NUTRITIVE VALUE OF MECHANICALLY PRESSED COTTONSEED MEAL COMPARED

WITH SOYBEAN MEAL FOR LACTATING DAIRY COWS

by

## **NATHAN WEBB**

(Under the Direction of John Bernard)

## **ABSTRACT**

The objective of this trial was to determine the nutritive value of cottonseed cake (CSC) compared with solvent extracted (SBM) or expeller soybean meal (ESM). The CSC was produced by dry extruding whole cottonseed at 121 to 149 °C for 12-20 sec. Treatments includes three dietary protein sources: SBM and ESM (CONT); CSC and ESM (CSBM); and CSC and SBM (CESM). No differences were observed in intake, milk yield or percentage of milk fat, lactose, or solid-not-fat. However, milk protein percentage was lower for CSBM compared with CONT but not different to CESM. Milk protein percentage was lower for CSBM compared with CONT but not different to CESM. Milk urea nitrogen (MUN) concentrations were lower for CSBM compared with CONT and CESM. The results of this trial indicate that the CSC can be substituted for soybean meal or heat-treated soybean meal without affecting intake, milk yield and composition. The lower MUN observed for CSBM suggest that substituting the CSC for SBM may have limited degradable nitrogen.

INDEX WORDS: mechanically pressed cottonseed cake, soybean meal, expeller soybean meal, milk yield, milk composition

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# **DEDICATION**

I would like to dedicate this thesis to my parents, who have always supported me and pushed me to always do things as for the glory of God. Thank you from the bottom of my heart.

# **ACKNOWLEDGEMENTS**

I would like to acknowledge the following individuals and groups for helping make this research trial possible. First, I would like to acknowledge and thank my major professor Dr. John Bernard for his support and guidance during my time completing my degree. I greatly appreciate all his efforts to make me successful during my time at the University of Georgia and in preparing me to enter the dairy nutrition industry. Secondly, I would like to thank the Georgia Cotton Commission for providing partial funding in support of this project and the Furst-McNess Company for supplying the cotton cake and other commodities used in the trial. Lastly, I would like to thank everyone involved in the trial at the University of Georgia Research Dairy Center and in the laboratory.

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## CHAPTER 1

## INTRODUCTION

Whole cottonseed is a feed ingredient commonly found in the southern and southwest regions of the United States were cotton production is one of the major crops grown. Whole cottonseed has been extensively fed to dairy cattle as a source of protein, high-quality fiber, and fat (Broderick et al., 2013). Whole cottonseed is bulky which makes it difficult to handle and transport and limits it use in many feed mills. Cottonseed meal is a byproduct from the extraction of oil from whole cottonseed and contains a blend of fat, protein, fiber, and minerals. The oil derived from whole cottonseed is a desirable product for the production of human foods. An increase in the use of cottonseed for biodiesel production has further increased markets for cottonseed oil and the resulting byproducts.

In recent years, several smaller scale feed mills have been constructed which use a dry extruder to extract cottonseed oil with the byproduct referred to as cottonseed cake. Unlike traditional processing methods using solvent to extract the oil leaving a relatively low amount of oil in the resulting meal, the dry extruder uses a screw and pressure to squeeze the oil from the cottonseed. This process does not remove all the oil from the seed and additionally generates heat that alters the cotton protein structure which results in an increase resistance to ruminal degradation in the resulting meal. The resulting cottonseed cake differs in composition and feeding value compared to solvent extracted meal, but data are limited on the actual feeding value for modern lactating dairy cows.

## **CHAPTER 2**

## LITERATURE REVIEW

## **Oil Extraction Process**

Cottonseed meal is the term used to describe the residual meal remaining after oil has been extracted from whole cottonseed. There are several processes used to extract oil with the most common being solvent extraction. The solvent extraction method required that the seed first be cleaned and then cracked. The cracked material is flacked to reduce the particle size which improves oil removal. The flaked material is heated or cooked to free the oil before it goes into an expeller or solvent extraction to harvest the oil. Most commercial mills used hexane as a solvent to extract the oil (KMEC Engineering, 2019). Cottonseed meal resulting from solvent extraction has lower concentration of fat than that from expeller processing.

Dry extrusion can be also be used to process oilseed. This process does not include a solvent extraction step which results in higher concentrations of oil and lower crude protein concentrations in the residual meal. Because of these differences, the resulting byproduct is commonly referred to as cotton cake rather than cottonseed meal as it has lower concentrations of crude protein (approximately 33%) and higher ether extract (approximately 5 to 7%), depending on the machine settings.

#### **Protein**

Feed protein can be divided into two categories, rumen degradable and rumen undegradable protein. As these names suggest, rumen degradable protein is potentially degraded

