

The effects of feeding corn containing an alpha-amylase gene on the performance and digestibility of growing cattle

by

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## Abstract

Two growth performance studies and two digestibility trials were conducted to evaluate the effects of feeding Enogen feed Corn silage and corn grain to growing cattle. In Experiment 1, there were a total of four diets offered for *ad libitum* intake. The four diets consisted of two varieties of corn (Enogen Feed Corn (EFC) vs. negative isoline control corn (CON)) with two different methods of corn processing (dry-rolled corn (DRC) vs. whole-shelled corn (WC)) and were formulated to provide 1.13 Mcal NEg/kg DM. ADG and final BW tended to be greater for calves fed EFC ( $P < 0.10$ ). Feed efficiency was greater for calves fed EFC ( $P < 0.01$ ), improving by 5.50% over calves fed CON corn. In Experiment 2, a digestibility trial was conducted using 7 cannulated Holstein steers fed the same diets from Experiment 1. Ruminant pH was not affected by corn variety ( $P > 0.82$ ). Liquid passage rate was greater for CON-fed calves, which resulted in decreased digestibility. Total tract organic matter (OM) and dry matter (DM) digestibility was greater for EFC-fed calves ( $P < 0.04$ ). In Experiment 3, there were a total of four diets offered for *ad libitum* intake. Diets consisted of two varieties of corn silage (EFC vs. CON) and two varieties of DRC (EFC vs. CON) and were formulated to provide 1.11 Mcal NEg/kg DM. ADG was greater ( $P < 0.01$ ) for calves fed EFC silage and feed efficiency tended to be greater for calves fed EFC silage ( $P < 0.14$ ). Feed efficiency of calves receiving EFC silage improved by 3.30% and ADG improved by 6.00%. In Experiment 4, a digestibility trial was conducted using 8 cannulated beef steers fed the same diets from Experiment 3. Liquid passage rate ( $P > 0.20$ ), ruminant pH ( $P > 0.23$ ), and ruminant VFA concentrations ( $P > 0.35$ ) were unaffected by treatment. Numerical differences showed a 2.5% and a 2.2% increase in total tract DM digestibility and total tract OM digestibility, respectively, for calves fed the EFC silage diets.

Key Words: Enogen Feed Corn, alpha-amylase, growing cattle, corn silage, dry-rolled corn

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# **Chapter 1 - Review of Literature**

## **Introduction**

With the size of commercial feed yards today, the management of higher-risk and younger cattle becomes more difficult (Close, 2019). Approximately 17.1 million cattle were on feed in 2018 (NASS, 2019). The beef industry is constantly changing and facing different production trends. Because of this, commercial feeders are increasingly relying on backgrounding and grower yards to precondition cattle in order to improve health and immunity while achieving a desired performance outcome while in the feedlot. The stocker/backgrounder/grower segment plays a critical role in the flexibility of the beef industry to deal with cyclic and seasonal variation, along with market shocks and being able to substitute fiber for grain. Three general ways for a stocker cattle operation to generate revenue are to focus on the value of gain, upgrading cattle quality, and speculation on market trends (Peel, 2003). According to Blasi et al. (2008), only 17.2% of operations in the U.S. that background cattle are ‘pure’ stocker operations, meaning that they are only involved in stocking and backgrounding cattle. This sector of the beef industry is attractive to producers because of the low initial cost of calves and the potential for a higher return in the end. Many risks come with backgrounding lightweight cattle due to the multitude of stressors placed on calves prior to and during the receiving period, which play a critical role in the health and immunity of the animal. Receiving diets become of great significance to the health and performance of these newly arrived stressed calves because feed intake is likely to be low (Galyean and Hubbert, 1995; Galyean et al., 1999). To compensate for low feed intakes, diets should be formulated to increase the nutrient density to meet the requirements of the animal and aid in immune function.

Originally genetically engineered for the production of ethanol, Syngenta Enogen® Feed corn (EFC) contains a thermo-tolerant alpha-amylase enzyme that is activated during salivation/mastication during consumption by the animal and fermentation in the rumen, which converts starch into fermentable sugars to provide energy for the animal. The presence of this enzyme affords the calf the opportunity to digest the grain more efficiently and gain more energy. Increasing digestibility of the corn ultimately improves the feed efficiency and gain in growing calves.

### **Newly Received Stocker Calves**

Stress consists of external body forces, either climatic, nutritional, social, or internal, that tend to interfere with homeostasis and, consequently, have a negative impact on the immune system (Stott, 1981). Any novel or rare experience for an animal can be a source of stress causing physiologic and psychologic changes (Fike and Spire, 2006). An animal's reactions are governed by an interaction of genetic factors and previous experiences (Grandin, 1997). For example, Brahman-influenced cattle naturally have higher cortisol levels (indicators of stress) than English cross calves and mature bulls have naturally lower cortisol levels than steers, cows, or heifers (Grandin, 1997). Calves are subjected to many possible stressors during the receiving phase including duration of transportation, dehydration, starvation, temperature extremes, commingling during marketing, unfamiliarity with feeding and watering facilities, and weaning. To keep these potential issues under control, calves must be evaluated based on their nutritional and health status to be able to get started on the right management program. Receiving management is critical to reduce stress and maximize the performance of newly arrived calves. Producers have practiced some form of preconditioning program on calves for over 50 years

(Dhuyvetter, 2004). Preconditioning means many things to different producers, but in a typical preconditioning program calves are weaned for at least 45 days, vaccinated for clostridial and viral diseases, castrated and/or dehorned, and are trained to use bunks and automatic waterers prior to shipping. The basic concept is to have a weaning program that improves the health of the calf before they are exposed to pathogens and stressors. Preconditioning of calves has the potential to generate a \$14/calf increase in return compared to selling calves at weaning (Dhuyvetter, 2004). Arthington et al. (2008) evaluated 4 different preweaning management techniques on steers and found that early weaned steers had improved performance and feed intake on arrival compared to steers that were weaned directly prior to shipping. Step et al. (2008), found that single source ranch calves participating in a 45-day weaning program prior to shipping resulted in decreased morbidity and health costs during the receiving period compared to multiple-source calves or calves that were weaned at shipping. In some cases, preconditioning calves is not economically feasible for cow/calf producers with the additional feed costs, labor, and facilities required (Cole, 1985). Fifty percent of the total cost of preconditioning is nutrition (Cole, 1985). This is where backgrounding operations come into the system, preconditioning cattle for entry into feedlots. Commercial feeders have embraced backgrounding operations as a collection point for cattle to grow and mature, while reaching a desired weight and readiness to perform in the feedlot (Close, 2019).

After already being subjected to weaning, transportation, commingling during marketing, and feed and water deprivation, calves are then introduced upon arrival to unfamiliar feeding facilities and noises, processing, social hierarchy, mud and manure, and additional pathogens. According to a survey involving 24 consulting feedlot nutritionists, approximately 28% of receiving calves are classified as high risk (Samuelson et al., 2016). A common practice for high-

risk receiving cattle is metaphylactic treatment on arrival to decrease the incidence of an anticipated disease outbreak. The majority of feed yards in the U.S. use metaphylactic treatment for high-risk receiving calves (Samuelson et al., 2016). In their review, Fike and Spire (2006) found that mass treatment and body temperature-based treatments of calves immediately after transport reduced the percentage of calves that were later treated for respiratory disease. Another potential issue when dealing with high-risk calves is the occurrence of persistently infected (PI) calves, which can be tested for by taking an ear notch from each high-risk calf and sending the samples to a PI lab (multiple locations across the U.S.) to be analyzed (Nickell et al., 2011). PI calves contract the bovine viral diarrhea (BVD) virus from an infection during the fetal stage in utero. The calf will then continue to shed the virus throughout its lifetime. PI calves pose a great threat to calves without the virus, especially highly stressed receiving calves that are already immunocompromised (Handel et al., 2011).

### ***Transportation***

The distance calves travel from ranch to feed yard can vary substantially, reaching over 2,000 km (Fike and Spire, 2006). The greatest physiologic indicators of stress are observed during loading and unloading of the trucks and at the start of transport (Fike and Spire, 2006). Various economic impacts of transportation include losses from morbidity and mortality, shrink, and carcass quality. Cattle transportation is normally associated with some sort of feed and water deprivation, which has been thought to have an effect on ruminal characteristics and concentrations to explain low feed intakes and poor performance on arrival (Cole and Hutcheson, 1985 and Galyean et al., 1981). However, this assumption was proved false by Fluharty et al. (1996) and Fluharty et al. (1994a) who found no decreases in cellulolytic or total bacteria in the

rumen of newly weaned calves after a period of feed and water deprivation of 72 h and 8 h of trucking. Loerch and Fluharty (1999) concurred with the previous results, finding that the ruminal microbial population is able to efficiently digest substrate immediately following weaning, transport, and food and water deprivation. Cernicchiaro et al. (2012) conducted a study with cattle originating from 21 different feedlots over a 12-year period to analyze the effect of distance traveled (< 250 km - > 1000 km) and season on bovine respiratory disease (BRD) morbidity, total mortality, and performance parameters of cattle. These researchers concluded that as distance traveled increased, BRD morbidity and overall mortality significantly increased and hot carcass weight (HCW) and average daily gain (ADG) significantly decreased. There was also a significantly greater risk for BRD during the summer months. Arthington et al. (2003) determined that transportation and commingling increases the acute-phase protein response in newly weaned heifer calves as well as increases plasma cortisol concentrations, which is a key indicator of stress. Blecha et al. (1984) conducted research to determine the effect of shipping stress on immune responses in feeder calves and found that shipped steers have suppressed lymphocyte blastogenic responses. Consequently, immune function is suppressed in calves that have endured weaning and/or transport stress (Fike and Spire, 2006).

Overall, different marketing, transportation, and management programs result in calf exposure to various pathogens and stressors, which leads to a high incidence of BRD in newly received calves. BRD is easily the most detrimental problem in the cattle industry, with *Mannheimia haemolytica* being the most prevalent BRD-causing pathogen (Lofgreen et al., 1975). Each year, BRD costs the U.S. cattle industry an estimated \$800-\$900 million (Chirase and Greene, 2001) taking into account death loss, treatment costs, and losses in performance. According to Smith (1998) as cited by Edwards (2010), BRD accounts for approximately 75% of

total morbidity and 45% of total mortality in feedlots, most of which occurs during the first 45 days after arrival. Jensen et al. (1976) were in agreement, stating that the majority of cattle that died of BRD were within the first 45 days after arriving at the feedlot, while deaths occurring after 45 days on feed were attributed to digestive disorders. Being able to successfully identify visual signs of BRD morbidity in cattle is essential. Typical visual symptoms include labored breathing, lethargy, anorexia, depression, and nasal or ocular discharge.

### ***Feed Intake***

The single most important driving force affecting performance in feeder cattle is feed intake, and its prediction is one of the greatest challenges facing researchers and cattle producers alike (Galyean and Hubbert, 1995). The most significant impacts of stress are on feed intake and immunocompetency (Loerch and Fluharty, 1999). Feed intake tends to be low for newly received stressed calves or calves that have contracted BRD (Galyean and Hubbert, 1995; Galyean et al., 1999). Lofgreen et al. (1980) compared feeding free-choice alfalfa hay containing different amounts of concentrate and concluded that feeding 50-75% concentrate in the diet promoted more rapid and efficient gains and that the demand for readily available energy outweighed any cravings for roughage, compared to diets containing less than 50% concentrate. Feeding a higher roughage diet tends to decrease BRD morbidity, but also decreases ADG and dry matter intake (DMI) (Rivera et al., 2005). The small decrease in BRD morbidity does not make up for the losses in performance when feeding higher amounts of roughage in receiving diets. It is recommended that the nutrient content of receiving diets be formulated to adjust for low feed intakes (Duff and Galyean, 2007). Stressed calves eat greater quantities of high-energy dense diets (Lofgreen et al., 1975; Lofgreen, 1983; Loerch and Fluharty, 1999), and diets with higher

concentrate levels improve performance of newly received cattle (Galyean et al., 1999; Fluharty et al., 1994b; Duff and Galyean, 2007; Lofgreen and Kiesling, 1985). Feeding high-energy receiving diets do not show any detrimental impacts towards receiving cattle health, and they have the potential to provide a positive energy balance for the animals (Fluharty and Loerch, 1996; Spore et al., 2018)

## **Receiving Diets**

### ***Concentrates as an Energy Source***

Cattle diets consist of concentrates and roughage. Today, concentrate in the diet commonly consists of cereal grains or their byproducts that typically possess a high energy density. Roughage is important because it provides a “scratch factor” to stimulate rumen motility and increased saliva production to improve the buffering capacity of the rumen, therefore decreasing the incidence of acidosis. In receiving diets, roughage is frequently included at 30% or more for light-weight and yearling cattle (Samuelson et al., 2016). Concentrates provide an efficient and abundant source of energy for ruminant animals. A main benefit of feeding grain-based instead of roughage-based diets is the price per unit of energy in grain vs. forage. Lightweight and stressed receiving cattle have high energy requirements that are hard to achieve because of low feed intake and possible nutrient deficiencies upon arrival.

Receiving cattle are typically fed diets containing 50-75% concentrates (Lofgreen et al., 1980; Samuelson et al., 2016). The most common grains fed to cattle are corn, sorghum, wheat, barley, and oats. In order of starch content: wheat (77%) > corn and sorghum (72%) > barley and oats (58%) (Huntington, 1997). In order of ruminal digestibility: wheat (94%) = oats (94%) > barley (93%) > corn (73%) > sorghum (66%) (Huntington, 1997). Starch granules are very

susceptible to digestion by ruminal bacteria. The digestibility of cereal grains is generally improved as the degree of processing increases, because of increases in the surface area of the grain for ruminal microbes to attach and the gelatinization of the starch in the grain disrupting the granular structure.

Corn grain is the primary ingredient used in both receiving and finishing rations in the U.S. (Samuelson et al., 2016). Ninety million acres of corn are planted annually in the United States, which accounts for over 95% of feed grain production and use (USDA-ERS, 2018). Based on availability, processing flexibility, and consistent starch content, corn is more economically feasible to include in receiving cattle diets and there is less risk of digestive upset compared to feeding other grains, which vary in starch content (Herrera-Saldana et al., 1990) and might be less readily available in the U.S.

### ***Dietary Characteristics of Corn Grain***

Corn contains 72% starch, which is responsible for the high energy content of the grain. The corn kernel is made up of three components: the pericarp, the germ, and the endosperm. The pericarp and germ are responsible for water uptake and contain very little starch. The majority of the starch remains in the endosperm, which is made up of three layers: the aleurone layer, the subaleurone layer, and the floury endosperm. The floury endosperm has very little cellular structure and has the highest density of starch granules, which are the most susceptible to enzymatic hydrolysis (Kotarski et al., 1992). Starch is a glucan composed of amylose and amylopectin. The majority of cereal starches generally contain 20-30% amylose and 70-80% amylopectin, which are held together in starch granules by hydrogen bonding (Rooney and Pflugfelder, 1986). Waxy grains have starches high in amylopectin and are more readily digested

than nonwaxy grains (Kotarski et al., 1992). The digestion of starch as energy occurs in 3 phases (Owens et al., 1986; Harmon et al., 2004; Huntington, 1997). The process begins in the lumen of the duodenum by alpha-amylase secreted from acinar cells in the pancreas, which initiates starch breakdown of amylose and amylopectin to produce dextrans and linear oligosaccharides. The second phase moves to the brush border membrane to yield glucose from brush border carbohydrases and the final phase is the transport of glucose out of the lumen of the intestine and into the blood stream. Seventy percent of starch that is enzymatically digested into glucose in the small intestine appears in the blood stream (Huntington et al., 2006).

