>
5	4084.889	3156.250 <sup>ab</sup>	3137.769 <sup>ab</sup>
6	4117.597	3189.194 <sup>ab</sup>	3169.083 <sup>ab</sup>
7	4101.097	3177.903 <sup>ab</sup>	3164.347 <sup>ab</sup>
8	4102.102	3174.125 <sup>ab</sup>	3155.306 <sup>ab</sup>
<i>p</i> -value	0.1147	0.0157	0.0111

<sup>1</sup> Carcass weight without giblets, before chilling the carcass in an ice bath.

<sup>2</sup> Carcass weight without giblets, after chilling the carcass for two hours.

<sup>a-b</sup> Treatment means not connected by the same subscript are significantly different at  $p < 0.05$ .

Table 20. Fat Pad Weights of 50 Day Old Male Broilers

Treatment	Fat Pad Weight
1	48.917
2	47.847
3	47.375
4	48.917
5	50.903
6	46.875
7	45.750
8	48.625
<i>p</i> -value	0.9203

Table 21. Skin Weights of 50 Day Old Male Broilers

Treatment	Skin Weight
1	114.931
2	112.736
3	117.000
4	117.000
5	118.278
6	116.944
7	114.514
8	113.139
<i>p</i> -value	0.3821

Table 22. Processing Weights (g) of Male Broilers

Treatment	Live	Rack	Breast	Tender	Wing	Leg Quarter
1	4159.708	746.375	913.750	175.500 <sup>a</sup>	316.694	946.556
2	4067.292	721.069	893.417	167.069 <sup>b</sup>	307.681	923.694
3	4046.083	722.458	870.792	170.042 <sup>ab</sup>	312.014	917.847
4	4115.889	738.889	903.569	173.417 <sup>ab</sup>	316.264	931.444
5	4084.889	730.069	882.889	170.417 <sup>ab</sup>	309.125	912.431
6	4117.597	740.444	892.167	171.514 <sup>ab</sup>	317.694	933.444
7	4101.097	720.069	901.208	172.208 <sup>ab</sup>	316.458	932.806
8	4102.102	734.819	885.000	170.472 <sup>ab</sup>	312.139	929.361
<i>p</i> -value	0.1147	0.1727	0.0304	0.0081	0.0372	0.0355

<sup>a-c</sup> Treatment means not connected by the same subscript are significantly different at  $p < 0.05$ .

Table 23. Processing Yields (%) of White Meat from 50 Day Old Male Broilers

Treatment	Rack	Breast	Tender	Total White Meat
1	23.18	28.38	5.45	33.83
2	23.03	28.54	5.34	33.88
3	23.20	27.96	5.46	33.42
4	23.22	28.39	5.45	33.84
5	23.27	28.14	5.43	33.57
6	23.36	28.15	5.41	33.56
7	22.77	28.48	5.44	33.92
8	23.29	28.05	5.40	33.45

Table 24. Processing Yields (%) of Dark Meat from 50 Day Old Male Broilers

Treatment	Rack	Wing	Leg Quarter
1	23.18	9.84	29.40
2	23.03	9.83	29.51
3	23.20	10.02	29.47
4	23.22	9.94	29.26
5	23.27	9.85	29.08
6	23.36	10.02	29.45
7	22.77	10.00	29.48
8	23.29	9.89	29.45

Table 25. White Striping and Woody Breast Scores from Broilers Selected for Processing on Day 50

Treatment	White Striping	Woody Breast
1	1.194	0.938
2	1.160	0.917
3	1.111	0.979
4	1.118	0.958
5	1.139	0.924
6	1.181	0.847
7	1.118	0.917
8	1.035	0.986
<i>p</i> -value	0.8295	0.4603

Table 26. Mean Treatment Values for Part Weights (g)