by the rumen microbes. The microbes initially breakdown the protein into oligopeptides, and eventually into individual amino acids. The peptides and amino acids can be incorporated directly into microbial crude protein or can be deaminated to release ammonia and a carbon skeleton that can be fermented by ruminal to provide energy (Russel, 2002). The released ammonia can be subsequently utilized by microbes to synthesize amino acids for microbial crude protein. However, excess ammonia derived from amino acid deamination is absorbed across the ruminal epithelium and transformed into the blood stream where it is transported to the liver and converted into urea. The blood (or plasma) urea can be recycled through saliva into the rumen and excess urea will be excreted in either urine or milk (Lapierre and Lobley, 2001). The degradation process has several potential inefficiencies because of the wasted energy from degrading proteins down to ammonia and their alpha keto acids, and then resynthesizing amino acids from these components. This anabolic process can also be used to incorporate ammonia derived from non-protein nitrogen with carbon skeletons to synthesize microbial crude protein. However, the microbial population can effectively improve the quality of feedstuffs that contain lower quantities of essential amino acids. In contrast, rumen undegradable protein is a more efficient protein, in terms of providing amino acids profiles to the abomasum of cattle that more closely match the original feedstuff composition. Typical rumen undegradable protein altered in their structure so that the microbial enzymes have greater difficulty accessing and cleaving the bonds that hold amino acids together (Andrade-Montemayor et al., 2009). Therefore, these proteins pass through the rumen relatively undegraded. Upon reaching the abomasum, hydrochloric acid denatures the proteins so that the animal's proteolytic enzymes in the small intestine can degrade these proteins into oligopeptides and single amino acids. Heat or chemical treatment can alter the structure of proteins to reduce their susceptibility to microbial

degradation. A high correlation has been observed with increased rumen undegradable protein when temperature of processing has increased and ammonia levels in the rumen have decreased. Pressure also proved to be a factor when whole cottonseed was autoclaved as opposed to dry heating (Tagari et al., 1986)

Whole cottonseed (WCS) protein is very degradable without additional physical or chemical treatment, and as much as 76.5% of the total protein being degradable (Park et al., 1998). When WCS is physically extruded, the heat and the pressure alter the structure of the crude proteins making them more resistant to ruminal microbial degradation. Meyer et al. (1999) indicated that extruded-expelled cottonseed meal is a valuable and useful source of rumen undegradable protein RUP. This was reported by Pena et al. (1986), when they reported a decrease in the ruminal ammonia and MUN concentrations rumen undegradable protein was fed to lactating dairy cows. The effects of the shift from degradable to undegradable protein in the ration of cattle was studied by Imaizumi et al. (2015) when ruminally degradable soybean meal was replaced with protein supplements high in rumen undegradable protein. Twenty-two of the 29 comparisons made in 15 metabolic trials reported decreased microbial protein synthesis in response to an increase in rumen undegradable protein (Imaizumi et al., 2015). Researchers have also observed that a greater proportion of amino acids and non-ammonia nitrogen escape the rumen when rumen undegradable protein is fed and that those amino acids are subsequently absorbed in the small intestine (Broderick et al., 2013). Despite the fact that cottonseed is lower in lysine content compared with soybean meal, the higher level of RUP in cottonseed meal appears to negate this (Brito and Broderick, 2007). Park et al. (1998) reported that the RUP fraction of expelled cottonseed meal is very digestible. Even though MUN concentrations tended

to be depressed with expelled cottonseed meal, concentrations of milk components were similar for all treatments.

# Milk Urea Nitrogen

Protein is degraded in the rumen into ammonia, peptides, and other nitrogen containing compounds. As the protein is degraded, ammonia is released. The ammonia can be captured by other microbes and attached to a carbon back bone to synthesize amino acids. However, when excess ammonia is produced, it is absorbed across the ruminal wall into the blood stream.

Ammonia is converted to urea in the liver to reduce its toxicity. Urea is transported throughout the body and is incorporated into milk and is measured as milk urea nitrogen (MUN).

Measurement of MUN provides` a metric for evaluating protein utilization. When MUN levels are depressed, there could be a reduced soluble protein concentration in the feed. This was evidenced in reports by Schepers and Meijer (1998) that MUN concentrations are positively correlated with the rumen degraded protein balance. As protein solubility decreases, so does MUN. In the study by Park (1998), whole cottonseed was replaced with express cottonseed which resulted in reduced MUN but had no effect on milk protein. This would suggest that the rumen undegradable protein level was high enough to maintain milk protein.

## **Fats from Cottonseed and Cottonseed Meal**

Feeding extruded oilseeds increases the fatty acid (FA) content of the diet (Dhiman et al., 1999). Feeding supplemental fat has been shown to increase milk fat percentages, but the results are not consistently. Dhiman et al. (1999) reported that cows fed extruded oilseed meals produced milk with lower concentrations of milk fat. Bauman and Griinari (2003) proposed the

biohydrogenation theory where certain conditions result in the partial biohydrogenation of polyunsaturated FA that results in the formation of trans-10, cis-12 CLA. When rumen pH is lower than optimal or excess polyunsaturated FA are fed, the normal pathway for biohydrogenation changes to one that produces trans-10, cis-12 CLA from linoleic acid. The partially hydrogenated FA escape from the rumen and are absorbed into the blood stream and then absorbed from the blood by the mammary gland. Trans-10, cis-12 CLA inhibits normal de novo synthesis of short chain FA by the mammary gland depressing milk fat (Chilliard et al., 2010). Under normal conditions, the mammary gland synthesis all of the short chain and most of the medium chain FA found in milk. Dietary long chain saturated and polyunsaturated FA are absorbed from blood and incorporated into milk fat (Lock et al., 2007).