According to a review conducted by Harmon et al. (2004), focusing on high-concentrate diets fed to growing cattle, approximately 77% of starch ingested is digested in the rumen. Fermentation in the rumen produces three major volatile fatty acids (VFA): acetate, propionate, and butyrate. Diets high in fermentable grain starch are associated with large propionate proportions that are responsible for providing energy to the animal (Martin et al., 1999; Orskov, 1986). Propionic acid also acts as a major hydrogen-sink product, which decreases methane production in the rumen. Energy from starch fermentation in the rumen is necessary to increase ruminal outflow of microbial protein to provide amino acids to the host animal (Huntington, 1997; Rowe et al., 1999). Starch digesting ruminal microbes, *Streptococcus bovis*, *Butyrivibrio fibrisolvens*, and *Bacteroides rumenicola*, readily attach to carbohydrates (Cotta, 1992) forming a microbial biofilm that covers the surface of feed particles (McAllister et al., 1994). The microbial population in the rumen responds quickly to carbohydrates, as they are extremely susceptible to ruminal fermentation. This can also result in digestive upsets, like acidosis, from a decrease in pH due to a rapid increase in VFA production in tandem with a reduction in ruminal motility. Acidosis results from an accumulation of lactate in the rumen, which causes the

decrease in ruminal pH. Low ruminal pH from high-starch diets can depress fiber digestion as well because of decreased ruminal motility, decreased acetate:propionate ratio, and the inactivation of fiber digesting microbes in a lower pH environment (Orskov, 1986). However, to avoid digestive upsets, some of the concentrate in the diet can be in the form of byproducts which contain highly digestible fiber rather than starch.

A review by Owens et al. (1986) of 40 different research trials suggests that between 18 and 42% of dietary starch from corn and sorghum grains fed to cattle reaches the small intestine for digestion. Increasing quantities of starch digested in the small intestine can also increase the percentage of starch that reaches the large intestine for digestion. Digestion in the large intestine is the least efficient and must be avoided (Harmon and McLeod, 2001; Mayes and Orskov, 1974; Owens et al., 1986). Ruminal conversion of starch to glucose in the rumen is approximately 64% as energetically efficient as the conversion of starch to glucose in the small intestine (Huntington et al., 2006). Although small intestinal digestion can provide more energy compared to ruminal fermentation, the extent of small intestinal digestion is limited compared to the capacity of the rumen to ferment starch (Owens et al., 1986; Huntington, 1997; Huntington et al., 2006). Digestion capacity in the small intestine is limited by the secretion of pancreatic amylase (Owens et al., 1986; Kreikemeier et al., 1991; Mayes and Orskov, 1974). Clary et al. (1969) and Van Hellen et al. (1978) found that pancreatic amylase secretions in steers increase with increasing concentrate in the diet and amylase secretion is low in newborn ruminants and increases with age (Siddons, 1968). Limits to digestion of starch in the small intestine might be explained by a rapid passage rate, which can result from feeding highly palatable diets with an increased degree of grain processing. An increase in processing increases digestibility in the rumen, therefore decreasing the amount of starch reaching the small intestine. In diets containing

72% corn, Galyean et al. (1979) observed an increase in ruminal digestion and a decrease in intestinal digestion, compared to feeding less concentrate in the diet. Because so many questions remain about the aspects and limitations of small intestinal starch digestion, nutritionists tend to focus on increasing ruminal starch fermentation in order to increase digestion and energy efficiency (Brake and Swanson, 2018). However, with more research, small intestinal digestion in cattle has the potential to significantly improve the energy efficiency of starch because more energy is produced when glucose is absorbed post-ruminally.

### ***Grain Processing***

Increasing the degree of processing improves digestibility of the grain by increasing the surface area for ruminal microbes to attach to in order to break down feed particles. The primary processing method used in receiving diets by feed yards in the U.S. is steam flaking (65.2%) followed by dry rolling (30.4%) (Samuelson et al., 2016). However, whole corn does not need to be processed for growing cattle to digest it as fully as processed corn (Gorocica-Buenfil and Loerch, 2005; Kaiser, 1999). Reinhardt et al. (1998) found no negative effects on rumen health or productivity when they fed whole corn to receiving and growing cattle. Younger cattle are able to efficiently break down the pericarp of corn kernels during mastication, although this becomes a more difficult task as cattle age and lose their teeth. Beauchemin et al. (1994) came to a similar conclusion, that processing corn may not be necessary to optimize digestion after observing that the majority of kernels were broken during the consumption and mastication of whole-shelled corn when it was fed to cows. In some cases, whole-shelled corn even outperformed dry-rolled corn (Chester-Jones et al., 1991). Owens et al. (1997) reported that

metabolizable energy value for whole corn grain is greater than for rolled corn grains, which might be explained by an increase in retention time.

### ***Syngenta Enogen® Feed Corn***

Syngenta is a global company that strives to advance sustainable agriculture through crop protection, seeds, seed treatments, and improved genetics, while working closely with small growers across the world. After being established in 2000, Syngenta soon launched their Enogen corn hybrids, which were designed specifically for ethanol production. These corn hybrids contain a bacterial transgene that produces an alpha-amylase enzyme in the grain, therefore eliminating the need to add liquid alpha-amylase. This enzyme is responsible for breaking down corn starch into fermentable sugar, which reduces the viscosity of the corn mash during the ethanol process. This new innovation provided the opportunity for ethanol plants to increase yield, production efficiency, and flexibility, while reducing production costs associated with the use of a liquid enzyme, natural gas, water, electricity, and maintenance chemicals (Urbanchuk et al., 2009). Ethanol from corn has been proven to reduce greenhouse gas emissions by up to 60%, water use by 7.7%, electricity use by 1.8%, and natural gas use by 8.9% (Urbanchuk et al., 2009). In order to reap the benefits of this alpha-amylase enzyme, only 15% of the total corn grind in ethanol plants needs to be Enogen grain (Syngenta, 2019a). The enzyme is activated by high temperatures, moisture, and acidic environments. Enogen corn allows growers to supply the enzyme to their local ethanol plants and earn a bushel premium. In 2018, bushel premiums for growers generated \$28.5 million (Syngenta, 2019b). In 2013, Syngenta announced Enogen Feed corn (EFC) for cattle (Griekspoor, 2018). The thermo-tolerant alpha-amylase enzyme, isolated from gene fragments of three different organisms, in the EFC grain improves the digestibility of

the corn by converting starch into fermentable sugars to provide energy for the animal. The enzyme is activated during salivation/mastication during consumption by the animal and fermentation in the rumen. With amylase thought to be the limiting factor in small intestinal starch digestion, an increased supply of amylase should enhance intestinal starch digestibility and absorption of glucose (Owens et al., 1986; Kreikemeier et al., 1991). Maximizing starch digestion in cattle is directly related to maximizing efficiency for producers.

In the field, EFC hybrids offer improved genetics and strong agronomic characteristics (Urbanchuk et al., 2009). Enogen corn can be used for grain or chopped for silage, unlike other hybrids designed specifically for silage use. Syngenta management recommendations for growing EFC include no-tillage operations with lower insecticide and nitrogen administration, yet an increase in herbicide, phosphorus pentoxide ( $P_2O_5$ ) and potassium chloride ( $K_2O$ ) (Urbanchuk et al., 2009). These recommended increases for herbicide and fertilizer are offset by other input reductions and adjusted agronomic practices. These hybrids are beneficial for producers that grow their own grain or silage. Commercial agreements are established through the Syngenta Stewardship program for growers in order to store EFC in separate bins/silos and plant buffer strips of control corn around EFC fields. These identity preservation measures prevent EFC from entering into the human food supply. Because of the rapid starch breakdown in the grain, the corn is not suitable for food processing and could have adverse effects on food quality and performance i.e. crumbly corn tortillas and corn chips.

### ***Feed By-Products***

In 2018, the U.S. ethanol industry generated 41.3 million metric tons (mmt) of corn distillers grains, corn gluten feed and corn gluten meal (RFA, 2019a). The increase in the production of ethanol is helping the U.S. to reduce foreign oil imports, lower fuel prices, and

lower greenhouse gas emissions (RFA, 2019b). Because of the rise in demand for ethanol production, corn prices have increased, which makes corn by-products an economically acceptable protein and energy source for beef cattle. Distillers grains by-products decrease the risk of digestive upsets because energy is in the form of digestible fiber and fat instead of highly fermentable starch. Wet distillers by-products have 97 to 147% the energy value of corn (Stock et al., 2000). After the starch is removed during the dry milling process, the remaining nutrients become more concentrated. Most ethanol in the U.S. is produced from dry milling due to cost and flexibility in type and quality of grain that can be used (Stock et al., 2000).

The dry milling process (Stock et al., 2000) begins with grinding grain into a meal and adding water to form a mash. Enzymes then break down the starch into fermentable sugars. Yeast is added and the sugars are converted to alcohol. After the fermentation process is complete, the mash is distilled, and feed particles are separated from the alcohol. The remaining feed slurry is called stillage. The coarse grain particles in the stillage are removed and sold as either wet distillers grains (WDG) or dried and sold as dried distillers grains (DDG). The remaining thin grain particles from the stillage are evaporated and produce condensed distillers solubles which can be dried and added to DDG to produce dried distillers grains with solubles (DDGS) or added to WDG to produce wet distillers grains with solubles (WDGS).

Crude protein (CP) content increases from 9% in the original corn grain to 27% in whole stillage (Stock et al., 2000). The majority of feed yards use corn by-products, mainly wet distillers grains (58%), as the main source of CP in receiving diets and some consulting nutritionists recommend levels as high as 18% CP to compensate for low feed intake (Samuelson et al., 2016). Oil is not removed in the dry-milling process, so distillers by-products are also higher than corn in fat content. Fiber, fat, S, and P are increased nearly 3 times compared to the

original levels in corn (Klopfenstein et al., 2008). High S levels exceeding the maximum tolerable level of 0.4% in cattle diets can be problematic for growing and finishing cattle (NRC, 2000; Sarturi et al., 2013). Larson et al. (1993) conducted research on yearling and calf finishing trials comparing diets with up to 40% wet distillers byproducts vs. cattle fed a 79% dry-rolled corn diet. Net energy increased 47% and 29% respectively, when wet distillers byproducts were fed to yearlings and calves at the 40% inclusion level and averaged 169% and 128% the value of corn. The energy value of distillers by-products and corn gluten feed is higher when fed in the wet form than when dried (Ham et al., 1994).

In the wet milling process (Stock et al., 2000), the corn grain is separated into its basic components of starch, protein, and fiber in a process called steeping. The isolated starch is fermented into ethanol in a process similar to dry milling or converted into fructose for corn syrups and corn sweeteners. Corn germ is then separated from the slurry and the oil is extracted to make corn germ meal. Bran and steep liquor remain. Water is removed from the bran and the steep liquor and distillers solubles are evaporated to produce corn gluten feed, which can be sold wet or dry.

### ***Corn Silage***

Over the past 40 years, silage acres have declined, but production has not due to corn hybrids and improvements in growing conditions. Since 2000, acres harvested for silage in the U.S. have averaged 6.3 million/year yielding 19.1 ton/acre (Warner, 2018). For producers that own or rent crop ground, feeding silage at 50% or more of the diet DM can be beneficial for backgrounding operations (Warner, 2018). Dependent on current feeder cattle and grain prices, farmer feeders can get more value from their crops by marketing feedstuffs through cattle. In doing so, producers can manage risk, add value to their crops, and diversify their operations

(Lawrence and Loy, 1999). Farmers that harvest their own corn silage have an economic freight advantage as opposed to other larger feed yards (Sprague, 2018). A survey based on the Midwest and Northern Plains regions of the United States found that over 90% of operations produced their own feed and the major crops grown were corn, corn silage, and alfalfa (Asem-Hiablie et al., 2016). The Northern Plains region reported larger feed yards and more backgrounded cattle than the Midwest region, however, they also reported a lower corn grain production, indicating that more corn is purchased by feedlots in the Northern Plains. Corn silage-based diets can be used successfully in receiving cattle (Fluharty and Loerch, 1996). The second most common roughage source used in receiving diets among U.S. feed yards is corn silage, with alfalfa hay being the primary roughage source (Samuelson et al., 2016). Silage is a high quality, energy dense feed that provides diet flexibility during drought seasons when hay is scarce or when the prices of grain and forage increase.

Stage of maturity and DM content are the two factors that help determine when to harvest silage. It is recommended to harvest silage with a chop length of 3/8 inch without a processor and 1/2 to 3/4 inch with a processor at 30-35% DM when the corn grain is at two-thirds milk-line (Johnson and Harrison, 2001; Saxe, 2007). If silage is harvested too wet, fermentation losses occur because butyric acid is produced instead of lactic acid. Butyric acid causes silage to have a foul odor with a slimy texture, which causes seepage, carrying soluble nutrients away while decreasing palatability (Bagg, 2012). DMI of young beef calves increased as silage DM content increased from 20 to 38% (Wilkinson et al., 1978). As the whole plant silage matures, DM, CP, neutral detergent fiber (NDF), and acid detergent fiber (ADF) increase, along with the plant yield and the proportion of grain in the plant (Wistuba, 1999). Essentially, as the whole corn plant increases in maturity, lignin content (indigestible fiber) increases, which decreases digestibility.

Unlike the grain portion of the plant, stover decreases in quality with advanced maturity (Russell, 1986; Hunt et al., 1989). An in-line kernel processor to damage the corn grain increases flexibility of the harvest window. Processing the whole plant increases the nutritive value and density of silage at all maturities (Johnson and Harrison, 2001; Wistuba, 1999; Young, 1998). At advanced maturities, processing the whole plant increases starch and fiber digestibility of the plant. Wistuba (1999) evaluated six silage diets comparing 50% milk-line, 80% milk-line, and 7 days post black layer either processed or unprocessed. Processing the whole plant and harvesting at 80% milk-line maximized DM yield and nutrient utilization. DM, OM, and starch digestibilities also increased due to the increased surface area from processing. Rojas-Bourrillon et al. (1987) and Miller et al. (1969) reported an increase in DM digestibility and DMI in corn silage diets when the silage was processed as well.

Silage fermentation occurs in 4 phases (Johnson and Harrison, 2001); first is the aerobic phase accompanied by a high pH. Then the fermentation phase begins when the microbial population starts to increase until all of the soluble sugars are consumed by the microbes, producing organic acids (lactate and acetate), which decreases the pH to a level that inhibits microbial proliferation. The use of homofermenter or heterofermenter inoculants may speed up the fermentation process and increase the shelf life of silage during feed-out (Saxe, 2007). In a study by Dalke et al. (1994), inoculating corn silage resulted in increased DM recovery, fermentation efficiency, and gain of the animals. The fermentation phase takes approximately 4-6 weeks. The third phase of silage fermentation has minimal biological activity and is referred to as the stable phase. The fourth and final phase is feed-out, which is the primary phase responsible for the loss of silage quality and DM due to mold and yeast growth. Limiting the time that silage is exposed to oxygen is a key component in decreasing the amount of losses that

occur. Other practices to help avoid losses are using proper harvesting and storage techniques, increasing the feed-out rate, and scraping the silage face downward (Johnson and Harrison, 2001).