Trt	Live Weight	Hot WOG	Fat Pad	Chilled WOG	Wings	Breast	Tenders	Legs and Thighs	Skin	Rack
1	4159.7083	3235.8889	48.9167	3219.4722	316.6944	913.7500	175.5000	946.5556	114.9306	746.3750
2	4067.2917	3150.1944	47.8472	3130.5833	307.6806	893.4167	167.0694	923.6944	112.7361	721.0694
3	4046.0833	3132.4861	47.3750	3114.5000	312.0139	870.7917	170.0417	917.8472	117.0000	722.4583
4	4115.8889	3201.1667	48.9167	3182.8056	316.2639	903.5694	173.4167	931.4444	117.0000	738.8889
5	4084.8889	3156.2500	50.9028	3137.7685	309.1250	882.8889	170.4167	912.4306	118.2778	730.0694
6	4117.5972	3189.1944	46.8750	3169.0833	317.6944	892.1667	171.5139	933.4444	116.9444	740.4444
7	4101.0972	3177.9028	45.7500	3164.3472	316.4583	901.2083	172.2083	932.8056	114.5139	720.3889
8	4102.1019	3174.1250	48.6250	3155.3056	312.1389	885.0000	170.4722	929.3611	113.1389	734.3889
<i>p</i> -value	0.1147	0.0157	0.9203	0.0111	0.0372	0.0304	0.0081	0.0355	0.3821	0.1727

### ***Economic Analysis***

Examination of the average bird body weight, adjusted feed conversions, and processing results determine that the feed costs for the proprietary blend must decrease in order to remain competitive. A price premium for the exclusive product is not supported due to the inability to generate superior feed conversions or weight gains when compared to an AV control diet. In order to ensure a profitable future, integrators must consider the costs of the protein supplement and necessary feed supplements before purchasing the feedstuffs for commercial use.

## **Conclusions**

The consumer demand for vegetarian-fed chicken raised without the use of antibiotics is steadily increasing. With such demand from consumers and restaurants alike, many integrators have concluded they have no choice but to respond to these requests. An integrators' response can entail one of two options: either find alternative ways to produce AV-fed, RWA meat in a healthy manner or ignore these new consumer demands and risk losing market share. Although not ideal for integrators due to higher production costs, in the long-run, changing poultry diets to AV may be the best economic decision to ensure consumers do not purchase substitution products such as beef or pork. For integrators that primarily use conventional grow-out methods, production settings with AV, antibiotic-free diets involve special attention to bird health and animal welfare to guarantee birds are free from suffering. The additional attentiveness to broilers and their wellbeing is onset by broilers' inability to efficiently and effectively digest required amino acids, such as methionine, from all-vegetable diets. Intensifying the problems related to AV feeding regimens, flocks have greater risks of mortality when raised without antibiotics due to an increased possibility of infectious diseases. Consequently, integrators must quickly scramble to find alternatives to antibiotics as well as vegetarian supplements that increase gut health and feed efficiency. This study intended to find a direct-fed microbial and fat emulsifier to enhance both gut health and weight gain. The research also investigated the economic effects of a proprietary all-vegetable protein supplement on broiler performance. Research objectives and subsequent findings are listed in the order presented.

### ***Objective 1***

Although the direct-fed microbial was included in the diet in order to improve the birds' gut health, the supplement did not increase body weights. On day 42, body weights were

significantly ( $p < 0.0001$ ) lower for those fed the PB basal diet, DFM, and FE. Likewise, the addition of strictly the DFM and proprietary protein supplement resulted in significantly lower body weights. Adjusted feed conversion ratios were not significantly different. Tender weight was significantly ( $p = 0.0081$ ) lower by 8.431 g/bird for those fed the traditional all-vegetable diet supplemented with the direct-fed microbial. Results suggest that the direct-fed microbial does not produce desired results with respect to weight gain, adjusted feed conversion ratios, or part weights.

### ***Objective 2***

The fat emulsifier was supplemented in the diet to increase fat absorption. Unknowingly, the inclusion of this supplement to the diet resulted in significantly ( $p < 0.0001$ ) lower body weights by 0.085 kilograms. Processing weights reveal significant differences at  $p = 0.0157$  and  $p = 0.0111$  for pre- and post- chill WOG weight, respectively, when comparing the traditional diet with and without the fat emulsifier. No significant differences were observed among the supplements when independently observing the basal diet containing the proprietary protein supplement. Processing weights did not differ for those fed the fat emulsifier; therefore, results show that it is not economically efficient to use the project fat emulsifier in a commercial setting.