Past research has shown that whole cottonseed increased milk fat production (Depeters et al., 1985, Noftsger et al., 2000) Whole cottonseed has been shown to actually limit the denovo synthesis of milk fat because of the partially biohydrogenated fatty acids escaping the rumen, reducing the synthesis of short chain FA (Smith et al., 1981). The three primary FA found in cottonseed oil are linoleic (C18:2), palmitic (C16:0), and oleic (C18:1), with linoleic representing over 50% of the total FA (Gunstone, 2011). This would potentially contribute to the production of trans-10, cis-12 CLA and cause milk fat depression under certain conditions. Whole cottonseed's fatty acids are released more slowly because the hull must be ruptured before rumen microbes can access fatty acids (Bernard, 1999). This was observed by Dhiman (1999) who fed the extruded cottonseed meal and observed a decrease in milk fat yield due to an increase in conjugated linoleic acid (CLA). However, Depeters et al (1985) reported that milk fat percent was increased with the introduction of whole cottonseed. Extrusion cracks the seed,

overloading the rumen's ability to properly biohydrogenate the polyunsaturated fatty acids (Mohamed et al., 1988).

Unsaturated FA exert an antimicrobial effect and can alter the microbiome of the rumen (Jenkins and Lock, 2008). Fatty acids are absorbed into the cell membrane of certain microbial species and causes a disorganization within the cell membrane leading to death of the microbe (Jenkins, 2002). Pantoja et al. (1994) reported that feeding unsaturated FA decreased fiber digestion in the rumen. Gerson et al. (1985) suggested that the bacteria that are responsible for fiber digestion, also biohydrogenated the unsaturated FA. Acetate is the VFA that is the primary precursor for denovo synthesis of fatty acids and is used to synthesize milk fat. A shift in the microbiome results also shifts VFA production in the rumen. Acetate, produced primarily from the fermentation of fiber, is decreased reducing the acetate to propionate ratio (Russell, 2002). Bernard and Calhoun (1997) reported that free oil in the rumen also increased propionate levels in the rumen, furthering reducing the acetate to propionate ratio.

# Gossypol

Cottonseed contains gossypol which is a phenolic compound located in the pigment glands of the stem, leaves, seeds and flowers of cotton (Rogers et al., 2002). There are two isomers of gossypol, a positive and negative isomer, where the negative isomer is the more bioactive of the two (Lordelo et al., 2007). Only free gossypol exerts a toxic effect (Bernard, 1999). Even though cottonseed meal is a high protein high oil feedstuff, it should only be fed to mature ruminants as monogastric animals are more susceptible to gossypol. Young, immature ruminants are also susceptible to gossypol because they do not have a functioning rumen (Gadelha et al., 2014). Gossypol has been shown to decrease DMI, milk production, and

increases red blood cell fragility in addition to other negative effects. When fed at high concentrations over time, gossypol can cause sudden death (Blasi and Drouillard, 2002). Gossypol also negatively impacts reproduction in both males and females. Males have decreased spermatogenesis (Randel et al., 1992), while females show a disruption of the follicular growth, pregnancy and early embryonic development (Lin et al., 1985). Cottonseed naturally have lower concentrations of lysine (Meyer et al., 2001) and lysine bioavailability is reduced through binding of gossypol to the epsilon group of lysine (Blackwelder et al., 1998). Extruded cottonseed has the lowest concentration of free gossypol of all the cottonseed products (Bernard and Calhoun, 1997). Processing cottonseed reduces free gossypol concentrations and the potential for gossypol toxicity when consumed by animals (Noftsger et al., 2000).

# **Summary**

Cottonseed is an extremely versatile feed ingredient. Pressing whole cottonseed is an economic way to reduce oil content. Cotton has a high polyunsaturated fatty acid profile that could reduce milk fat if not properly monitored. Add in the effect that the ruptured seed has because those fatty acids are dumped all at one instead of a gradual release with whole cottonseed. When WCS is pressed to remove oil, gossypol is bound up to the epsilon group of lysine. Reducing the already negligible impact gossypol would have to ruminants, especially at the levels that we are expecting to feed at. The binding of gossypol also highlights the altered protein structure that this form of processing has. Whole cottonseed has a high percentage of rumen degradable protein, but when processed with heat and pressure, the protein's structure becomes more resistant to microbial degradation. This is evidenced by the reduced milk urea nitrogen and decreased ammonia levels when feeding an expelled cotton cake.

We expected to see a reduction in milk urea nitrogen when replacing a rumen degradable protein source, in our case soybean meal, with this extruded cotton cake. We do not expect to see a decrease in milk fat because the research diets that are compiled do not reach the threshold of fats in the diet. All other parameters of production such as milk yield, solids-not-fat, lactose and milk fat should not be affected by the mechanically separated cottonseed meal.