When all harvest and storage costs are considered, bagging silage tends to be the least costly compared to bunker, pile, or upright storage systems (Holmes, 1998). However, according to a survey by Asem-Hiablie et al. (2016), most corn silage in feed yards was stored in covered bunkers or piles (44%) and only 15% used bagged silage. For bagged silage, DM losses can be as low as 4% compared to 12-15% in bunker silos. Density varies throughout the bag and is greatest at the bottom middle of the bag and least towards the top outside of the bag (Saxe et al., 2007). Recommendations for using a bagged silage system include having a well-drained site with a solid and flat foundation such as concrete or asphalt that is protected from wildlife. Bags should be inspected regularly for punctures. The main benefits of using bagged silage are having a smaller feed-out face with low labor requirements, longer chop length, improved speed of packing and handling, decreased cost, decreased DM losses, and having a high-quality feed stored in reserve.

## **Conclusions**

Management of newly arrived calves to receiving facilities can be a challenging task for producers in the beef industry. Many risks are involved based on the multitude of stressors placed on calves prior to and during the receiving period that compromise the immune system and decrease feed intakes upon arrival. This puts a lot of responsibility on the formulation of receiving diets to meet the high nutrient and energy requirements of these highly stressed and immunocompromised animals. With the increasing technological advances in the U.S. today,

companies like Syngenta and the ethanol industry are able to introduce products like Enogen Feed corn and distillers by-products to help improve the way producers feed cattle, significantly increasing feed efficiency and gains without having negative impacts on the health of the animal.

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# **Chapter 2 - Effects of Feeding Corn Containing an Alpha-Amylase Gene on the Performance and Digestibility of Newly Received Growing Steers**

## **Introduction**

Management of high risk and newly received cattle presents a great challenge to the beef industry. Feed intakes tend to be low upon arrival based on a multitude of stressors and decreased immunity from those stressors. Once on full-feed, it is important from a profitability perspective to achieve maximum feed efficiency not only during the growing phase, but in the subsequent feedlot phase as well. Diets should be formulated to meet the large nutrient and energy requirements of the animal and aid immune function.

Although the amylolytic activity of ruminal microbes is able to increase two-fold with the addition of grains to the diet, this increase is relatively minor in comparison to the use of exogenous amylases (Rojo et al., 2005). Therefore, the use of external amylase enzymes has the potential to increase the efficiency of starch digestion instead of trying to manipulate the activity of microbes in the rumen (Rojo et al., 2005). Data on feeding a corn hybrid containing an alpha-amylase enzyme to cattle is limited and previous research involving supplementing exogenous alpha-amylase in cattle diets has been extremely variable (Tricarico et al., 2007; Tricarico et al., 2005; Hristov et al., 2008; DeFrain et al., 2005). However, research conducted by Jolly-Breithaupt et al. (2018) has shown that feeding Enogen Feed Corn (EFC) to feedlot cattle has the potential to improve feed efficiency by 5.5%. The relative value of EFC as a source of energy either as a silage and/or grain for newly arrived and growing beef cattle is unknown.

## Materials and Methods

All procedures involving the use of animals were approved by the Kansas State University Institutional Animal Care and Use Committee.

### ***Experiment 1. Performance Study***

A total of 426 English crossbred steers (BW = 244 kg  $\pm$  90 kg) were purchased from Oklahoma, Texas, and Missouri and assembled at a farm in Lazbuddie, Texas and then shipped 909 kilometers to the Kansas State University Beef Stocker Unit on May 15, 2017. The steers were used in a completely randomized design with a 2 x 2 factorial arrangement of treatments to examine the effects of feeding two corn types (Enogen Feed Corn (EFC) vs. yellow #2 corn (CON)) with two methods of corn processing (dry-rolled (DRC) vs. whole-shelled (WC)) on the performance of stocker cattle in a 90-d receiving and growing study. EFC DRC and CON DRC were analyzed for particle size (Kansas State University Swine lab, Manhattan KS), which was 1633 microns and 1920 microns, respectively. The four treatment diets, EFC/DRC, EFC/WC, CON/DRC, and CON/WC were formulated to provide 1.13 Mcal NEg/kg DM. Diets contained 28.6% corn, 6.4% supplement, 17.5% alfalfa hay, 17.5% prairie hay, and 30% wet distillers grains on a DM basis (Golden Triangle Energy Cooperative, Craig, MO). Wet distillers grains was utilized as a protein source and to help limit the incidence of acidosis. Previous research has shown that replacing dietary corn with corn by-products that are high in fermentable fiber instead of starch can decrease the risk of acidosis in cattle and has the potential to increase animal performance (Corrigan et al., 2009; Bremer et al., 2011; Owens et al., 1997; Krehbiel et al., 1995). All diets were offered for *ad libitum* intakes (Table 2.1). EFC containing the alpha-amylase enzyme was provided by Syngenta Crop Protection, LLC (Greensboro, NC). All diets

had similar starch content. Upon arrival, calves were individually weighed using a hydraulic squeeze chute on load cells (Silencer, Moly Manufacturing Inc., Lorraine, KS) and given an individual visual identification ear tag and a radio-frequency identification (RFID) button tag. Thirty-two steers on the lower end of the weight spectrum and 10 steers on the higher end of the weight spectrum were removed from the research population. The remaining 384 steers were stratified by individual arrival weight and randomly assigned to 32 pens containing 12 steers. Each of the 32 pens was provided long-stem hay and *ad libitum* access to water via automatic waterers. Pens were then randomly assigned to one of four treatments, which equaled 8 pens/treatment. Pens were soil surfaced and of equal size (9.1 x 15.2 m) with concrete bunks measuring 9.1-m in length attached to a 3.6-m apron.

The morning after arrival (d 0), calves were weighed, ear tagged with a pen number, and vaccinated for viral and clostridial diseases. Vision 7 Somnus with Spur (Merck Animal Health, Omaha, NE) was used for protection against clostridial pathogens and Pyramid 5 + Presponse (Boehringer Ingelheim Vetmedica Inc., St. Joseph, MO), a modified-live vaccine protecting against infectious bovine rhinotracheitis (IBR), bovine viral diarrhea types 1 and 2 (BVDI-II), parainfluenza 3 (PI3), and bovine respiratory syncytial virus (BRSV), was used for protection against respiratory pathogens. The calves were also treated for internal parasites with Safe-Guard containing 10% Fenbendazole (Merck Animal Health, Madison, NJ). On day 21, all research animals were revaccinated for respiratory diseases with Bovishield Gold 5, a modified-live virus vaccine protecting against IBR, BVDI-II, BRSV, and PI3 (Zoetis, Parsippany, NJ).

Animals were fed their respective diets once daily at approximately 0700 using a Roto-Mix feed wagon (model 414-14B), which was thoroughly cleaned between each diet. Feed delivery was adjusted based on daily refusals to ensure *ad libitum* intakes without excess of

unconsumed feed. Individual animal weights were measured on d -1 (arrival), d 0 (initial processing), d 21 (revaccination), d 56/57 (fecal grab sampling) and d 91 (final weights). Fecal samples were obtained individually from steers in 16 pens d 56 and individually from steers in the remaining 16 pens d 57 and analyzed for starch and organic matter the same week (Table 2.8). Pen weights were collected on d 7, d 14, d 35, d 63, and d 77. Individual ingredient samples were collected weekly and composited for analysis and total mixed diet samples from each treatment were collected weekly and analyzed individually (moisture, DM, CP, ADF, NDF, calcium, phosphorus, potassium, magnesium, relative feed value (RFV), metabolizable energy (ME), digestible energy (DE), and starch) (Table 2.2) by a commercial laboratory (SDK Laboratories, Hutchinson, KS).

Animals were observed each day for signs of morbidity, such as depression, decreased appetite, and nasal or ocular discharge. Steers showing any of these signs were removed from the pen and herded to the treatment area. Once restrained in the chute, rectal temperature was measured and a clinical illness score (CIS) was assigned. CIS was assessed on a scale of 1 to 4: 1 = normal and healthy; 2 = slightly ill with mild depression/gauntness; 3 = moderately ill with severe depression/labored breathing/ocular or nasal discharge; and 4 = severely ill to the point of death with little response to human approach. Animals with a rectal temperature  $> 39.9^{\circ}\text{C}$  and a CIS  $> 1$  were treated. Treatment protocol was as follows: first treatment, Resflor Gold (300 mg/mL florfenicol and 16.5 mg/mL flunixin meglumine; Merck Animal Health, Madison, NJ); second treatment, Baytril 100 (100 mg/mL enrofloxacin; Bayer Animal Health, Shawnee Mission, KS); third treatment, Biomycin (200 mg/mL oxytetracycline; Boehringer Ingelheim Vetmedica, Inc., St. Joseph, MO). At the third treatment, animals were considered chronic and removed from the research population.

### ***Experiment 2. Intake and Digestibility Study***

Seven ruminally cannulated Holstein steers (BW = 198 kg  $\pm$  10kg) were used in an incomplete 4 x 4 Latin Rectangle design to determine diet digestibility and digestion characteristics. Data from one steer in the second period was removed due to issues with the rumen cannula. Experimental diets were the same as in Exp. 1 (Table 2.1). The study consisted of four consecutive 15-d periods consisting of a 10-d diet adaptation, 4-d fecal collection, and 1 d for ruminal fluid sampling. As the loads of feed were mixed daily for Exp. 1, the amount needed for Exp. 2 was removed from the beginning of each load and feed samples were analyzed independently from those in Exp. 1 (Table 2.2).

Animals were housed in individual outdoor pens (12.2 x 15.2 m). Each steer had *ad libitum* access to tank waterers, which were filled daily. Animals were fed once daily at approximately 1000 h. Diets were fed for *ad libitum* intake to target at least a 10% refusal. Total mixed ration (TMR) samples were collected on d 10 through 14 and composited for each period for analysis. TMR and weekly individual ingredient samples from Exp. 1 overlapping with the sampling week were sent to an independent laboratory (SDK Laboratories, Hutchinson, KS) for nutrient analysis (moisture, DM, CP, ADF, NDF, calcium, phosphorus, potassium, magnesium, RFV, ME, DE, and starch). On d 4 through 14, chromium oxide (Cr<sub>2</sub>O<sub>3</sub>) (10 g) was top-dressed and hand mixed into each animal's diet as a marker to calculate digestibility. Refusals were collected on d 11 through 15 and composited for each animal for each period. Fecal samples were also collected on d 11 through 14, taken from the rectum of the steers every 8 h with the sampling time increasing by 2 h each day so that every 2-h interval after feeding was represented. Fecal samples were stored frozen (-20°C) for later analysis. Refusal and fecal samples were composited for each steer in each period and sent to an independent commercial

lab (SDK Laboratories, Hutchinson, KS) for analysis (moisture, DM, CP, ADF, NDF, calcium, phosphorus, potassium, magnesium, RFV, ME, DE, and starch).

Refusal samples were dried at 55°C, air equilibrated, and ground through a 1-mm screen using a Wiley mill. Fecal samples were dried at 105°C and ground through a 1-mm screen using a Wiley mill. Fecal and refusal samples were weighed (0.5g) into 50-mL crucibles and ashed in a muffle oven at 600°C for 4 h. Chromium concentrations were determined by atomic absorption spectrophotometry according to the procedures of Williams et al. (1962).

On d 15 of each period, ruminal fluid samples were collected from 4 different locations in the rumen at 0, 2, 4, 6, 8, 12, 18, and 24 h after feeding and pooled within sampling time. Following the 0 h sampling, 3 g of Co-EDTA (0.4 g Co) dissolved in 200 mL of water was dosed into the rumen. Rumen samples were analyzed for pH with a transportable pH meter (Orion Model 230A (Beverly, MA)) and strained through 8 layers of cheesecloth. Strained rumen fluid was pipetted into four 2-mL micro-centrifuge tubes containing 0.25 mL of *m*-phosphoric acid and then frozen at -20°C for later analysis of VFA concentrations by gas liquid chromatography (GLC) and ammonia (Broderick and Kang, 1980). Additionally, 20 mL of strained rumen fluid was collected and frozen at -20°C for later analysis of Co concentration to determine liquid passage rate. Co concentrations were analyzed in the ruminal fluid and in the original dose by atomic absorption spectrophotometry. Liquid passage rate was determined by calculating the ruminal cobalt concentrations at 2 through 18 h after dosing Co-EDTA into the rumen and regressing the natural logarithm of Co against time for each steer in each period using the nonlinear procedure in SAS. Passage rate was based on the slope of the line, and the 0-h intercept was back converted from the log-transformed value and used to calculate ruminal liquid volume as dose divided by the Co concentration predicted at the time of dosing.

### ***Experiment 3. Performance Study***

A total of 362 crossbred steers of Tennessee origin (BW = 298 kg  $\pm$  75 kg), previously backgrounded for 63 days on a common diet at the Kansas State University Beef Stocker Unit, were used in a completely randomized 2 x 2 factorial design to determine the effects of feeding two varieties of corn silage (Enogen Feed Corn, E111F1-5122A-EZT0 (EFC) vs. Mycogen corn, TMF14L46 (CON)) and two varieties of DRC (EFC vs. yellow #2 corn (CON)) on the performance of stocker cattle in a 91-d growing study. EFC DRC and CON DRC were analyzed for particle size (Kansas State University Swine lab, Manhattan KS), which was 2628 microns and 3206 microns, respectively. There was a gut-fill equalization period of 14 days at the end of the trial (d77-d91), where all animals were limit-fed at 2.2% of BW daily a control diet containing 38% dry-rolled corn, 40% wet corn gluten feed (Sweet Bran; Cargill Animal Nutrition, Blair, NE), 8% supplement, and remainder of alfalfa and prairie hay on a DM basis (Table 2.5). The four treatment diets, EFC/EFC silage (EFC/ES), EFC/CON silage (EFC/CS), CON/EFC silage (CON/ES), and CON/CON silage (CON/CS), were formulated to contain 1.11 Mcal NE<sub>g</sub>/kg DM and contained 38.5% corn, 7.5% supplement, 7.5% alfalfa hay, 7.5% prairie hay, and 40% corn silage. All diets were offered for *ad libitum* intakes (Table 2.3). Twenty-five acres of dryland EFC silage (Enogen Feed Corn, E111F1-5122A-EZT0) and 6.5 acres of dryland CON silage (Mycogen, TMF14L46) was harvested in August of 2017 at 2/3 milk-line, chopped to a length of 20 mm, kernel processed to 2mm with an on-board processor, and bagged (SILOBOLSA Plastar Premium silage bags (Buenos Aires, Argentina)) the same day using a 550 horsepower Versa bagger (Astoria, OR). At harvest, EFC and CON silage averaged 34% and 29% DM, respectively. Each silage type was ensiled for approximately 147 days. CON silage and EFC silage yielded approximately 11 tons DM/acre and 9 tons DM/acre, respectively.