### ***Objective 3***

Given the results of the live performance (feed conversion ratios and feed consumed) and processing data, the proprietary all-vegetable blend does not produce desirable outcomes. The lack of significant differences in feed conversion ratios, live slaughter weights, and profit generating parts, such as breasts, determine that the proprietary blend is a commodity. Thus, the feed company should consider a price discount for this product in order to remain competitive in the poultry industry.



#### ***Objective 4***

A comparison of the traditional all-vegetable diet to the proprietary blend showed the two basal diets were not significantly different at the 5% significance level. The inclusion of both a direct-fed microbial and fat emulsifier to the basal diet failed to produce higher body weights. Comparisons of body weights for broilers fed the PB without supplements to those fed the PB with both the DFM and FE revealed a significantly ( $p < 0.0001$ ) lower mean body weight (3.340 kg/bird vs 3.243 kg/bird). Significantly different results were not detected when evaluating the adjusted feed conversion ratios. Additionally, no significant differences were identified for part weights when comparing the treatment means.

#### ***Future Research***

Although current findings based on the experimental results suggest inclusion of the direct-fed microbial, fat emulsifier, and all-vegetable proprietary protein supplement do not provide monetary gain for the commercial integrator, further research should be conducted. A deeper understanding of the inclusion of these products is necessary in the poultry industry as consumers and restaurants continue to demand all-vegetable poultry products raised without the use of antibiotics. A comparison of the treatments to a diet containing meat products will allow a better comparison of live performance and processing results between the conventional and AV feeding regimens. Additional feed analyses can also determine if diets are properly formulated with respect to metabolizable energy and digestible amino acids. Moreover, the PB should be tested in different production settings, perhaps with more replications and a greater sample size. The feed company should also consider processing each broiler in forthcoming studies. Future research practices will allow additional conclusions to be drawn by integrators hoping to improve gut health and feed conversion ratios for flocks raised without antibiotics and with

vegetarian diets. The threat of diminishing profits to companies within the industry should motivate those within to continue to improve live management practices as the price premium fades with time.

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## Appendix

Table 27. Summary of Lifespan Adjusted Feed Conversion by Treatment, Containing Outliers

Treatment		Mean	Std. Dev	Min	Max	Median	N	
All-vegetable	Control	1	1.659	0.029	1.592	1.714	1.663	18
	DFM	2	1.650	0.023	1.607	1.702	1.648	18
	FE	3	1.642	0.037	1.543	1.683	1.660	18
	DFM + FE	4	1.646	0.020	1.606	1.671	1.652	18
Proprietary Blend	Control	5	1.667	0.028	1.613	1.720	1.673	18
	DFM	6	1.668	0.029	1.622	1.740	1.667	18
	FE	7	1.670	0.037	1.614	1.753	1.673	18
	DFM + FE	8	1.662	0.018	1.618	1.690	1.668	18

Table 28. Summary of Day 49 Average Bird Weight (kg) by Treatment, Containing Outliers

Treatment		Mean	Std. Dev	Min	Max	Median	N	
All-vegetable	Control	1	4.176	0.131	3.878	4.370	4.175	18
	DFM	2	4.066	0.122	3.790	4.217	4.103	18
	FE	3	4.118	0.089	3.956	4.290	4.108	18
	DFM + FE	4	4.132	0.126	3.805	4.322	4.150	18
Proprietary Blend	Control	5	4.148	0.124	3.940	4.521	4.133	18
	DFM	6	4.062	0.159	3.665	4.265	4.071	18
	FE	7	4.087	0.092	3.860	4.210	4.113	18
	DFM + FE	8	4.093	0.073	3.925	4.200	4.097	18