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# CHAPTER 3

# NUTRITIVE VALUE OF MECHANICALLY PRESSED COTTONSEED MEAL COMPARED WITH SOYBEAN MEAL FOR LACTATING DAIRY COWS $^{\rm 1}$

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## **ABSTRACT**

Forty-eight lactating Holstein cows were used in a randomized block trial to evaluate the nutritive value of mechanically pressed cottonseed cake (CSC) compared with solvent extracted (SBM) or expeller soybean meal (ESM). The CSC was produced by dry extruding whole cottonseed at 121 to 149 °C for 12-20 sec. The resulting CSC contains approximately 33% CP with 40% rumen degradable and 60% rumen undegradable protein (DM basis). All cows were fed a control diet during the first 3 wk and data collected were used as a covariate in the statistical analysis. At the end of the preliminary period, cows were fed experimental diets for the following 8 wk. Treatments included: 1) control diet supplemented with SBM and ESM (CONT); 2) CSC substituted for SBM (CSBM); and 3) CSC substituted for ESM (CESM). No differences (P > 0.10) were observed in DMI (27.2, 28.1, and 28.1 kg/d), milk yield (33.0, 32.8, 33.4 kg/d), milk fat (3.96, 3.96=5, and 4.00%), lactose (4.67, 4.67, and 4.69 %), solids-not-fat (8.62, 8.52, and 8.57 %) or ECM (35.1, 34.7, and 35.7 kg/d) were not different among CONT, CSBM, or CESM, respectively. Milk protein percentage was lower (P = 0.0418) for CSBM compared with CONT but not different to CESM (2.96, 2.84, and 2.90% for CONT, CSBM and CESM, respectively). Milk urea nitrogen concentrations were lower (P = 0.0003) for CSBM compared with CONT and CESM: 8.56, 7.58, and 9.27 mg/dL for CONT, CSBM, and CESM, respectively. No difference (P > 0.10) were observed in BW change (31.7, 32.5, and 28.1 kg for)CONT, CSBM, and CESB, respectively). The results of this trial indicate that the CSC can be substituted for soybean meal or heat treated soybean meal without affecting intake, milk yield and composition. The lower MUN observed for CSBM suggest that substituting the CSC for SBM may have limited degradable nitrogen.

Key words: mechanically pressed cottonseed cake, soybean meal, expeller soybean meal, milk yield, milk composition

## INTRODUCTION

Whole cottonseed is an ingredient commonly found in the southern and southwest regions of the United States were cotton production is one of the major crops grown. Whole cottonseed has been extensively fed to dairy cattle as a source of protein, high-quality fiber, and fat (Broderick et al., 2013). Whole cottonseed is bulky which makes it difficult to handle and transport and limits it use in many feed mills. The oil in cottonseed is a desirable product for the human food markets. An increase the use of cottonseed for biodiesel production has also led to the increased markets for cottonseed oil and the resulting byproducts.

Cottonseed meal is a byproduct from the extraction of oil from whole cottonseed and contains a blend of fat, protein, fiber, and minerals. In recent years, several smaller scale mills have been constructed which use an expeller for extracting the oil with the byproduct referred to as cottonseed cake. Unlike tradition processing methods that use a solvent to extract the oil leaving a relatively low amount of oil in the resulting meal, the expeller process uses a screw and pressure to squeeze the oil out of the cottonseed. This process inevitable does not remove all the oil from the seed and generates heat that alters the cotton protein to increase resistance to ruminal degradation in the resulting meal. The resulting cottonseed cake differ in composition and feeding value compared to solvent extracted meal, but data are limited on the actual feeding value for modern lactating dairy cows.

# **MATERIALS AND METHODS**

# Animal and Feeding Management

All methods were reviewed and approved by the University of Georgia Animal Care and Use Committee prior to conducting the trial. Thirty-six multiparous and 12 primiparous Holstein cows were blocked by lactation number, days in milk (DIM) and milk yield and assigned randomly to 1 of 3 dietary treatments in the 11 wk randomized block designed experiment. Multiparous cows were enrolled at an average of 180 DIM and 78.4lbs, primiparous cows were enrolled at an average of 220 DIM and 72.9lbs. The first 3 weeks were a preliminary period and included as a covariate in the statistical analysis. At the end of the preliminary period, cows were switched to their respective treatment. Dietary treatments included a control diet that contained soybean meal and expeller soybean meal (CON), cotton cake replaced soybean meal (RDP) or cotton cake replaced the expeller soybean meal (RUP). Experimental diets were formulated to meet NRC requirements and are described in Table 1.

Animals were trained to eat through Calan (American Calan, Northwood, NH) prior to beginning the trial. Cows were housed in a 4 row, sand bedded free stall barn equipped with fans and misters to provide evaporative cooling when the temperature-humidity index was above 68. Cows were fed once daily at 1300 h and had free access to water. Refusals were collected once daily and the amount of feed offered adjusted to provide a minimum of 5% refusals. A base mix consisting of forages, molasses, and ground corn were mixed using a mixer wagon (Knight Khun Model 3120, Khun North America, Brodhead, WI) which was transferred to a Data Ranger (American Calan). Concentrate and the remaining dietary ingredients were added and mixed before individual feeding. Feed was pushed up three times each day.

# Sampling

Samples of individual feed ingredients, orts and experimental diet samples were collected 3 times each week. Dry matter was determined using a forced air drying oven set at 55 °C for 48h. Samples were first ground through a 6-mm screen using a Wiley mill (Thomas Scientific, Swedesboro, NJ). Samples were composited by experimental week. The composite sample was ground to pass through a 2-mm screen using a Wiley mill and submitted for chemical analysis. Samples were shipped to Cumberland Valley Analytical Laboratory for analysis of DM, ash, ether extract, ADF, (AOAC International, 2000), NDF adjusted for ash and organic matter (Van Soest et al., 1991), protein (LECO FP-5258 Nitrogen Analyzer, St. Joseph, MO), and minerals (AOAC International, 2000). Samples of cotton cake, soybean meal, and expeller soybean meal were subjected to the multi-step protein evaluation procedure described by Ross et al. (2013) to soluble protein, rumen degradable protein (RDP), rumen undegradable protein (RUP), intestinal digested protein, and total tract digested and undigested protein. Body weights were measured on 3 consecutive days before the beginning of the trial after the 1600 h milking. Water was restricted after milking until weighing was completed. Weights were averaged to determine average BW. Body condition score was assigned according to Wildman et al. (1982). Final BW and BCS were measured at the end of the trial.