The ten heaviest steers were removed from the research population. The remaining 352 steers were stratified by weight and randomly assigned to pens composed of 11 animals. Pens were then randomly allocated to one of four treatments, which equaled 8 pens/treatment for a total of 32 pens. Pens used were the same as in Exp. 1. On d -6, calves were allocated to pens based on individual weight measured using a hydraulic squeeze chute with load cells (Silencer, Moly Manufacturing Inc., Lorraine, KS). On d 0, calves were individually weighed and tagged with a pen number. All calves were vaccinated for viral and clostridial diseases at the start of the previous 63-day backgrounding phase at the Kansas State University Beef Stocker Unit. Vision 7 Somnus with Spur (Merck Animal Health, Omaha, NE) was used for protection against clostridial pathogens and Bovishield Gold 5 (Zoetis, Parsippany, NJ), a modified-live vaccine protecting against infectious bovine rhinotracheitis (IBR), bovine viral diarrhea types 1 and 2 (BVDI-II), parainfluenza 3 (PI3), and bovine respiratory syncytial virus (BRSV), was used for protection against respiratory pathogens. Zuprevo 18% was used as a metaphylaxis on arrival (180 mg/mL Tildipirosin; Merck Animal Health, Madison, NJ) for the treatment of BRD associated with *Mannheimia haemolytica*, *Pasteurella multocida*, and *Histophilus somni*. The calves were also treated for internal parasites with Safe-Guard containing 10% Fenbendazole (Merck Animal Health, Madison, NJ) at the start of the previous 63-day backgrounding phase.

The steers were fed their respective diets once daily at approximately 0700 h using a Roto-Mix feed wagon (model 414-14B). Feed delivery was adjusted based on daily refusals to ensure *ad libitum* intakes without an excess of left-over feed. Individual ingredient samples were collected weekly and composited for analysis and total mixed ration samples from each diet were collected weekly and analyzed individually (moisture, DM, CP, ADF, NDF, calcium, phosphorus, potassium, magnesium, RFV, ME, DE, and starch) (Table 2.4) by a commercial

laboratory (SDK Laboratories, Hutchinson, KS). Individual animal weights were measured on d -6 (allocation), d 0 (initial processing), d 49 (fecal grab sampling), and d 91 (final weights). Fecal samples were obtained individually on day 49 and sent to a commercial laboratory (SDK Laboratories, Hutchinson, KS) to be analyzed for starch and organic matter the same week (Table 2.13). Pen weights were collected on d 14, d 28, d 42, d 56, d 70, d 77, and d 91. Animals were observed daily for morbidity and treated according to the same protocol from Exp. 1.

#### ***Experiment 4. Intake and Digestibility Study***

Eight ruminally cannulated, predominantly Angus, beef steers (BW = 211 kg  $\pm$  30 kg) were used in a 4 x 4 Latin rectangle design to determine diet digestibility and digestion characteristics. However, data from one steer was removed from the first period due to rumen cannula issues. Experimental diets were the same as in Exp. 3 (Table 2.3). The study consisted of four consecutive 15-d periods made up of a 10-d diet adaptation, 4-d fecal collection, and 1 d for ruminal fluid sampling. As loads of feed were mixed daily for Exp. 3, the amount needed for Exp. 4 was removed from the beginning of each load and feed samples were analyzed independently from those in Exp. 3.

Animals were housed in individual outdoor pens (6.1 x 15.2 m). Each steer had *ad libitum* access to tank waterers, which were filled daily. Steers were fed once daily at approximately 1000 h. Diets were fed for *ad libitum* intake to target at least a 10% refusal. Total mixed diet samples were collected on d 10 through 14 and composited for each period for analysis. Overlapping individual ingredient samples from Exp. 3 coinciding with the sampling week were sent to a commercial laboratory (SDK Laboratories, Hutchinson, KS) for nutrient analysis (moisture, DM, CP, ADF, NDF, calcium, phosphorus, potassium, magnesium, RFV, ME, DE, and starch) (Table 2.4). On d 4 through 14, Cr<sub>2</sub>O<sub>3</sub> (10 g) was top-dressed and hand

mixed into each animal's diet as a marker to calculate digestibility. Refusals and fecal samples were collected on d 11 through 14 and ruminal fluid samples were collected on d 15. Refusal, fecal, and ruminal fluid samples were collected and analyzed following the same procedures as Exp. 2.

### ***Statistical Analysis***

In Exp. 1, performance measures were analyzed using the MIXED procedure in SAS with the fixed effects of variety, processing, and variety  $\times$  processing. Fecal starch parameters and net energy calculations were analyzed using the GLIMMIX procedure in SAS (ver. 9.4; SAS inst. Inc., Cary, NC) with the fixed effects of variety, processing, and variety  $\times$  processing.

In Exp. 2, concentrations and proportions of VFA, ammonia, pH, and digestibility were analyzed in a linear mixed model fit in Proc GLIMMIX with fixed effects of variety, processing, sampling hour as well as their two- and three-way interactions. Period was included as a fixed effect, animal as a random effect, and sampling hour was modeled as a repeated measure with period  $\times$  animal as the subject. The covariance structure for the repeated measures was selected from first order ante-dependent, compound symmetry, heterogeneous compound symmetry, unstructured, Toeplitz, and heterogeneous Toeplitz based on AIC values for each response variable.

In Exp. 3, performance measures and net energy calculations were analyzed using the MIXED procedure in SAS with the fixed effects of corn, silage, and corn  $\times$  silage. Fecal starch parameters were analyzed using the GLIMMIX procedure in SAS with the fixed effects of corn, silage, and corn  $\times$  silage. Silage DM and total starch was also analyzed using the GLIMMIX procedure in SAS with the fixed effect of silage and date of sampling as a block.

In Exp. 4, ruminal parameters and digestibility were analyzed in a linear mixed model fit in Proc GLIMMIX with fixed effects of corn, silage, sampling hour as well as their two- and three-way interactions. Period was included as a fixed effect, animal as a random effect, and sampling hour was modeled as a repeated measure with period  $\times$  animal as the subject. The covariance structure for the repeated measures was selected from first order ante-dependent compound symmetry, heterogeneous compound symmetry, unstructured, Toeplitz, and heterogeneous Toeplitz based on AIC values for each response variable.

## **Results and Discussion**

### ***Experiment 1. Performance Study***

Little morbidity and no mortality were observed in this experiment (Table 2.6). One animal was treated for foot rot, 1 for bloat, and 1 for pinkeye; all animals recovered. Two steers were treated for chronic respiratory illness and removed from the research population as was one steer treated for lameness.

Performance results from Exp. 1 are presented in Table 2.7. Over the entire 91-d trial, DMI of calves fed EFC tended to be lower ( $P < 0.09$ ) than calves fed CON. This difference was especially apparent through d 14, where CON-fed calves consumed significantly more feed than their EFC-fed counterparts ( $P < 0.01$ ). Average daily gain (ADG) and off-test weights tended to be greater ( $P < 0.10$ ) for calves fed EFC over the entire 91-d trial. Gain:feed (G:F) was greater in calves fed EFC ( $P < 0.01$ ). As early as d 35, feed:gain (F:G) and G:F tended to be better for EFC-fed steers than for CON-fed steers ( $P < 0.07$ ). For the remainder of the study (d 63 and d 91), feed efficiency was significantly better for calves fed EFC ( $P < 0.01$ ). The efficiency of feed conversion (F:G) of calves receiving EFC was improved by 5.5%. This data agrees with results

published by Jolly-Breithaupt et al. (2018), which showed a 5.7% increase in feed efficiency when feeding EFC DRC containing an alpha-amylase enzyme. However, another experiment by Jolly-Breithaupt et al. (2018) revealed no differences in performance when cattle were fed EFC as DRC, which they concluded could be a result of specific growing conditions of the corn hybrid. Other researchers found no differences in DMI, ADG, or feed efficiency when feeding a ground corn hybrid (CA3272) containing an alpha-amylase enzyme when included at 0, 10, or 20% of the diet DM (Schoonmaker et al., 2014). These researchers hypothesized that no differences were observed because the control diet allowed adequate capacity to hydrolyze starch due to the extensive level of corn processing and that the alpha-amylase enzyme may show better results in whole corn.

There were no effects of corn processing observed for final DMI ( $P < 0.57$ ) or final body weight ( $P < 0.24$ ). However, final ADG ( $P < 0.14$ ), F:G ( $P < 0.10$ ), and G:F ( $P < 0.13$ ) tended to be better for DRC than for WC, but were not a significant factor in improving the performance of the cattle. This supports research done by Siverson et al. (2014) who fed similar diets and found no significant differences in performance between DRC and WC when included at 29% of the diet DM in addition to 30% wet corn gluten feed (WCGF). When considering EFC/DRC vs. CON/DRC, there was a 2.2% improvement in F:G and a 2.7% improvement in G:F for EFC/DRC vs. CON/DRC. Jolly-Breithaupt et al. (2018) found similar results when comparing EFC/DRC to CON/DRC, which was a 2.2% increase in G:F for Enogen corn compared to control corn.

Overall, feed efficiency tended to be best for the EFC/WC treatment diet ( $P < 0.09$ ). Calves fed EFC/WC did not significantly differ from those fed EFC/DRC, but did however outperform calves on the CON/DRC diet ( $P < 0.09$ ). When considering numerical differences

between EFC/WC and EFC/DRC, the WC led to 2.2% better F:G and 2.4% better G:F. These results suggest that feeding EFC/WC results in equal or improved performance in growing calves as opposed to feeding DRC of either variety. This suggests that mastication by the animal is sufficient to break down WC kernels with the alpha-amylase trait and that processing EFC may not be necessary to optimize digestion. Research by Beauchemin et al. (1994) supports this inference after observing that the majority of corn kernels were broken during the consumption and mastication of WC when it was fed to cows. When considering health, cost, and performance of the animal, the least amount of processing is best (Orskov, 1986).

The fecal starch analyses from d 56/57 (Table 2.8) show a processing effect with WC leading to a greater fecal starch concentration than DRC ( $P < 0.01$ ), meaning less starch was digested and utilized by the animal when WC was fed. The EFC treatments led to lower starch concentrations in the feces than did CON ( $P < 0.01$ ), showing a better starch digestibility for EFC. Fecal starch is a good indicator of starch digestion in cattle (Fredin et al., 2014 and Zinn et al., 2007).

Net energy values, (calculations from Galyean (2019) based on NRC (1996) requirements), (Table 2.7) demonstrated significant differences between corn varieties. Calves fed the EFC treatments had greater NEm (net energy of maintenance) and NEg (net energy of gain) concentrations than the CON treatments ( $P < 0.01$ ). Net energy concentrations tended to differ among treatments as well. EFC/WC and EFC/DRC diets had similar net energy values, followed by CON/DRC, and CON/WC had the lowest NEm and NEg values ( $P < 0.13$ ). These net energy values were lower than what was originally formulated in the diets, which could reflect a number of factors that might have affected animal performance independent of the true dietary energy density.

In conclusion, the efficiency of feed conversion (F:G) of calves receiving EFC was improved by 5.50% compared to calves receiving CON corn. This response became apparent by day 35 and was a significant factor throughout the remainder of the study. There were no negative observations regarding the health or behavior of the calves when feeding EFC. By using a variety of corn that is more energy dense and requires less processing, producers can potentially produce gain with greater economic efficiency.

### ***Experiment 2. Intake and Digestibility Study***

The results from this intake and digestibility trial supports the effects of treatments on growth efficiency in Experiment 1. Digestion and ruminal fermentation results are presented in Tables 2.9 and 2.10. There was no effect for corn processing on digestion and ruminal parameters other than a small tendency for pH to be lower for DRC than for WC ( $P < 0.15$ ). This agrees with Exp. 1 and reiterates the results determined by Siverson et al. (2014) and Beauchemin et al. (1994), where no differences in digestibility were found between DRC and WC. Ruminal pH ( $P > 0.82$ ) and ruminal ammonia concentration ( $P > 0.30$ ) were not affected by corn variety (Table 2.9). Ruminal ammonia and pH measurements over time after feeding can be seen in Figures 2.2 and 2.1, respectively. There was a treatment x time interaction for both ruminal pH and ruminal ammonia, which is to be expected based on eating behavior of the animals. As the steer consumes the specified treatment diet and fermentation begins, ammonia is produced and the pH declines in the rumen. This resulted in an increase in ammonia and a decrease in pH in the rumen after feeding. Liquid passage rate was faster for CON-fed calves than for EFC-fed calves ( $P < 0.01$ ), which might in part explain the tendency for DMI to be

highest for CON/DRC ( $P < 0.11$ ), and also supports the greater DMI for calves fed the CON treatments than for the EFC treatments in Exp. 1.

Passage rate can be inversely related to digestibility because faster passage allows less time for ruminal digestion. Passage rate was greater for calves fed CON diets than for those fed EFC diets, whereas digestibility was less for EFC than for CON. Total tract organic matter (OM) and dry matter (DM) digestibility were greater for EFC-fed calves than for CON-fed calves ( $P < 0.04$ ), representing an 8% and 9% increase, respectively. More energy should be available to the animal when digestibility increases, so the differences in digestibility between EFC and CON may explain differences in feed efficiency observed in Exp. 1. In agreement with this study, Jolly-Breithaupt et al. (2018) observed an increase in total tract OM and DM digestibility with the feeding of EFC ( $P < 0.08$ ), but they also observed greater total tract starch digestibility with EFC ( $P < 0.01$ ), which we did not observe. Rojo et al., (2005) supplemented alpha-amylase from *Bacillus licheniformis* to lambs, observing an increase in total tract DM, OM, and starch digestion. Although there were no significant effects for starch digestibility in our study, numerical differences resulted in a 5.9% greater starch digestion for the EFC/WC treatment when compared to the other 3 treatments.

VFA concentrations are shown in Tables 2.9 and 2.10 and Figure 2.3. There were no effects of corn processing ( $P > 0.28$ ), corn source ( $P > 0.28$ ), or interactions ( $P > 0.29$ ) for ruminal concentrations of propionate, butyrate, isobutyrate, valerate, or isovalerate. Ruminal acetate concentration was lower in EFC-fed calves than in CON-fed calves ( $P < 0.05$ ), and this effect was more pronounced in calves fed the DRC than for those fed WC ( $P < 0.06$ ). Previous research involving supplementing exogenous alpha-amylase in cattle diets has been extremely variable. Researchers have either discovered an increase in acetate (Tricarico et al., 2005; Rojo et

al., 2005), an increase in propionate (Vander Pol et al., 2009), or found no effects on ruminal VFA concentrations in ruminant animals supplemented with alpha-amylase (Jolly-Breithaupt et al., 2018 and Hristov et al., 2008).

As a percent of total VFA, valerate tended to increase for EFC treatments ( $P < 0.10$ ), being the greatest for EFC/DRC ( $P < 0.09$ ). Isobutyrate percent tended to be higher for the EFC/DRC treatment as well ( $P < 0.07$ ). When analyzing the *in vitro* VFA profile, Horton et al. (2018) also observed an increase in percent valerate for blends of EFC ( $P < 0.06$ ). A corn  $\times$  processing interaction revealed a tendency for total VFA to be greatest for CON/DRC and EFC/WC ( $P < 0.14$ ), suggesting that processing corn is not necessary to achieve greater VFA concentrations.

### ***Experiment 3. Performance Study***

Little morbidity and no mortality was observed for this experiment (Table 2.11). One animal was treated for respiratory illness and 2 were treated for bloat; all animals recovered. Two steers were also treated for lameness and removed from the experiment.