Table 29. Body Weight of Male Broilers, Reported Weekly, Containing Outliers

Trt	Day 0 (g)	Day 7 (kg)	Day 14 (kg)	Day 21 (kg)	Day 28 (kg)	Day 35 (kg)	Day 42 (kg)	Day 49 (kg)
1	37.374	0.155	0.468	0.978 <sup>a</sup>	1.741 <sup>a</sup>	2.528 <sup>a</sup>	3.383 <sup>a</sup>	4.176
2	37.349	0.150	0.457	0.949 <sup>ab</sup>	1.702 <sup>abc</sup>	2.447 <sup>bc</sup>	3.300 <sup>abcd</sup>	4.066
3	37.508	0.151	0.452	0.925 <sup>c</sup>	1.692 <sup>ab</sup>	2.449 <sup>bc</sup>	3.315 <sup>abcd</sup>	4.118
4	37.719	0.155	0.465	0.967 <sup>ab</sup>	1.713 <sup>ab</sup>	2.469 <sup>ab</sup>	3.337 <sup>abc</sup>	4.132
5	37.539	0.149	0.465	0.941 <sup>abc</sup>	1.688 <sup>abc</sup>	2.437 <sup>bc</sup>	3.340 <sup>ab</sup>	4.148
6	37.861	0.149	0.458	0.918 <sup>c</sup>	1.669 <sup>bc</sup>	2.396 <sup>bc</sup>	3.256 <sup>bcd</sup>	4.062
7	37.699	0.152	0.455	0.928 <sup>bc</sup>	1.683 <sup>bc</sup>	2.394 <sup>bc</sup>	3.255 <sup>cd</sup>	4.087
8	37.487	0.151	0.458	0.920 <sup>c</sup>	1.652 <sup>c</sup>	2.383 <sup>c</sup>	3.243 <sup>d</sup>	4.093
<i>p</i> -value	0.7455	0.0924	0.5805	<0.0001	<0.0001	<0.0001	<0.001	0.0446

<sup>a-d</sup> Treatment means not connected by the same subscript are significantly different at  $p < 0.05$ .

Table 30. Body Weight of Male Broilers on Day of Diet Change, Containing Outliers

Treatment	Day 0 (g)	Day 14 (kg)	Day 28 (kg)	Day 42 (kg)	Day 49 (kg)
1	37.374	0.468	1.741 <sup>a</sup>	3.383 <sup>a</sup>	4.176
2	37.349	0.457	1.702 <sup>abc</sup>	3.300 <sup>abcd</sup>	4.066
3	37.508	0.452	1.692 <sup>ab</sup>	3.315 <sup>abcd</sup>	4.118
4	37.719	0.465	1.713 <sup>ab</sup>	3.337 <sup>abc</sup>	4.132
5	37.539	0.465	1.688 <sup>abc</sup>	3.340 <sup>ab</sup>	4.148
6	37.861	0.458	1.669 <sup>bc</sup>	3.256 <sup>bcd</sup>	4.062
7	37.699	0.455	1.683 <sup>bc</sup>	3.255 <sup>cd</sup>	4.087
8	37.487	0.458	1.652 <sup>c</sup>	3.243 <sup>d</sup>	4.093
<i>p</i> -value	0.7455	0.5805	<0.0001	<0.001	0.0446

Table 31. Adjusted Feed Conversion Ratios<sup>1</sup> of Male Broilers Grown to 49 Days of Age, Containing Outliers