Cows were milked 3 times e daily at 0800, 1600, and 2400 h. Milk weights were electronically recorded each milking (DelPro, Deleval, Kansas City, MO) and summed daily. Milk samples were collected once each week from three consecutive milking. Samples were refrigerated and shipped next day to Dairy One Cooperative (Ithaca, NY) for analysis of fat,

protein, lactose, and MUN using a Foss 400 instrument (Foss North America, Eden Prairie, MN) as described by AOAC International (2000).

# Statistical Analysis

Production data were subjected to repeated analysis of variance using PROC MIXED procedures of SAS (SAS Institute, Cary, N. C.). The model included covariate, block, treatment, week and the interactions of week and treatments. Data from the preliminary period were included as covariates in the analysis of production data. Cow within treatment was included as a random variable and week as a repeated variable. The first-order autoregressive covariance structure was used according to Littell et al. (1998). Initial BW, BCS, changes in BW and BCS, and nutrient digestibility data were subjected to analysis of variance. The model included the effect of treatment within cow as a random effect. Significance was declared at P < 0.05 and a trend when P > 0.05 and < 0.1.

#### RESULTS AND DISSCUSSION

The chemical composition of CSC, SBM and ESM is outlined in Table 2. The CSC had lower concentrations of CP and more soluble CP than SBM and ESM. Concentrations of RUP and RDP as measured using the procedures of Ross et al (2013) were intermediate between SBM and ESM. The intestinal digested protein concentration in CSC was 30.20% of CP which was slightly higher than SBM (25.85% of CP) but less than that observed for ESM (65.90% of CP). Total tract digestible protein was less for CSC compared with SBM and ESM: 75.55, 90.88, and 87.93% of CP, respectively. The RUP for SBM and ESM reported by NRC (2001) are 42.6% and

46.3% of CP, respectively, which is higher than that measured for the SBM and ESM used in this trial. Concentrations of CP, NDF, ADF and ether extract in solvent extracted cottonseed meal summarized by NRC (2001) were 44.9, 30.5, 19.9, and 1.9% of DM, respectively. In contrast, CSC has less CP and more NDF, ADF, and ether extract than solvent extracted cottonseed meal and reflect the difference in expeller and solvent extraction methods.

The chemical composition of the experimental diets is shown in Table 3. The CP concentrations for CONT, CSBM and CESM were 14.52% ( $\pm$  0.66), 14.05% ( $\pm$  0.65) and 15.07% ( $\pm$  0.48) respectively. This was approximately 1.5 to 2.0% units lower than formulated. The reason for the difference is not apparent. The supplements contained similar concentrations of CP as used for initial diet formulation. Concentrations of other nutrients were within expected ranges based on diet formulations.

There were no differences (P > 0.10 here and elsewhere) among treatments in DMI (Table 4), however, there was a treatment by wk interaction (P = 0.0128) as cows consuming CONT had lower DMI during wk 1 and 4 compared with those fed CSBM (Figure 1). Another study that was conducted comparing extruded cottonseed meal with whole, roasted and roasted pelleted cottonseed also observed no difference in DMI between treatments (Bernard and Calhoun, 1997). This is supported by Broderick et al. (2013) in that they reported that substituting extruded cottonseed meal for soybean meal resulted in decreased DMI. These authors suggested that increased oil content resulted in an increase in net energy of lactation, depressing intake. When mechanically pressed cottonseed meal replaces soybean meal (Bernard and Tao, 2017), no differences were observed in DMI among treatments.

No differences were observed (P > .10) in milk yield or percentage of milk fat, lactose or SNF among treatments. Average milk yield and percentage fat, lactose, SNF was 33.1 kg/d,

3.97%, 4.68% and 8.57%, respectively. Osti and Pandey (2006) reported that feeding cottonseed meal increased milk fat percentage in cows that were 40-50 DIM. When soybean meal was replaced with cottonseed meal (Imaizumi et al., 2015), milk fat percentage tended to increase quadratically. Contrary to other research where the oil seeds were cracked allowing for easier access to the oils in the whole seed, Dhiaman et al. (1999) reported decreased milk fat percentage. When soybean meal was replaced with CSC, milk fat percentage was depressed (Bernard and Tao, 2017). Perfield et al. (2007) also reported decreased milk fat percentage when extruded oil seeds were fed and attributed the decrease to increased partially hydrogenated conjugated linoleic acids escaping the rumen. In our current trial, the diets did not contain high concentrations of free oil so we would not have overloaded the rumen to allow partially hydrogenated fatty acids to escape from the rumen. If diets were not formulated to account for the increased and greater accessible oils, one could speculate that there would have been a reduction in milk fat percentage.

Milk protein percentage was lower (P = .0418) for CSBM (2.84%) compared with CONT (2.96%) but was not different from CESM (2.90%). This is consistent with previous research where soybean meal was replaced by cottonseed meal. Bernard and Calhoun (1997) reported a trend for depressed milk protein percentage when extruded whole cottonseed replaced whole cottonseed. Tashev and Todorov (1981) incorporated cottonseed meal into lactating dairy cow diets and observed decreased milk protein percent, but no differences in milk protein yield. Cottonseed meal supplementation further depressed milk protein and milk yield of Australian dairy cows (Grainger et al., 2010). Whole cottonseed was incrementally increased in the diet from 0 to 10% of DM and decreased milk protein percentage when fed at 10%. The decline in milk protein percentage was greatest for the highest producing cows (Depeters et al., 1985). In

our current trial, no differences (P > .10) were observed in milk protein yield among treatments. Yield of ECM and efficiency of milk production (ECM/DMI) was not different (P > 0.10) among treatments and averaged 35.2 kg/d and 1.27, respectively.