Performance results from Exp. 3 are shown in Table 2.12. No significant effects of DRC grain type were noted for the overall 91-d feeding trial, nor were any significant interactions between corn silage type and DRC grain type observed. Starting as early as d 42 and continuing through d 70, ADG tended to be greater for EFC silage ( $P < 0.07$ ) than for the CON silage, and this was significant throughout at d 77 and 91 ( $P < 0.01$ ). DMI tended to be greater ( $P < 0.08$ ) for calves fed EFC silage over the entire 91-d trial. This difference was significant at d 56 ( $P = 0.03$ ). Lara et al. (2018b) researched the effects of feeding corn silage diets with or without an amylolytic enzyme supplemented to lambs. They found that providing 602 dextrinizing units of alpha-amylase/kg DM in the TMR had no effect on DMI ( $P = 0.90$ ) or ADG ( $P = 0.15$ ), and,

although not significant, feed efficiency was improved by 4.8% for lambs fed corn silage with an alpha-amylase supplement. Feed efficiency (G:F and F:G) over the full 91-d study tended to be better in calves fed EFC silage ( $P = 0.14$ ). There was also a tendency for final body weight to be greater for EFC silage ( $P = 0.10$ ). In agreement with the performance results in our experiment, Leahy et al. (1990) observed an increase in ADG ( $P < 0.01$ ), feed efficiency ( $P < 0.01$ ), and final body weight ( $P < 0.05$ ) in beef heifers when fed corn silage treated with alpha-amylase at 0.05% (wet basis) before ensiling, resulting in 11% increases in performance in ADG and G:F for heifers fed the alpha-amylase treatment.

Results from the d 49 fecal sampling showed no effects of corn source or silage source on fecal starch concentration (Table 2.13). Starch concentration of the EFC silage was 6.0 percentage units greater than the CON silage. Additionally, the EFC silage had a greater DM concentration than the CON silage (34 vs. 30%;  $P < 0.01$ ), which may have played a role in the performance differences between silages. Net energy calculations based on growth performance (calculations from Galyean (2019) based on NRC (1996) requirements), did not differ among treatments (Table 2.12). The NE concentrations calculated from performance were less than originally formulated in the diets, which could simply be due to the inefficiency of humans to be able to accurately predict energy values in cattle. These net energy values were lower than what was originally formulated in the diets, which could reflect a number of factors that might have affected animal performance independent of the true dietary energy density. The purpose of a gut-fill equilibration period is to reduce variability. The two-week diet adaptation period at the conclusion of the study apparently narrowed the differences in gain between the CON silage-fed calves.

Overall, feed efficiency of calves receiving EFC silage was improved by 3.30% and average daily gain improved by 6.00% compared to calves receiving CON silage. No significant effects of corn grain type were noted over the entire 91-d trial, nor any overall significant interactions between corn silage type and corn grain type. There were no negative observations regarding cattle health or behavior with the feeding of EFC silage.

#### ***Experiment 4. Intake and Digestibility Study***

The results from this intake and digestibility trial help to explain the results from Experiment 3. These results are shown in Tables 2.15 and 2.16. CON silage had a significantly higher DMI than EFC silage ( $P < 0.01$ ), which is notably opposite of the effect observed in the corresponding performance study (Exp. 3). There were no effects of corn, silage, or corn  $\times$  silage on liquid passage rate ( $P > 0.20$ ), ruminal pH ( $P > 0.23$ ), or digestibility of DM, OM, NDF, ADF, or starch ( $P > 0.24$ ). Ruminal pH at times after feeding are presented in Figure 2.4. There was a treatment  $\times$  time interaction, which is to be expected based on eating behavior of the animals. As the steer consumes the specified treatment diet, fermentation begins and the pH will decrease accordingly. CON corn tended to lead to higher ammonia concentrations than did EFC ( $P < 0.06$ ). This is indicative of fermentation in the rumen. Ruminal ammonia measured over time post-feeding is presented in Figure 2.5. There was a treatment  $\times$  time interaction, which was expected based on eating behavior of the animals. As the steer consumes the specified treatment diet, fermentation begins and ammonia is produced in the rumen, which resulted in higher concentrations of ammonia after feeding.

VFA concentrations are presented in Tables 2.15 and 2.16 and Figure 2.6. Ruminal concentrations (mM) of acetate, isobutyrate, and valerate were not affected by corn, silage, or by an interaction of the two ( $P > 0.35$ ). In contrast to the results observed in Exp. 2, ruminal

propionate concentrations (mM) were greater for EFC silage treatments ( $P < 0.01$ ). Lara et al. (2018a) researched the effects of feeding corn silage diets with or without an amylolytic enzyme supply to cannulated wethers. When providing 602 dextrinizing units of alpha-amylase/kg DM in the TMR, molar proportions of propionic acid increased ( $P < 0.01$ ). Effects of silage source tended to be present for ruminal butyrate concentration (mM) ( $P < 0.10$ ) and total VFA concentration ( $P < 0.13$ ), with greater concentrations observed for the EFC silage. Horton et al. (2018) observed similar results with ensiled high-moisture EFC. In their study, *in vitro* fermentations with ruminal microbes led to greater production of butyrate and total VFA for the high-moisture EFC than for high-moisture CON corn ( $P < 0.05$ ). DRC source tended to have an effect on ruminal propionate concentration (mM), which was greater for CON DRC compared to EFC DRC ( $P < 0.09$ ).

Molar % propionate was greater for EFC silage treatments ( $P < 0.01$ ) and CON silage treatments had a greater % acetate ( $P < 0.01$ ). DRC source had an effect on molar % acetate ( $P < 0.03$ ), which was greater for EFC, and molar % propionate ( $P < 0.03$ ), which was greater for CON. Acetate molar % and propionate molar % differed significantly among treatments ( $P < 0.01$ ). Calves fed CON/ES had the highest molar % of propionate and calves fed CON/CS had the least, and the opposite was true for molar % acetate. These results agree with the findings of Exp. 2, where ruminal acetate concentrations were greater for EFC treatments. CON silage treatments had a tendency for a higher molar % isovalerate ( $P < 0.11$ ). Calves fed the CON/CS treatment had the highest molar % isovalerate and calves fed CON/ES had the least amount ( $P < 0.01$ ).

Numerical differences showed a 2.5% increase in total tract DM digestibility and a 2.2% increase in total tract OM digestibility for EFC silage, which helps to explain the increased

performance of calves fed EFC silage in Exp. 3. Jolly-Breithaupt (2018) compared feeding EFC as DRC vs. CON DRC with either WCGF or modified distillers grains (MDGS) and 15% CON corn silage included in all diets. They found no interactions among treatments between DM, OM, or starch digestibility. Conversely, Lara et al. (2018a) observed an increase in apparent OM and DM digestibility by wethers when corn silage was supplemented with an alpha-amylase enzyme in the diet.

### **Implications**

There were no negative observations regarding the health or behavior of the calves when feeding EFC or EFC silage. Relative to CON, there were significant advantages in feed efficiency when feeding EFC as grain or silage. Under our circumstances, cattle fed EFC/WC had either equal or improved performance as opposed to feeding EFC/DRC or CON/DRC. Because younger cattle are able to successfully masticate whole corn, feeding EFC/WC has the potential to be beneficial to the stocker/grower sector of the beef industry by eliminating processing costs without sacrificing performance or digestibility. Digestibility of the corn grain was increased with the addition of the alpha-amylase enzyme present in the Enogen Feed Corn. Overall, the results of these studies indicate that using a hybrid of corn containing an alpha-amylase enzyme generally improved feed efficiency in growing calves.

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**Table 2.1 Diet Composition (Exp. 1 and 2)**

Ingredient <sup>1</sup>	% of DM
Corn (variety x processing) <sup>2</sup>	28.57
Wet Distillers Grains	30.00
Alfalfa Hay	17.50
Prairie Hay	17.50
Supplement <sup>3</sup>	6.43

<sup>1</sup>Diets were formulated to contain 1.74 Mcal NEm/kg DM and 1.13 Mcal NEg/kg DM

<sup>2</sup>Corn Type: EFC (Enogen Feed Corn) vs. CON (Yellow #2 corn) and fed as either WC (whole corn) or DRC (dry-rolled corn)

<sup>3</sup>Supplement pellet was formulated to contain (DM basis) 11.09% crude protein, 8.50% calcium, 0.42% phosphorus, 5.50% salt, 0.80% potassium, 0.57% magnesium, 1.70% fat, 11.04% acid detergent fiber, and 331 mg/kg lasalocid (Bovatec; Zoetis, Parsippany, NJ).

**Table 2.2 Diet Nutrient Analysis (Exp. 1 and 2)**

Nutrient, % of DM	Corn Source				
	CON <sup>1</sup>		EFC <sup>2</sup>		
	Corn Processing				
	DRC <sup>3</sup>	WC <sup>4</sup>	DRC <sup>3</sup>	WC <sup>4</sup>	
Exp. 1					
DM, %	57.9	57.6	54.3	53.2	
CP	17.4	17.3	18.2	18.7	
NDF	27.4	29.1	30.1	30.6	
ADF	17.3	19.1	17.5	19.1	
Starch	25.5	26.6	23.0	25.8	
Ca	1.16	1.18	1.08	1.19	
P	0.42	0.45	0.42	0.47	
Exp. 2					
DM, %	59.9	59.1	56.7	58.0	
CP	17.3	17.9	18.4	18.5	
NDF	29.0	28.1	30.0	29.4	
ADF	16.6	15.7	16.8	17.2	
Starch	25.5	26.6	23.0	25.8	
Ca	1.16	1.17	1.14	1.15	
P	0.42	0.43	0.45	0.44	

<sup>1</sup>Yellow #2 corn<sup>2</sup>Enogen Feed Corn<sup>3</sup>Dry-rolled corn<sup>4</sup>Whole-shelled corn

**Table 2.3 Diet Composition (Exp. 3 and 4)**

Ingredient <sup>1</sup>	% of DM
Corn <sup>2</sup>	38.50
Corn silage <sup>3</sup>	40.00
Alfalfa Hay	7.00
Prairie Hay	7.00
Supplement <sup>4</sup>	7.50

<sup>1</sup>Diets formulated to contain 1.72 Mcal NEm/kg DM and 1.11 Mcal NEg/kg DM.

<sup>2</sup>Dry-rolled Corn Type: EFC (Enogen Feed Corn) vs. CON (Yellow #2 corn)

<sup>3</sup>Corn Silage: EFC (Enogen Feed Corn) vs. CON (Mycogen corn)

<sup>4</sup>Supplement pellet was formulated to contain (DM basis) 8.80% crude protein, 5.68% calcium, 1.00% phosphorus, 3.78% salt, 1.89% potassium, 0.47% magnesium, 3.08% fat, 11.9% acid detergent fiber, and 231 mg/kg monensin (Rumensin; Elanco, Greenfield, IN).

**Table 2.4 Diet Nutrient Analysis (Exp. 3 and 4)**

Item	Corn Silage Source			
	CON <sup>1</sup>		EFC <sup>2</sup>	
	Dry-rolled Corn Source			
	CON <sup>3</sup>	EFC <sup>2</sup>	CON <sup>3</sup>	EFC <sup>2</sup>
Composition, % of DM (Exp. 3)				
DM, %	50.9	51.3	54.6	54.3
CP	13.4	13.4	13.0	13.2
NDF	27.9	27.9	25.8	28.1
ADF	19.0	18.7	17.2	18.0
Starch	35.8	36.2	39.3	37.1
Ca	0.88	0.90	0.81	0.86
P	0.31	0.31	0.28	0.28
Composition, % of DM (Exp. 4)				
DM, %	52.9	51.2	54.7	54.7
CP	12.9	12.9	12.9	12.8
NDF	28.1	29.1	27.8	27.6
ADF	19.0	19.2	18.2	18.1
Starch	39.1	38.5	39.1	39.3
Ca	0.80	0.79	0.80	0.79
P	0.29	0.29	0.28	0.28

<sup>1</sup>Mycogen corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Yellow #2 corn

**Table 2.5 Gut-Fill Equalization Diet Composition and Nutrient Analysis**

Ingredient	% of DM
Corn <sup>1</sup>	38.82
Sweet Bran <sup>2</sup>	40.00
Alfalfa	6.50
Prairie Hay	6.50
Supplement <sup>3</sup>	8.18
Composition, % of DM	
DM, %	70.85
CP	16.18
NDF	24.99
ADF	12.01
Ca	0.31
P	0.66

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<sup>1</sup>Dry-rolled yellow #2 corn

<sup>2</sup>Cargill Animal Nutrition, Blair, NE

<sup>3</sup>Supplement pellet was formulated to contain (DM basis) 8.80% crude protein, 5.68% calcium, 1.00% phosphorus, 3.78% salt, 1.89% potassium, 0.47% magnesium, 3.08% fat, 11.9% acid detergent fiber, and 231 mg/kg monensin (Rumensin; Elanco, Greenfield, IN).

**Table 2.6 Effects of Enogen Feed Corn and corn processing on health (Exp. 1)**

Disease Diagnosis	Treatment Diet <sup>1</sup>
Chronic bovine respiratory disease	CON/WC
Chronic bovine respiratory disease	EFC/WC
Foot rot	CON/WC
Bloat	EFC/WC
Pinkeye	EFC/DRC
Lameness	CON/DRC

<sup>1</sup>CON/WC: Yellow #2 corn/whole-corn,  
EFC/WC: Enogen Feed Corn/whole-corn,  
CON/DRC: Yellow #2 corn/dry-rolled corn,  
EFC/DRC: Enogen Feed Corn/dry-rolled corn

**Table 2.7 Effect of Enogen Feed Corn and corn processing on performance (Exp. 1)**

Item	Corn Source				SEM	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Process	Source	Process x Source
	Corn Processing							
	DRC <sup>3</sup>	WC <sup>4</sup>	DRC <sup>3</sup>	WC <sup>4</sup>				
No. of pens	8	8	8	8				
No. of animals	95	95	96	93				
Body weight, kg								
d 0	244	245	244	245	1.08	0.33	0.77	0.59
d 7	258	259	259	259	1.91	0.70	0.80	0.85
d 14	277	274	275	275	1.96	0.14	0.49	0.14
d 35	307	305	307	306	2.53	0.17	0.32	0.54
d 63	344	341	348	344	3.99	0.09	0.07	0.64
d 77	360	360	367	364	6.89	0.62	0.10	0.68
d 91	385	380	386	386	4.29	0.24	0.10	0.34
ADG, kg/d								
d 0-7	2.08	2.04	2.07	2.07	0.28	0.87	0.93	0.91
d 0-14	2.37	2.12	2.20	2.18	0.15	0.06	0.41	0.09
d 0-35	1.80	1.71	1.80	1.77	0.07	0.07	0.37	0.38
d 0-63	1.58	1.53	1.64	1.58	0.06	0.05	0.09	0.75
d 0-77	1.51	1.49	1.59	1.55	0.09	0.52	0.11	0.75
d 0-91	1.55	1.49	1.56	1.55	0.04	0.14	0.09	0.25
Average DMI, kg/d								
d 0-7	6.62	6.57	6.45	6.24	0.25	0.31	0.03	0.55
d 0-14	7.71	7.61	7.47	7.30	0.19	0.13	0.01	0.72

d 0-35	8.58	8.54	8.56	8.12	0.30	0.09	0.11	0.14
d 0-63	9.08	9.20	9.02	8.78	0.35	0.72	0.15	0.27
d 0-77	9.13	9.34	9.03	8.90	0.37	0.83	0.11	0.32
d 0-91	9.44	9.69	9.30	9.24	0.37	0.57	0.09	0.37
F:G, kg/kg								
d 0-7	3.26	3.28	3.18	3.08	0.19	0.82	0.46	0.76
d 0-14	3.28	3.61	3.42	3.37	0.10	0.20	0.66	0.07
d 0-35	4.78	4.99	4.78	4.60	0.11	0.86	0.08	0.08
d 0-63	5.74	6.00	5.50	5.60	0.12	0.14	0.01	0.48
d 0-77	6.06	6.26	5.70	5.84	0.19	0.38	0.05	0.84
d 0-91	6.10	6.49	5.97	5.97	0.11	0.10	0.01	0.09
G:F, kg/kg								
d 0-7	0.314	0.311	0.325	0.332	0.020	0.92	0.44	0.81
d 0-14	0.307	0.279	0.294	0.299	0.009	0.19	0.68	0.07
d 0-35	0.210	0.201	0.211	0.218	0.005	0.81	0.07	0.10
d 0-63	0.175	0.167	0.182	0.180	0.003	0.15	0.01	0.51
d 0-77	0.165	0.160	0.176	0.174	0.005	0.48	0.02	0.76
d 0-91	0.164	0.154	0.168	0.168	0.003	0.13	0.01	0.11
NEm, Mcal/kg <sup>5</sup>	1.51	1.45	1.54	1.54	0.02	0.19	0.01	0.13
NEg, Mcal/kg <sup>5</sup>	0.91	0.86	0.94	0.94	0.02	0.19	0.01	0.13

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<sup>1</sup>Yellow #2 corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Dry-rolled corn

<sup>4</sup>Whole-shelled corn

<sup>5</sup>Net energy calculations from Galylean (2019) based on NRC (1996) requirements.

