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EVALUATION OF SUPPLEMENTING BREWER'S YEAST TO LACTATING DAIRY COWS

BY

TAYLOR CHRISTINE AUBREY

A thesis submitted in partial fulfillment of the requirements for the

Master of Science

Major in Biological Sciences

Specialization in Dairy Science

South Dakota State University

2017

EVALUATION OF SUPPLEMENTING BREWER'S YEAST TO LACTATING DAIRY COWS

TAYLOR CHRISTINE AUBREY

This thesis is approved as a creditable and independent investigation by a candidate for the Master of Science degree in Biological Sciences and is acceptable for meeting the thesis requirements for this degree. Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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ACKNOWLEDGEMENTS

It takes a community to succeed, and my accomplishments are not my own. I would like to thank Dr. Jill Anderson, Dr. David Schingoethe, Dr. Vikram Mistry, and Dr. Weiwei Zhang for serving on my graduate committee. I appreciate your patience, guidance, and willingness to work with me to achieve my graduate school goals. In particular, I would like to thank Dr. Jill Anderson for adopting me into her graduate group and serving as my academic and research advisor. Her ability to accommodate my project and give me direction are some of the things I admire the most from her leadership qualities. Thank you, Jill for supporting me through my graduate studies. I want to thank Dr. David Casper for initially taking me on at SDSU and his assistance in setting-up the trial. In addition, I would like to thank the head of the Dairy and Food Science Department, Dr. Vikram Mistry for his unyielding support throughout my graduate work, and the rest of the department for providing me with learning experiences I will retain for the rest of my life.

I would be remiss to not thank the SDSU Dairy Research and Training Facility farm crew and employees, especially Pete Linke, Melissa Schmitt, and Laura McMartin. It would have been impossible for this trial to have gone on without your support, communication, and assistance in maintaining cow health and the facility. Also, I want to thank those who sacrificed their time to help me sample and feed my cows (and even me at times): Dinesh Shrestha, the Kass family (all 5 of them), Kali Linville, and Caitlin Louwagie. I especially want to thank Carl Kass for being my right-hand man, helping me fix things and working through issues together made my time out at the DRTF a much more enjoyable one.

Thank you to my fellow colleagues and graduate students for providing me with wisdom, laughter, and friendship. Special thanks goes to Karla Rodriguez-Hernandez, Nirosh Seneviranthne, and Rhea Lawrence for their assistance and expertise with laboratory procedures.

I also want to thank those who provided me encouragement and academic support through scholarships and awards, such as SDSU Women & Giving, SD Farmers Union Foundation, Mrs. Robert Breazeale, the Brandt Family and Land O'Lakes. I remain deeply honored to represent SDSU and the dairy industry, and without the willingness and generosity of such donors, it is challenging for students to confidently advance themselves in our field. Therefore, I sincerely thank you for your continuing support of students in higher education.

Lastly, I want to thank my friends and family for their belief, words of encouragement, and undying support of me in all facets of life. I am truly humbled to be surrounded by such a great group of people who have believed in me from day one and have provided me with the comfort to be myself. My parents, Phil and Jo Ann, and my brother, Anthony, have been my role models and constant source of strength through good and bad times. Their love, laughter, and support have helped shape me into the strong, confident individual I am today.

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ABBREVIATIONS

AA Amino Acid

ADF Acid Detergent Fiber

ADIA Acid Detergent Insoluble Ash

BCS Body Condition Score

BW Body Weight

CLA Conjugated Linoleic Acid

CON Control Diet (no yeast)