Concentrations of MUN were lowest (P = .0003) for CSBM (7.58 mg/dL) compared with CONT (8.56 mg/dL) and CESM (9.27 mg/dL). The ideal rage for MUN is 8mg/dL to 12 mg/dL (Kohn et al., 2002). The CONT and CESM diets were in the lower end of this range, the CSBM diet was below this threshold. This is consistent with the lower milk protein percentage observed for CSBM and may reflect a deficiency soluble or degradable protein resulting from the lower than planned dietary CP concentration. Given that milk yield and other components was not different among treatments, this would suggest that replacing SBM with CSC resulted in limited rumen soluble protein or degradable protein which limited microbial protein synthesis as reflected by the reduced MUN and milk protein concentrations. These finding are consistent with previous research reported by Meyer et al. (2001) where mechanically processed cottonseed meal fed at 16 % of the diet replace soybean meal. Milk urea nitrogen is a measure of nitrogen efficiency and feed efficiency (Godden et al., 2001). There is a lack of research examining MUN concentrations when soybean meal is replaced with cottonseed meal, but clues that can be gleaned from previous research regarding RUP concentrations and the inferences that can be made to the resulting MUN levels. In general, as RUP increases there is a corresponding decrease in MUN because of the reduced amount of ammonia produced in the rumen (Wattiaux and Karg. 2004). This assumption is also supported by Arieli (1998) who was stated that an increase in RDP is associated with increased rumen ammonia concentrations. When cottonseed meal was replaced by extruded-expelled cottonseed meal, an increase in RUP was observed. This increase in RUP could have resulted in a decrease in MUN (Park et al., 1998). Pena et al. (1986)

observed that extrusion of cottonseed decreases rumen ammonia concentrations. Imaizumi et al. (2015) summarized 15 trials where a soybean meal was replaced by varying amounts of whole cottonseed and observed an increase in RUP occasionally resulting in inadequate RDP concentrations to optimize microbial synthesis and ruminal fermentation. This would also result in decreased MUN concentrations. No differences (P > 0.10) were observed in BW or body condition score among treatments and other researchers also observed no change in body weight (Bernard and Calhoun, 1997).

# **CONCLUSION**

Expeller cottonseed meal is a viable replacement for soybean meal and expelled soybean meal. Special precautions must be made when considering this feed ingredient. When CSC replaces SBM, the results of our trial suggest that rumen soluble protein may be depressed although concentrations of soluble CP were higher for CSC than SBM. This is evidenced by the depressed MUN concentrations. The lower milk protein percentage observed in the current trial was possible due to the lower dietary CP and apparently lower soluble protein concentrations. The question remains, if dietary CP concentrations would have been higher in the diet, would there have been a difference in milk yield as previously reported (Bernard and Tao, 2017).

Milk fat percentage was not affected by the inclusion of CSC because dietary fat levels were maintained at normal concentrations within the diet, but the higher concentrations of ether extract in CSC should be considered when formulating diets to avoid overfeeding dietary fat that would be reactive in the rumen. Milk protein percent was depressed, further research needs to be conducted to see if crude protein was limiting the scope of the results.

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Table 3.1. Composition of experimental diets without cottonseed cake (CONT) or with cottonseed cake replacing soybean meal (CSBM) or expeller soybean meal (CESB).

	CONT	CSBM	CESM
Ingredient		% of DM	
Corn silage	45.03	45.03	45.03
Bermudagrass hay	4.33	4.33	4.33
Soybean hulls	12.12	8.66	6.93
Soybean meal	5.20		6.23
Expeller soybean meal	4.50	4.50	
Cottonseed cake		8.66	8.66
Ground corn <sup>1</sup>	10.82	10.82	10.82
Molasses	4.33	4.33	4.33
Citrus pulp <sup>1</sup>	9.53	9.53	9.53
Urea <sup>1</sup>	0.43	0.43	0.43
Alimet <sup>1,2</sup>	0.10	0.10	0.10
Salt <sup>1</sup>	0.26	0.26	0.26
Calcium carbonate <sup>1</sup>	0.17	0.17	0.17
Calcium monophosphate <sup>1</sup>	0.35	0.35	0.35
Magnesium oxide <sup>1</sup>	0.52	0.52	0.52
Sodium bicarbonate <sup>1</sup>	0.95	0.95	0.95
Potassium carbonate <sup>1</sup>	0.43	0.43	0.43
Potassium magnesium sulfate <sup>1</sup>	0.09	0.09	0.09
OmniGen-AF <sup>1,3</sup>	0.19	0.19	0.19
Diamond V XP Yeast1, <sup>4</sup>	0.19	0.19	0.19
Zinpro Availa Zn 1201, <sup>5</sup>	0.02	0.02	0.02
Vitamin E, 44,052 IU/kg <sup>1</sup>	0.17	0.17	0.17
TM-vitamin premix, <sup>6</sup>	0.26	0.26	0.26

<sup>&</sup>lt;sup>1</sup>Ingredients combine into a concentrate for feeding.