**Table 2.8 Fecal Analysis (Exp. 1)**

Item	Corn Source				SEM	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Process	Source	Process x Source
	Corn Processing							
	DRC <sup>3</sup>	WC <sup>4</sup>	DRC <sup>3</sup>	WC <sup>4</sup>				
DM, %	18.84	19.92	16.74	17.92	0.39	0.01	<0.01	0.90
Starch, % of DM	11.98	21.84	6.09	13.78	1.41	<0.01	<0.01	0.45

<sup>1</sup>Yellow #2 corn<sup>2</sup>Enogen Feed Corn<sup>3</sup>Dry-rolled corn<sup>4</sup>Whole-shelled corn

**Table 2.9 Effects of Enogen Feed Corn and processing on digestibility and ruminal characteristics (Exp. 2)**

Item	Corn Source				SEM <sup>5</sup>	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Process	Source	Process x Source
	Corn Processing							
	DRC <sup>3</sup>	WC <sup>4</sup>	DRC <sup>3</sup>	WC <sup>4</sup>				
Number of observations	7	7	6	7				
DMI, kg/d	8.21	7.68	7.75	8.14	0.43	0.80	0.99	0.11
Ruminal								
pH <sup>6</sup>	5.81	5.93	5.84	5.87	0.06	0.15	0.82	0.37
Ammonia, mM <sup>6</sup>	2.79	2.38	3.63	2.80	0.73	0.32	0.30	0.73
Total VFA, mM <sup>6</sup>	109.4	107.0	102.1	109.5	5.27	0.45	0.45	0.14
Acetate, mM <sup>6</sup>	66.6	65.9	60.6	65.8	3.01	0.16	0.05	0.06
Propionate, mM <sup>6</sup>	28.1	26.7	26.3	28.9	2.68	0.73	0.90	0.29
Butyrate, mM <sup>6</sup>	10.6	10.2	10.9	10.4	0.72	0.28	0.52	0.86
Isobutyrate, mM <sup>6</sup>	1.31	1.47	1.40	1.41	0.12	0.29	0.82	0.38
Valerate, mM <sup>6</sup>	1.57	1.59	1.77	1.64	0.15	0.64	0.28	0.48
Isovalerate, mM <sup>6</sup>	1.19	1.21	1.15	1.17	0.14	0.82	0.68	0.97
Liquid passage rate, %/h <sup>7</sup>	9.52	8.84	7.43	8.38	0.67	0.77	0.01	0.09
Digestibility, % (total tract)								
DM	58.41	56.21	62.05	63.17	2.53	0.83	0.04	0.50
OM	61.51	59.28	64.84	66.02	2.45	0.82	0.04	0.46
NDF	51.14	46.96	51.52	53.51	4.18	0.79	0.41	0.46
ADF	48.49	42.05	50.01	54.52	5.20	0.85	0.17	0.28
Starch	84.66	85.14	86.43	90.43	2.90	0.37	0.16	0.47

<sup>1</sup>Yellow #2 corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Dry-rolled corn

<sup>4</sup>Whole-shelled corn

<sup>5</sup>Largest value of treatments reported

<sup>6</sup>Average of values collected at 0, 2, 4, 6, 8, 12, 18, and 24 h after feeding.

<sup>7</sup>Calculated values from samples collected at 2, 4, 6, 8, 12, and 18 after feeding.

**Table 2.10 Effects of Enogen Feed Corn and processing on ruminal VFA profile (Exp. 2)**

Item	Corn Source				SEM <sup>5</sup>	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Process	Source	Process x Source
	Corn Processing							
	DRC <sup>3</sup>	WC <sup>4</sup>	DRC <sup>3</sup>	WC <sup>4</sup>				
Number of observations	7	7	6	7				
Ruminal VFA, % of total								
Acetate <sup>6</sup>	62.0	61.4	60.8	60.3	1.33	0.68	0.34	0.95
Propionate <sup>6</sup>	24.5	25.1	24.4	26.4	1.38	0.33	0.65	0.60
Butyrate <sup>6</sup>	9.66	9.51	10.6	9.50	0.60	0.24	0.41	0.36
Isobutyrate <sup>6</sup>	1.22	1.38	1.40	1.30	0.07	0.62	0.47	0.07
Valerate <sup>6</sup>	1.42	1.45	1.69	1.44	0.09	0.18	0.10	0.09
Isovalerate <sup>6</sup>	1.11	1.13	1.17	1.06	0.12	0.65	0.99	0.52

<sup>1</sup>Yellow #2 corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Dry-rolled corn

<sup>4</sup>Whole-shelled corn

<sup>5</sup>Largest value among treatments reported.

<sup>6</sup>Average of values collected at 0, 2, 4, 6, 8, 12, 18, and 24 h after feeding expressed as a percentage of total VFA.

**Table 2.11 Effects of Enogen Feed Corn silage and corn on health (Exp. 3)**

Disease Diagnosis	Treatment Diet <sup>1</sup>
Bovine respiratory disease	EFC/ES
Bloat	CON/ES
Bloat	CON/CS
Lameness	CON/CS
Lameness	EFC/ES

<sup>1</sup>EFC/ES: Enogen Feed DRC/Enogen Feed Corn silage,  
CON/ES: Yellow #2 DRC/Enogen Feed Corn silage,  
CON/CS: Yellow #2 DRC/Mycogen corn silage

**Table 2.12 Effects of Enogen Feed Corn silage and corn on performance (Exp. 3)**

Item	Corn Silage Source				SEM	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Corn	Silage	Corn x Silage
	Dry-rolled Corn Source							
	CON <sup>3</sup>	EFC <sup>2</sup>	CON <sup>3</sup>	EFC <sup>2</sup>				
No. of pens	8	8	8	8				
No. of animals	88	87	87	88				
Body weight, kg								
d 0	301	299	297	297				
d 14	334	336	335	336	3.37	0.25	0.71	0.97
d 28	345	343	343	341	5.51	0.47	0.47	0.80
d 42	375	372	374	374	4.10	0.61	0.93	0.38
d 56	395	393	395	396	4.37	0.81	0.62	0.35
d 70	415	413	415	416	5.73	0.90	0.50	0.62
d 77	420	423	426	426	5.84	0.66	0.09	0.56
d 91	429	427	433	433	5.94	0.77	0.10	0.85
ADG, kg/day								
d 0-14	2.35	2.63	2.69	2.80	0.21	0.06	0.02	0.39
d 0-28	1.55	1.58	1.66	1.56	0.19	0.70	0.67	0.47
d 0-42	1.76	1.75	1.83	1.83	0.09	0.96	0.07	0.76
d 0-56	1.68	1.68	1.74	1.76	0.08	0.84	0.06	0.70
d 0-70	1.62	1.63	1.68	1.69	0.08	0.83	0.08	0.95
d 0-77	1.54	1.61	1.68	1.67	0.08	0.47	0.01	0.34
d 0-91	1.40	1.41	1.49	1.48	0.07	0.97	0.01	0.82
Average DMI, kg/day								
d 0-14	7.68	8.01	7.69	7.90	0.13	0.05	0.69	0.66
d 0-28	8.13	8.40	8.19	8.42	0.14	0.09	0.76	0.92

d 0-42	8.48	8.77	8.81	8.97	0.14	0.12	0.07	0.63
d 0-56	8.86	9.05	9.18	9.35	0.14	0.21	0.03	0.95
d 0-70	9.14	9.23	9.38	9.52	0.14	0.42	0.08	0.84
d 0-77	9.20	9.43	9.48	9.64	0.14	0.18	0.09	0.78
d 0-91	9.17	9.38	9.44	9.56	0.12	0.19	0.08	0.71
F:G, kg/kg								
d 0-14	3.28	3.08	2.88	2.84	0.10	0.24	<0.01	0.42
d 0-28	5.28	5.37	5.08	5.59	0.30	0.32	0.98	0.48
d 0-42	4.82	5.04	4.84	4.91	0.10	0.16	0.62	0.48
d 0-56	5.26	5.40	5.28	5.33	0.11	0.40	0.85	0.72
d 0-70	5.65	5.70	5.57	5.65	0.12	0.58	0.58	0.90
d 0-77	5.97	5.88	5.67	5.78	0.14	0.94	0.17	0.47
d 0-91	6.47	6.60	6.26	6.39	0.14	0.36	0.14	0.98
G:F, kg/kg								
d 0-14	0.306	0.328	0.350	0.354	0.011	0.24	<0.01	0.45
d 0-28	0.192	0.189	0.203	0.185	0.010	0.33	0.74	0.48
d 0-42	0.192	0.184	0.181	0.182	0.004	0.19	0.64	0.45
d 0-56	0.191	0.186	0.190	0.189	0.004	0.48	0.84	0.68
d 0-70	0.178	0.176	0.180	0.178	0.004	0.67	0.60	0.96
d 0-77	0.168	0.171	0.177	0.174	0.004	0.88	0.16	0.47
d 0-91	0.155	0.152	0.160	0.157	0.002	0.43	0.14	0.94
NEm, Mcal/kg <sup>4</sup>	1.61	1.58	1.62	1.60	0.02	0.31	0.42	0.89
NEg, Mcal/kg <sup>4</sup>	1.00	0.97	1.01	0.99	0.01	0.30	0.39	0.87

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<sup>1</sup>Mycogen corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Yellow #2 corn

<sup>4</sup>Net energy calculations from Galyean (2019) based on NRC (1996) requirements.

**Table 2.13 Fecal Analysis (Exp. 3)**

Item	Corn Silage Source				SEM	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>					
	Dry-rolled Corn Source							
	CON <sup>3</sup>	EFC <sup>2</sup>	CON <sup>3</sup>	EFC <sup>2</sup>		Corn	Silage	Corn x Silage
DM, %	18.6	19.3	18.6	19.2	0.68	0.38	0.99	0.94
Starch, % of DM	20.4	21.7	19.6	23.5	2.00	0.20	0.82	0.52

<sup>1</sup>Mycogen corn<sup>2</sup>Enogen Feed Corn<sup>3</sup>Yellow #2 corn

**Table 2.14 Ensiled Silage Analysis (Exp. 3)**

Item	Corn Silage Source		SEM	P-value
	CON <sup>1</sup>	EFC <sup>2</sup>		
DM, %	30.0	34.4	0.42	<0.01
Starch, % of DM	28.7	34.7	0.90	<0.01

<sup>1</sup>Mycogen corn<sup>2</sup>Enogen Feed Corn

**Table 2.15 Effects of Enogen Feed Corn silage and corn on digestibility and ruminal characteristics (Exp. 4)**

Item	Corn Silage Source				SEM <sup>4</sup>	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Corn	Silage	Corn x Silage
	Dry-rolled Corn Source							
	CON <sup>3</sup>	EFC <sup>2</sup>	CON <sup>3</sup>	EFC <sup>2</sup>				
Number of observations	7	8	8	8				
DMI, kg/d	7.91	7.93	7.46	7.18	0.51	0.49	<0.01	0.41
Ruminal								
pH <sup>5</sup>	6.37	6.47	6.32	6.37	0.09	0.27	0.23	0.67
Ammonia, mM <sup>5</sup>	3.92	3.45	3.87	2.94	0.45	0.06	0.44	0.51
Total VFA, mM <sup>5</sup>	106.9	106.6	113.4	111.3	5.52	0.82	0.27	0.85
Acetate, mM <sup>5</sup>	69.7	68.3	68.8	71.3	2.60	0.81	0.64	0.41
Propionate, mM <sup>5</sup>	20.7	21.5	27.4	22.4	1.35	0.09	<0.01	0.02
Butyrate, mM <sup>5</sup>	11.4	11.6	12.6	12.3	0.77	0.94	0.10	0.70
Isobutyrate, mM <sup>5</sup>	1.32	1.34	1.37	1.39	0.15	0.89	0.70	0.99
Valerate, mM <sup>5</sup>	1.46	1.30	1.42	1.37	0.11	0.35	0.86	0.58
Isovalerate, mM <sup>5</sup>	2.60	2.24	2.13	2.54	0.23	0.91	0.67	0.05
Liquid passage rate, %/h <sup>6</sup>	13.14	14.23	13.52	13.39	0.66	0.32	0.62	0.20
Digestibility, %								
DM	65.33	64.73	67.00	66.32	1.77	0.64	0.24	0.98
OM	67.66	67.14	69.19	68.56	1.73	0.66	0.26	0.97
NDF	58.58	60.18	61.01	60.56	2.01	0.75	0.44	0.56
ADF	59.75	60.66	61.62	60.55	2.04	0.96	0.63	0.58
Starch	84.59	83.55	85.85	83.94	2.51	0.31	0.57	0.76

<sup>1</sup>Mycogen corn

<sup>2</sup>Enogen Feed Corn

<sup>3</sup>Yellow #2 corn

<sup>4</sup>Largest value among treatments reported.

<sup>5</sup>Average of values collected at 0, 2, 4, 6, 8, 12, 18, and 24 h after feeding.

<sup>6</sup>Calculated values from samples collected at 2, 4, 6, 8, 12, and 18 after feeding.