Trt	Feed:Gain D0-14	Feed:Gain D14-28	Feed:Gain D28-42	Feed:Gain D42-49	Feed:Gain D0-42	Feed:Gain D0-49
1	1.131 (0.016)	1.464 (0.013)	1.773 (0.015)	2.077 (0.058)	1.565 <sup>ab</sup> (0.006)	1.659 <sup>ab</sup> (0.008)
2	1.124 (0.016)	1.471 (0.013)	1.769 (0.015)	2.025 (0.036)	1.564 <sup>ab</sup> (0.006)	1.650 <sup>ab</sup> (0.008)
3	1.142 (0.016)	1.465 (0.013)	1.747 (0.015)	1.995 (0.058)	1.560 <sup>b</sup> (0.006)	1.642 <sup>b</sup> (0.008)
4	1.127 (0.016)	1.463 (0.013)	1.770 (0.015)	2.025 (0.058)	1.564 <sup>ab</sup> (0.006)	1.652 <sup>ab</sup> (0.008)
5	1.094 (0.016)	1.497 (0.013)	1.783 (0.015)	2.033 (0.0)	1.580 <sup>ab</sup> (0.006)	1.667 <sup>ab</sup> (0.008)
6	1.097 (0.016)	1.494 (0.013)	1.808 (0.015)	2.214 (0.058)	1.589 <sup>a</sup> (0.006)	1.692 <sup>a</sup> (0.008)
7	1.121 (0.016)	1.473 (0.013)	1.808 (0.015)	2.027 (0.058)	1.585 <sup>ab</sup> (0.006)	1.670 <sup>ab</sup> (0.008)
8	1.107 (0.016)	1.499 (0.013)	1.794 (0.015)	1.974 (0.058)	1.584 <sup>ab</sup> (0.006)	1.662 <sup>ab</sup> (0.008)
<i>p</i> - value	0.9797	0.7851	0.5937	0.6408	0.0233	0.0658

<sup>1</sup> Feed:Gain ratio was adjusted for mortality was calculated by dividing the feed consumed by the pen weight including mortality weights

Table 32. Weekly Adjusted Feed Conversion Ratios<sup>1</sup> of Male Broilers Grown to 49 Days, Containing Outliers

Trt	Feed:Gain D0-7	Feed:Gain D7-14	Feed:Gain D14-21	Feed:Gain D21-28	Feed:Gain D28-35	Feed:Gain D35-42	Feed:Gain D42-49
1	1.040 (0.024)	1.181 (0.025)	1.525 (0.029)	1.430 (0.017)	1.786 (0.026)	1.772 (0.027)	2.077 (0.058)
2	1.057 (0.024)	1.168 (0.025)	1.543 (0.029)	1.429 (0.017)	1.803 (0.026)	1.743 (0.027)	2.025 (0.036)
3	1.053 (0.024)	1.191 (0.025)	1.571 (0.029)	1.405 (0.017)	1.795 (0.026)	1.731 (0.027)	1.995 (0.058)
4	1.018 (0.024)	1.184 (0.025)	1.523 (0.029)	1.431 (0.017)	1.825 (0.026)	1.740 (0.027)	2.025 (0.058)
5	1.007 (0.024)	1.137 (0.025)	1.592 (0.029)	1.443 (0.017)	1.862 (0.026)	1.732 (0.027)	2.033 (0.0)
6	1.008 (0.024)	1.138 (0.025)	1.638 (0.029)	1.411 (0.017)	1.883 (0.026)	1.754 (0.027)	2.214 (0.058)
7	1.008 (0.024)	1.180 (0.025)	1.577 (0.029)	1.412 (0.017)	1.887 (0.026)	1.761 (0.027)	2.027 (0.058)
8	1.012 (0.024)	1.155 (0.025)	1.594 (0.029)	1.445 (0.017)	1.862 (0.026)	1.743 (0.027)	1.974 (0.058)
<i>P</i> - value	0.7929	0.9995	0.8269	0.8433	0.2147	0.9458	0.6408

<sup>1</sup> Feed:Gain ratio was adjusted for mortality was calculated by dividing the feed consumed by the pen weight



MEMORANDUM

TO: Karen Christensen  
FROM: Craig N. Coon, Chairman  
DATE: 7/10/15  
SUBJECT: IACUC Approval  
Expiration Date: Jul 14, 2018

The Institutional Animal Care and Use Committee (IACUC) has APPROVED your protocol 15067 : " Effect of Feed Additives and Equipment on Performance of Commercial Broilers" with the start date of July 15, 2015

In granting its approval, the IACUC has approved only the information provided. Should there be any further changes to the protocol during the research, please notify the IACUC in writing (via the Modification form) prior to initiating the changes. If the study period is expected to extend beyond Jul 14, 2018 you must submit a newly drafted protocol prior to that date to avoid any interruption. By policy the IACUC cannot approve a study for more than 3 years at a time.

The IACUC appreciates your cooperation in complying with University and Federal guidelines involving animal subjects.

CNC/aem

cc: Animal Welfare Veterinarian