<sup>&</sup>lt;sup>2</sup>Methionine hydroxyl analogue (Novus International, Inc. St. Charles, MO)

<sup>&</sup>lt;sup>3</sup>Immune modulator (Phibro Animal Health Corp, Teaneck, NJ)

<sup>&</sup>lt;sup>4</sup>Yeast culture (Diamond V Mills, Cedar Rapids, IA)

<sup>&</sup>lt;sup>5</sup>Zinc amino acid complex (Zinpro Corporation, Eden Prairie, MN)

<sup>&</sup>lt;sup>6</sup>Mineral-vitamin premix contained (DM basis): 29.5% Ca; 0.06% P, 0.42% Mg; 0.31% S; 377 ppm Co; 3,472 ppm Cu; 530 ppm Fe; 388 ppm I; 23,882 ppm Mn; 110 ppm Se; 13,313 ppm Zn; 1,221,966 IU/kg Vitamin A; 129,456 IU/kg Vitamin D; 2,817 IU/kg Vitamin E

Table 3.2. Chemical composition of cottonseed cake (CSC), soybean meal (SBM) and expeller soybean meal (ESB).

	CSC	SBM	ESM	
DM, %	$93.65 \pm 1.47$	$90.42 \pm 1.58$	$89.48 \pm 1.58$	
		% of DM		
CP	$32.33 \pm 1.33$	$50.83 \pm 1.16$	$48.75 \pm 0.60$	
		% of CP		
Soluble CP	$16.73 \pm 5.77$	$12.93 \pm 3.24$	$8.13 \pm 1.84$	
$RUP^1$	$54.65 \pm 3.72$	$33.98 \pm 1.8$	$77.98 \pm 1.54$	
$RDP^2$	$45.35 \pm 3.72$	$66.03 \pm 1.68$	$22.03 \pm 1.54$	
$IDP^3$	$30.20 \pm 2.75$	$24.85 \pm 1.27$	$65.90 \pm 1.57$	
Total tract DP <sup>4</sup>	$75.55 \pm 1.18$	$90.88 \pm 0.61$	$87.93 \pm 0.53$	
		% of DM		
$aNDF_{OM}$	$43.78 \pm 1.24$	$8.03 \pm 0.46$	$17.75 \pm 3.13$	
ADF	$34.08 \pm 0.91$	$4.88 \pm 0.25$	$7.65 \pm 1.11$	
Ether extract	$5.53 \pm 0.26$	$1.9 \pm 0.2$	$1.32 \pm 0.02$	
NFC	$12.22 \pm 1.87$	$30.78 \pm 1.03$	$24.35 \pm 2.55$	
Ash	$6.14 \pm 0.50$	$8.56 \pm 1.77$	$7.83 \pm 0.70$	
Ca	$0.56 \pm 0.11$	$0.88 \pm 0.58$	$1.03 \pm 0.22$	
P	$0.88 \pm 0.02$	$0.83 \pm 0.15$	$0.77 \pm 0.01$	
Mg	$0.57 \pm 0.02$	$0.36 \pm 0.06$	$0.34 \pm 0.01$	
K	$1.69 \pm 0.02$	$2.84 \pm 0.08$	$2.74 \pm 0.08$	
Na	$0.12 \pm 0.03$	$0.55 \pm 0.34$	$0.09 \pm 0.04$	
		ppm		
Fe	$136 \pm 10$		$167 \pm 24$	
Mn	$60 \pm 6$	$123 \pm 89$	$50 \pm 8$	
Zn	$103 \pm 9$	$102 \pm 40$	$66 \pm 12$	
Cu	$18 \pm 3$	$25 \pm 6$	$19 \pm 2$	

<sup>&</sup>lt;sup>1</sup>RUP=Rumen Undegradable Protein

<sup>&</sup>lt;sup>2</sup>RDP= Rumen Degradable Protein

<sup>&</sup>lt;sup>3</sup>Intestinally digestible protein = protein that is rumen undegraded but digested in pepsin for 1 hr, then in trypsin, chymotrypsin, amylase and lipase for 24 h

<sup>&</sup>lt;sup>4</sup>Total tract digestible protein = total protein less intestinal undigested protein.

Table 3.3. Chemical composition of experimental diets without cottonseed cake (CONT) or with cottonseed cake replacing soybean meal (CSBM) or expeller soybean meal (CESB).