**Table 2.16 Effects of Enogen Feed Corn silage and corn on ruminal VFA profile (Exp. 4)**

Item	Corn Silage Source				SEM <sup>4</sup>	P-value		
	CON <sup>1</sup>		EFC <sup>2</sup>			Corn	Silage	Corn x Silage
	Dry-rolled Corn Source							
	CON <sup>3</sup>	EFC <sup>2</sup>	CON <sup>3</sup>	EFC <sup>2</sup>				
Number of observations	7	8	8	8				
Ruminal VFA, % of total								
Acetate <sup>5</sup>	64.9	64.6	60.8	64.3	0.77	0.03	<0.01	0.01
Propionate <sup>5</sup>	19.2	19.9	23.8	19.9	0.78	0.03	<0.01	<0.01
Butyrate <sup>5</sup>	10.9	10.9	11.0	10.9	0.61	0.88	0.81	0.76
Isobutyrate <sup>5</sup>	1.21	1.22	1.24	1.25	0.09	0.90	0.73	0.96
Valerate <sup>5</sup>	1.32	1.20	1.24	1.23	0.06	0.17	0.54	0.20
Isovalerate <sup>5</sup>	2.50	2.14	1.87	2.28	0.16	0.78	0.11	0.01

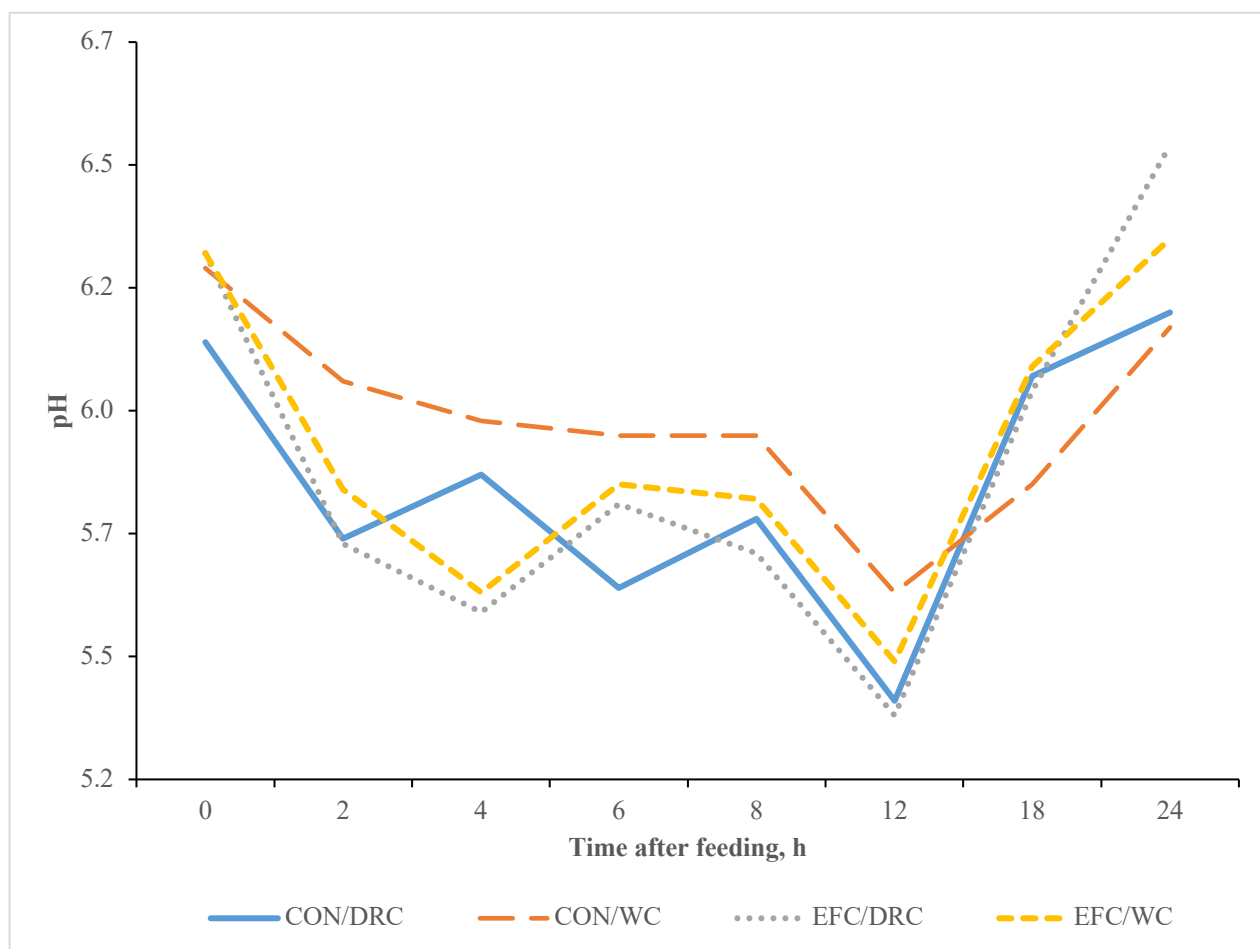
<sup>1</sup>Mycogen corn

<sup>2</sup>Enogen Feed Corn

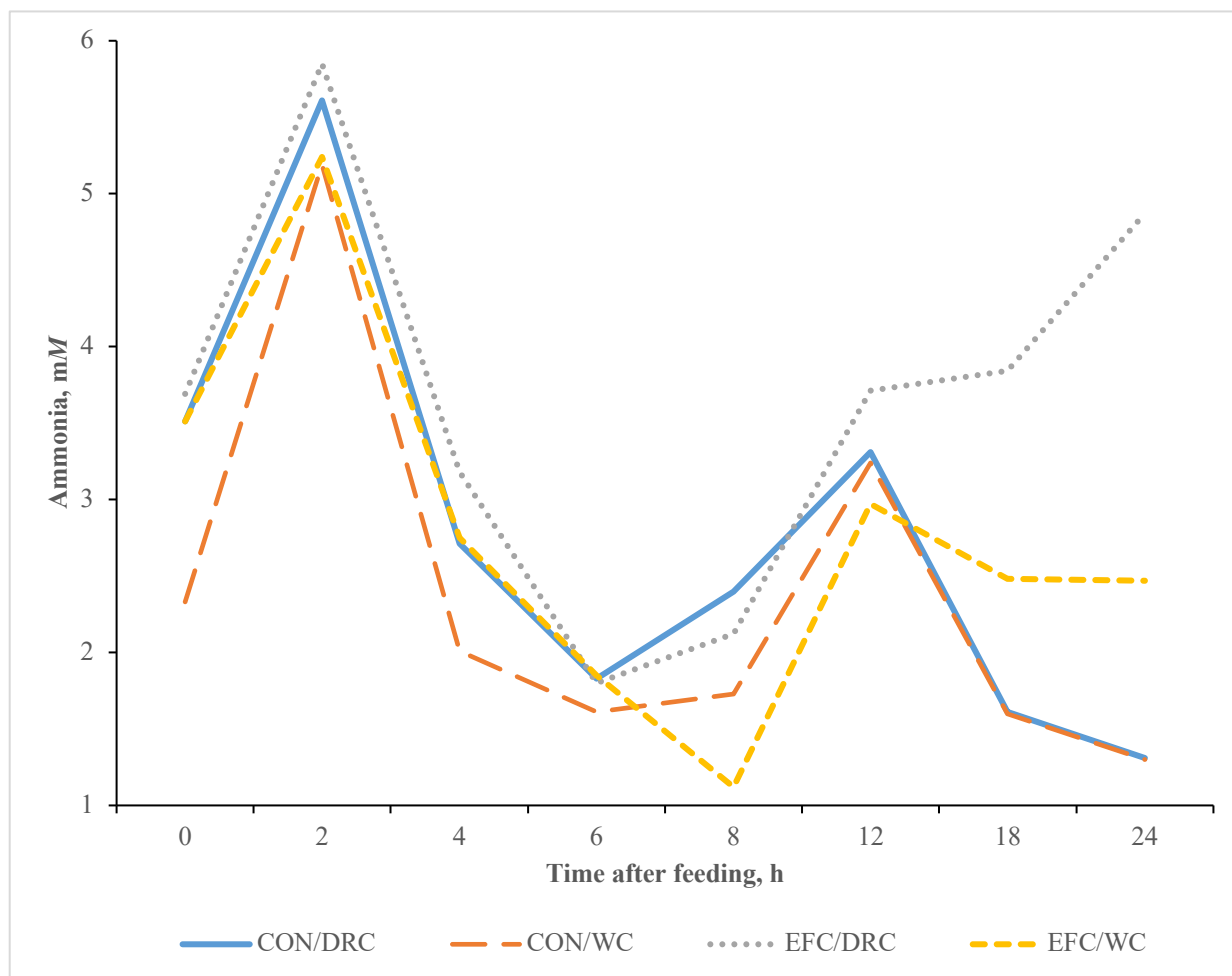
<sup>3</sup>Yellow #2 corn

<sup>4</sup>Largest value among treatments reported.

<sup>5</sup>Calculated values from samples collected at 0, 2, 4, 6, 8, 12, 18, and 24 h after feeding expressed as a percentage of total VFA.



**Figure 2.1 Effects of Enogen Feed Corn on ruminal pH measured over 24 h (Exp. 2).**  
 CON/DRC = Yellow #2 corn/dry-rolled corn. CON/WC = Yellow #2 corn/whole corn.  
 EFC/DRC = Enogen Feed Corn/dry-rolled corn. EFC/WC = Enogen Feed Corn/whole corn.  
 Corn effect ( $P = 0.82$ ) processing effect ( $P < 0.15$ ) corn x processing effect ( $P < 0.37$ ) hour  
 effect ( $P < 0.0001$ ) hour x corn effect ( $P < 0.01$ ) hour x processing effect ( $P = 0.40$ ) hour x  
 corn x processing effect ( $P = 0.64$ ).

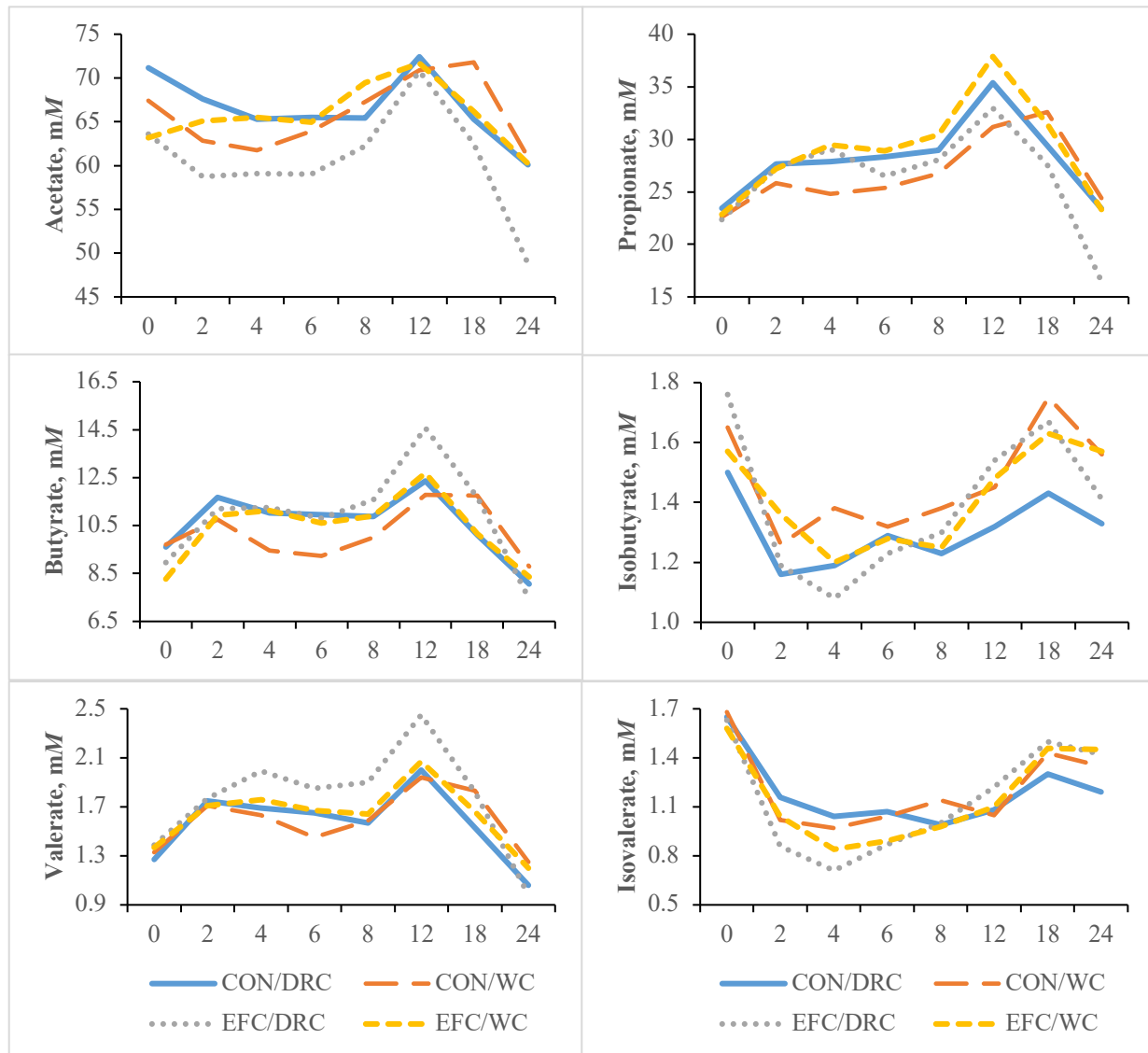


**Figure 2.2 Effects of Enogen Feed Corn on ruminal ammonia measured over 24 h (Exp. 2).**

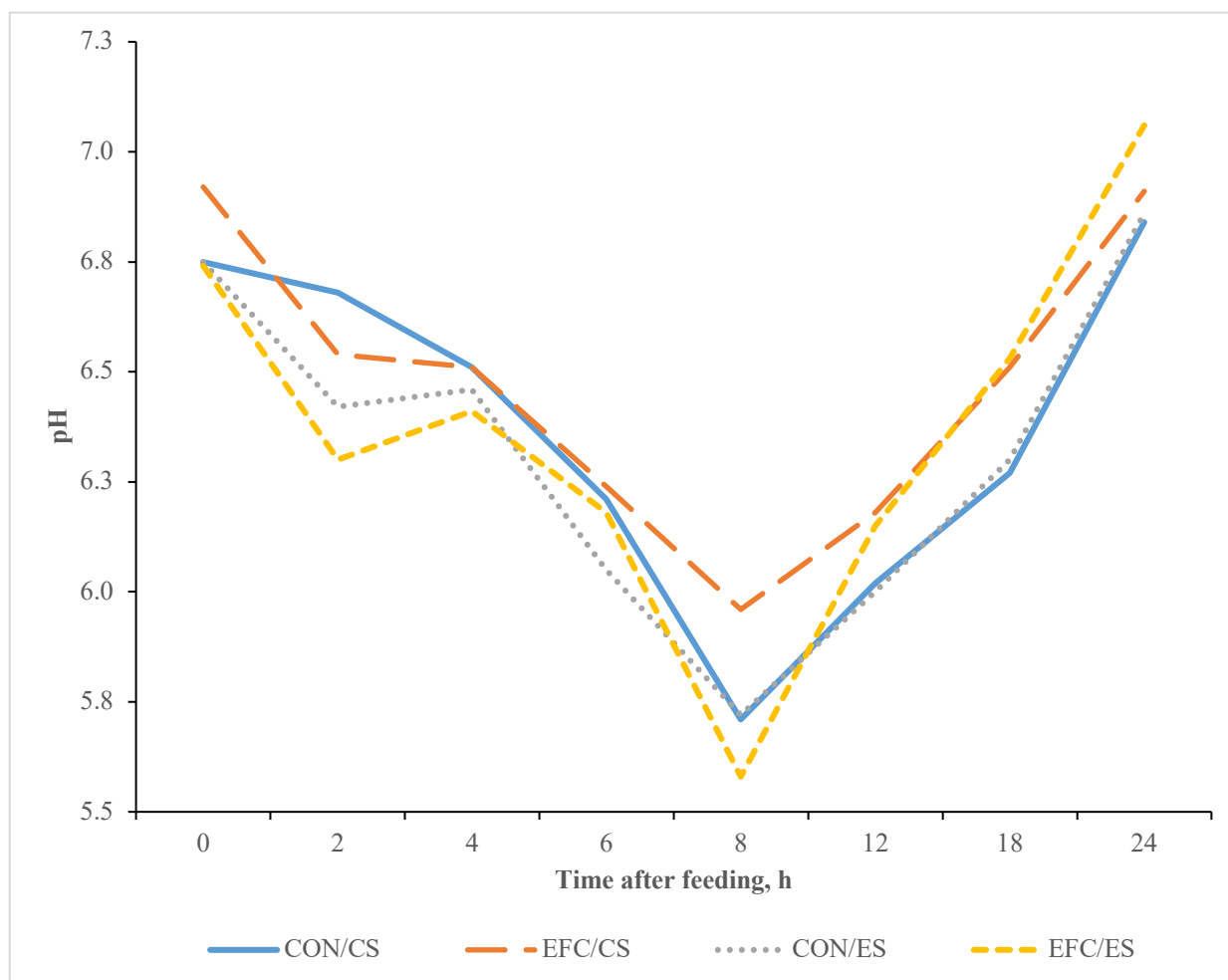
CON/DRC = Yellow #2 corn/dry-rolled corn. CON/WC = Yellow #2 corn/whole corn.