CP Crude Protein

DCAD Dietary Cation-Anion Difference

DDGS Distillers Dried Grains with Solubles

DHIA Dairy Herd Improvement Association

DIM Days in Milk

DM Dry Matter

DMI Dry Matter Intake

ECM Energy-Corrected Milk

EE Ether Extract

FA Fatty Acid

FCM Fat-Corrected Milk

FDA Food and Drug Administration

FE Feed Efficiency

GIT Gastrointestinal System

MUN Milk Urea Nitrogen

NDF Neutral Detergent Fiber

NEL Net Energy of Lactation

NFC Non Fiber Carbohydrate

NH₃-N Ammonia N

NRC Nutrient Requirements of Dairy Cattle

OM Organic Matter

peNDF Physically Effective Neutral Detergent Fiber

PUFA Polyunsaturated Fatty Acids

PUN Plasma Urea Nitrogen

rBST Recombinant Bovine Somatotropin

SARA Subacute Ruminal Acidosis

SCC Somatic Cell Count

SNF Solids-Not-Fat

SP Soluble Protein

TMR Total Mixed Ration

VFA Volatile Fatty Acid

VFD Veterinary Feed Directive

Y1 Concentrated Brewer's Yeast Diet

Y2 Concentrated Commercial Yeast Diet

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ABSTRACT

EVALUATION OF SUPPLEMENTING BREWER'S YEAST TO LACTATING DAIRY COWS

TAYLOR CHRISTINE AUBREY

2017

Probiotics have been used as effective, natural feed additives in the dairy industry to improve animal health and performance. Yeast product supplementation can be beneficial in the diet of lactating dairy cows by increasing feed efficiency, alleviating disease, and improving production performance under heat stress. Objectives of this study were to evaluate supplementing a concentrated brewer's yeast in the ration of a lactating dairy cow by assessing milk yield and composition, blood metabolites, rumen fermentation, and feed intake and efficiency. We hypothesized that diets containing a concentrated brewer's yeast supplement would increase milk and component yields, benefit rumen fermentation, and improve feed efficiency and nutrient utilization. Thirtysix Holstein cows (24 multiparous and 12 primiparous; DIM = 71.17 ± 16.42) were used in an 8-wk randomized complete block design experiment. Cows were blocked by milk yield, DIM, and parity. Treatments include: 1) control with no yeast (CON), 2) a concentrated brewer's yeast product (Y1), and 3) a commercial yeast product (Y2). Cows were fed a common TMR, except for yeast supplements (14.2 g/hd/d), once daily using the Calan Broadbent feeder system to determine daily individual dry matter intake (**DMI**). All milk weights were recorded daily and each week milk samples, body condition scores (BCS), and body weights were collected. Blood, rumen fluid, and fecal samples were taken during wk 7 and 8. Data were analyzed using MIXED procedures

with repeated measures and means were compared using Tukey's test. Dry matter intake was similar among treatments, but there was a treatment by week interaction (P < 0.01) with cows fed Y1 having greater DMI during wk 2, 3, 4 of the study. Milk production and components, including fatty acid composition were similar (P > 0.05) among treatments. Feed efficiencies, calculated as energy-corrected milk/DMI, were similar among treatments, but there was also a treatment by week interaction (P < 0.01). A treatment effect for plasma urea nitrogen (**PUN**) was detected (P < 0.01) and a treatment by group interaction for glucose was determined (P < 0.01). No statistical significance on treatment effects were determined for ruminal parameters and total-tract digestibilities. Yeast products maintained performance, rather than improving production as hypothesized.

Keywords: Yeast Supplement, Lactation Performance, Dairy Cow

INTRODUCTION

Animal agriculture and livestock production systems have come under immense scrutiny by society in recent years due to increasing demands over natural resources, such as land and water, for a growing human population. In addition, concerns over the environment and greenhouse gas emissions by livestock, as well issues pertaining to possible transfer of zoonotic diseases to people have caused humans to approach animal production more mindfully. These stressors have contributed to a concerted effort made by animal producers and scientists to deliver a sustainable, efficient product for current and future consumers. In order to create such products, various technologies and tools must be employed to alter the innate biology of the dairy cow in order to achieve production goals of maximum milk output, increased feed efficiency, and decreased environmental impacts. Therefore, feed additives, such as antibiotics, ionophores, and probiotics have been heavily researched and used by dairy producers to obtain these targeted areas of production.

In particular, fermented yeast and yeast cultures, such as *Saccharomyces cerevisiae*, also known as brewer's yeast, have been among the most scrutinized and utilized in numerous dairy cow, heifer, and calf diets to facilitate increases in dry matter intake (**DMI**) (Williams et al., 1991; Wohlt et al., 1991; Dann et al., 2000), milk production (Arambel and Kent, 1990; Piva et al., 1993; Wang et al., 2001), and stimulate rumen microbial fermentation and development (Moya et al., 2009; Alugongo et al., 2016). In addition to production benefits, supplementation of yeast in dairy cow diets has become more popular due to its naturally occurring state in the rumen, and growing concerns over antibiotic use in animal agricultural systems (Martin, 1998; Martin et al.,

1999). Antibiotics in animal feed have been banned in various regions of the world, and continue to face pressure through recent legislation, such as the Veterinary Feed Directive (**VFD**) in the United States, making it challenging to freely feed such products (Vohra et al., 2016). However, significant research and meta-analyses have shown inconsistent results when yeast is fed to cattle (Lescoat et al., 2000; Sauvant et al., 2004; Desnoyers et al., 2009).