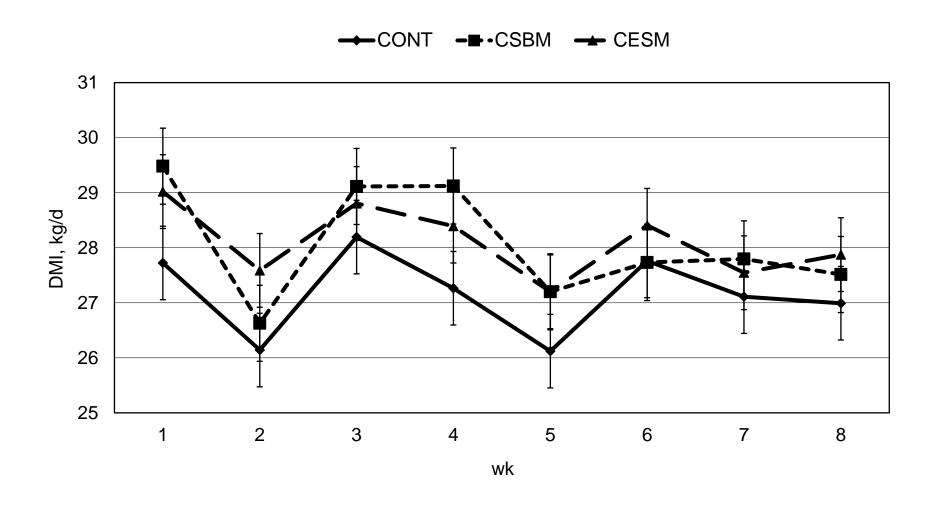
	CONT	CSBM	CESM			
DM, %	$54.90 \pm 5.04$	$55.33 \pm 5.22$	$55.28 \pm 5.28$			
		% of DM				
CP	$14.5\ 2\pm0.66$	$14.05 \pm 0.65$	$15.07 \pm 0.48$			
Soluble CP	$5.18 \pm 0.24$	$5.18 \pm 0.32$	$5.53 \pm 0.36$			
aNDF <sub>OM</sub>	$33.33 \pm 1.11$	$34.45 \pm 1.75$	$33.08 \pm 1.38$			
ADF	$22.18 \pm 0.77$	$23.38 \pm 1.17$	$22.40 \pm 0.66$			
Ether extract	$2.25 \pm 0.34$	$2.72 \pm 0.2.5$	$2.66 \pm 0.14$			
NFC	$41.48 \pm 1.97$	$40.21 \pm 2.79$	$40.65 \pm 1.91$			
Ash	$8.96 \pm 0.44$	$9.16 \pm 0.44$	$8.89 \pm 0.46$			
Ca	$1.09 \pm 0.09$	$1.07 \pm 0.10$	$0.96 \pm 0.11$			
P	$0.43 \pm 0.02$	$0.46 \pm 0.02$	$0.47 \pm 0.02$			
Mg	$0.59 \pm 0.05$	$0.62 \pm 0.06$	$0.59 \pm 0.04$			
K	$2.09 \pm 0.07$	$2.00 \pm 0.05$	$2.02 \pm 0.05$			
Na	$0.68 \pm 0.10$	$0.68 \pm 0.13$	$0.66 \pm 0.12$			
		ppm				
Fe	$619 \pm 21$	$611 \pm 48$	$584 \pm 61$			
Mn	$108 \pm 34$	$110 \pm 34$	$107 \pm 34$			
Zn	$117 \pm 31$	$115 \pm 27$	$113 \pm 33$			
Cu	$25 \pm 7$	$25 \pm 7$	$25 \pm 8$			

Table 3.4. Intake and performance of lactating dairy cows fed diets without cottonseed cake (CONT) or with cottonseed cake replacing soybean meal (CSBM) or expeller soybean meal (CESB).

	CONT	CSBM	CESM	SE	P
DMI, kg/d	27.2	28.1	28.1	0.6	0.4175
Milk, kg/d	33.0	32.8	33.4	0.5	0.7131
Fat, %	3.96	3.95	4.00	0.08	0.9246
Fat, kg/d	1.31	1.31	1.34	0.04	0.7183
Protein, %	$2.96^{a}$	2.84 <sup>b</sup>	$2.90^{ab}$	0.03	0.0418
Protein, kg/d	0.98	0.93	0.97	0.02	0.1781
Lactose, %	4.67	4.67	4.69	0.04	0.8711
Lactose, kg/d	1.54	1.53	1.57	0.03	0.7902
SNF, %	8.62	8.52	8.57	0.04	0.1893
SNF, kg/d	2.84	2.80	2.87	0.01	0.5766
ECM, kg/d <sup>1</sup>	35.1	34.7	35.7	0.7	0.6636
Efficiency, ECM/DMI	1.29	1.24	1.27	0.03	0.2494
MUN, mg/dL	$8.56^{a}$	$7.58^{b}$	$9.27^{a}$	0.27	0.0003
Initial BW, kg	614.8	636.6	647.0	18.6	0.4463
BW change, kg	31.7	32.5	28.1	5.1	0.8139
Initial BCS	3.23	3.12	3.18	0.06	0.4823
BCS Change	-0.09	0.06	-0.01	0.07	0.3949

abMeans in the same row with unlike superscripts differ (P < 0.05).

Figure 3.1. Interaction of dietary treatment and week (P = 0.0128) on trial for DMI of cows fed diets without cottonseed cake (CONT) or with cottonseed cake replacing soybean meal (CSBM) or expeller soybean meal (CESB).



# **CHPATER 4**

# **CONCLUSIONS**

Expeller cottonseed meal is a viable replacement for soybean meal and expelled soybean meal. Special precautions must be made when considering this feed ingredient. When CSC replaces SBM, the results of our trial suggest that rumen soluble protein may be depressed when substituted for SBM although concentrations of soluble CP were higher for CSC than SBM. This is evidenced by the depressed MUN concentrations. The lower milk protein percentage observed in the current trial was possible due to the lower dietary CP and apparently lower soluble protein concentrations. The question remains, if dietary CP concentrations would have been higher in the diet, would there have been a difference in milk yield as previously reported (Bernard and Tao, 2017).

Milk fat percentage was not affected by the inclusion of CSC because dietary fat levels were maintained at normal concentrations within the diet, but the higher concentrations of ether extract in CSC should be taken into account when formulating diets to avoid overfeeding dietary fat that would be reactive in the rumen. Milk protein percent was depressed when CSC replaced SBM. Further research needs to be conducted to see if crude protein was limiting the response of lactating dairy cows and would impact milk protein synthesis and MUN concentrations as observed in our current trial.