EFC/DRC = Enogen Feed Corn/dry-rolled corn. EFC/WC = Enogen Feed Corn/whole corn.

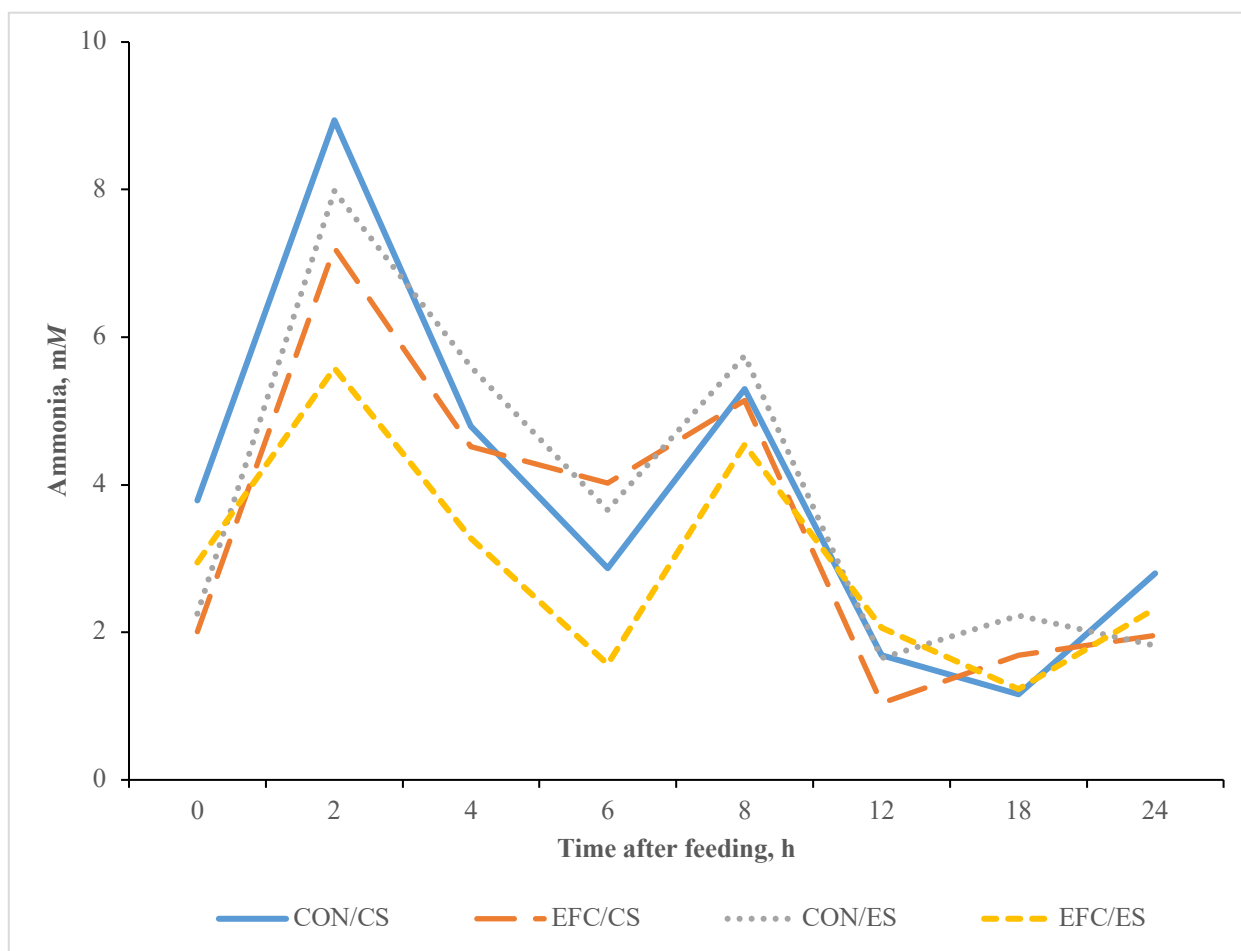
Corn effect ( $P < 0.30$ ) processing effect ( $P < 0.32$ ) corn x processing effect ( $P = 0.73$ ) hour effect ( $P < 0.0001$ ) hour x corn effect ( $P < 0.10$ ) hour x processing effect ( $P = 0.52$ ) hour x corn x processing effect ( $P = 0.67$ ).



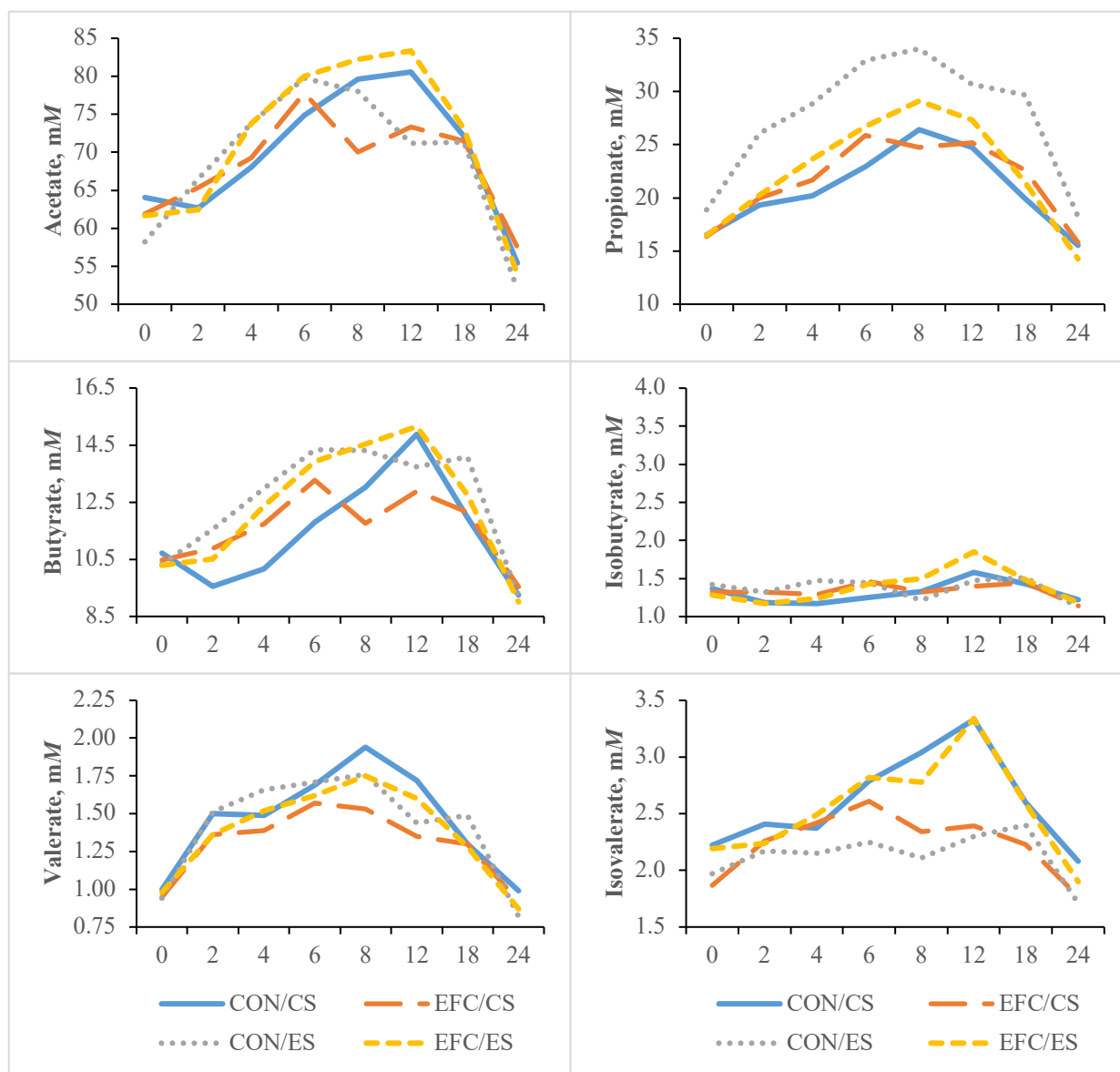
**Figure 2.3 Effects of Enogen Feed Corn on ruminal VFA concentrations over 24 h (Exp. 2).** CON/DRC = Yellow #2 corn/dry-rolled corn. CON/WC = Yellow #2 corn/whole corn. EFC/DRC = Enogen Feed Corn/dry-rolled corn. EFC/WC = Enogen Feed Corn/whole corn. For acetate, corn effect ( $P < 0.05$ ) processing effect ( $P < 0.16$ ) corn x processing effect ( $P < 0.06$ ) hour x corn effect ( $P = 0.79$ ) hour x processing effect ( $P = 0.52$ ) hour x corn x processing effect ( $P = 0.81$ ). For propionate, corn effect ( $P = 0.90$ ) processing effect ( $P = 0.73$ ) corn x processing effect ( $P < 0.29$ ) hour x corn effect ( $P = 0.56$ ) hour x processing effect ( $P = 0.76$ ) hour x corn x processing effect ( $P = 0.70$ ). For butyrate, corn effect ( $P = 0.52$ ) processing effect ( $P < 0.28$ ) corn x processing effect ( $P = 0.86$ ) hour x corn effect ( $P < 0.48$ ) hour x processing effect ( $P = 0.97$ ) hour x corn x processing effect ( $P = 0.70$ ). For isobutyrate, corn effect ( $P = 0.82$ ) processing effect ( $P < 0.29$ ) corn x processing effect ( $P < 0.38$ ) hour x corn effect ( $P < 0.33$ ) hour x processing effect ( $P = 0.63$ ) hour x corn x processing effect ( $P = 0.73$ ). For valerate, corn effect ( $P < 0.28$ ) processing effect ( $P = 0.64$ ) corn x processing effect ( $P = 0.48$ ) hour x corn effect ( $P = 0.83$ ) hour x processing effect ( $P = 0.59$ ) hour x corn x processing effect ( $P = 0.84$ ). For isovalerate, corn effect ( $P = 0.68$ ) processing effect ( $P = 0.82$ ) corn x processing effect ( $P = 0.97$ ) hour x corn effect ( $P = 0.39$ ) hour x processing effect ( $P = 0.93$ ) hour x corn x processing effect ( $P = 0.79$ ).



**Figure 2.4 Effects of Enogen Feed Corn silage and corn on ruminal pH measured over 24 h (Exp. 4).** CON/CS = Yellow #2 corn/Mycogen corn silage. EFC/CS = Enogen Feed Corn/Mycogen corn silage. CON/ES = Yellow #2 corn/Enogen Feed Corn silage. EFC/ES = Enogen Feed Corn/Enogen Feed Corn silage. Corn effect ( $P < 0.27$ ) silage effect ( $P < 0.23$ ) corn x silage effect ( $P = 0.67$ ) hour effect ( $P < 0.0001$ ) hour x corn effect ( $P = 0.90$ ) hour x silage effect ( $P = 0.82$ ) hour x corn x silage effect ( $P = 0.91$ ).



**Figure 2.5 Effects of Enogen Feed Corn silage and corn on ruminal ammonia measured over 24 h (Exp. 4).** CON/CS = Yellow #2 corn/Mycogen corn silage. EFC/CS = Enogen Feed Corn/Mycogen corn silage. CON/ES = Yellow #2 corn/Enogen Feed Corn silage. EFC/ES = Enogen Feed Corn/Enogen Feed Corn silage. Corn effect ( $P < 0.06$ ) silage effect ( $P = 0.44$ ) corn x silage effect ( $P = 0.51$ ) hour effect ( $P < 0.0001$ ) hour x corn effect ( $P = 0.91$ ) hour x silage effect ( $P = 0.87$ ) hour x corn x silage effect ( $P < 0.01$ ).



**Figure 2.6 Effects of Enogen Feed Corn silage and corn on ruminal VFA concentrations over 24 h (Exp. 4).** CON/CS = Yellow #2 corn/Mycogen corn silage. EFC/CS = Enogen Feed Corn/Mycogen corn silage. CON/ES = Yellow #2 corn/Enogen Feed Corn silage. EFC/ES = Enogen Feed Corn/Enogen Feed Corn silage. For acetate, corn effect ( $P = 0.81$ ) silage effect ( $P = 0.64$ ) corn x silage effect ( $P = 0.41$ ) hour x corn effect ( $P = 0.95$ ) hour x silage effect ( $P = 0.54$ ) hour x corn x silage effect ( $P < 0.32$ ). For propionate, corn effect ( $P < 0.09$ ) silage effect ( $P < 0.01$ ) corn x silage effect ( $P < 0.02$ ) hour x corn effect ( $P = 0.94$ ) hour x silage effect ( $P < 0.29$ ) hour x corn x silage effect ( $P = 0.41$ ). For butyrate, corn effect ( $P = 0.94$ ) silage effect ( $P < 0.10$ ) corn x silage effect ( $P = 0.70$ ) hour x corn effect ( $P = 0.98$ ) hour x silage effect ( $P < 0.28$ ) hour x corn x silage effect ( $P < 0.37$ ). For isobutyrate, corn effect ( $P = 0.46$ ) silage effect ( $P < 0.35$ ) corn x silage effect ( $P = 0.51$ ) hour x corn effect ( $P = 0.61$ ) hour x silage effect ( $P = 0.64$ ) hour x corn x silage effect ( $P < 0.34$ ). For valerate, corn effect ( $P < 0.35$ ) silage effect ( $P = 0.86$ ) corn x silage effect ( $P = 0.58$ ) hour x corn effect ( $P = 0.77$ ) hour x silage effect ( $P = 0.74$ ) hour x corn x silage effect ( $P < 0.04$ ). For isovalerate, corn effect ( $P = 0.91$ ) silage effect ( $P = 0.67$ ) corn x silage effect ( $P < 0.05$ ) hour x corn effect ( $P = 0.52$ ) hour x silage effect ( $P = 0.96$ ) hour x corn x silage effect ( $P < 0.01$ ).