Therefore, the objectives of this literature review are to discuss the purposes and previous literature findings of feeding yeast cultures to lactating dairy cows. In addition, the effects of supplementing yeast on production parameters will be highlighted and a justification for further research will be described. Objectives of the research project described herein were to evaluate supplementing brewer's yeast in the ration of a lactating dairy cow by assessing milk yield and composition, blood metabolites, rumen fermentation, and feed intake and efficiency. We hypothesized that diets containing a concentrated brewer's yeast supplement would increase milk and component yields, benefit rumen fermentation, and improve feed efficiency and nutrient utilization.

CHAPTER 1: LITERATURE REVIEW

Introduction

The expansion of the human population, income growth, and urbanization (Thornton, 2010) and the ability to feed people a high, animal-based protein source of food is causing the livestock industry to evolve. With limitations in land, water, and other essential resources, global food security has become challenging to achieve and sustain. Dairy products and other animal food sources have been widely recognized to meet and secure nutritional needs of a demanding global population (Murphy and Allen, 2003; Randolph et al., 2007). In addition, environmental awareness, and increasing concerns over greenhouse gas emissions of carbon dioxide, methane, and nitrous oxide have prompted consumers to become more conscious of products consumed (Bauman and Capper, 2011). Steinfeld et al. (2006) describes that approximately 9% of carbon dioxide, 37% of methane, and 65% of nitrous oxide emissions contribute directly or indirectly to livestock production. With mounting environmental challenges and continual growth of the human population, sustainability of animal agriculture, particularly the dairy industry, must be achieved through efficient production practices.

The United States dairy herd peaked at 26.5 million cows in 1944 with an average farm size of approximately 6 cows and an average daily milk production of less than 7 kg/cow (Capper et al., 2009). In contrast, modern day dairy farms employ around 500 cows to produce over 30 kg/d of milk, comprising about 60% of the total U.S. milk supply (USDA, 2007). These production efficiencies exemplify the current synergism between the understanding of dairy cow biological systems and advances in technology and farm management practices, such as implementation of artificial insemination (AI),

genetic selection programs, and improvements in feed analysis and diet formulation, to name a few (Bauman and Capper, 2011).

Therefore, it is essential for producers and scientists to work together to discover and implement practices beneficial for the producer, the animals, and the environment. Supplementation of feed additives has become common in feeding practices throughout U.S. dairies, demonstrating abilities to increase animal performance and health (Poppy et al., 2012), while decreasing the possibility of infection and disease (AlZahal et al., 2014). Common feed additives supplemented include antibiotics, ionophores, and probiotics, such as yeast cultures (Vohra et al., 2016). Due to various characteristics to be discussed, probiotics, such as yeast, are generally described to be an effective, natural strategy towards improving such parameters mentioned in animal production systems. This literature review will focus on the goals of feed additives, types of feed additives, and how yeast supplementation can positively influence dairy cow production.

Use of Feed Additives in Dairy Feeding

Feed additives are ingredients added to the diet of an animal to provide health, production, or environmental benefits (Vohra et al., 2016). When supplemented to ruminants, feed additives demonstrate a number of beneficial characteristics to the host that lead to increased productivity (i.e., increased milk, meat, and wool production) (Wallace and Newbold, 1995; Kung et al., 1997; Newbold, 2007; Nagaraja, 2012). Most of these advantages occur in the rumen by balancing the pH and preventing the production of lactate, which if over produced can lead to metabolic disease development, such as ruminal acidosis or bloat (Kung et al., 1997). Furthermore, reduction of ruminal pathogens in both neonates and older livestock can be achieved with feed additives. In

younger livestock, feed additives supplemented in calf starter have been demonstrated to improve rumen development and fermentation (Quigley et al., 1992; Brewer et al., 2014).

Feed additives also decrease ruminal methanogenesis and the acetate to propionate ratio without reducing milk fat synthesis, thereby improving ruminal energy utilization. In addition, improvements in ruminal nitrogen utilization are be marked by reduction of proteolysis, peptidolysis, and amino acid deamination, thereby minimizing production and losses of ammonia to the environment, and improving microbial protein synthesis (Hristov et al., 2009). Yeast cultures have also been shown to enhance ruminal microflora, leading to increased microbial fiber digestion due to increases in cellulose-digesting bacteria populations (Wiedmeier et al., 1987).

Nagaraja (2012) discussed how manipulations of ruminal fermentation by feed additive activities can decrease production of trans-fatty acids in dairy cows in order to offset decreased milk fat syndrome, also known as milk fat depression. This reduction in trans-fatty acids is countered by an increase in conjugated linoleic acids (**CLA**), which is a common acid found in meat and dairy products (Nagaraja, 2012). Conjugated linoleic acids have been extensively researched in dairy cows (Bauman and Griinari, 2003; Bauman et al., 2008), and certain isomers have been shown to inhibit milk fat synthesis (Bauman et al., 2008).

Manipulation of the rumen by supplementation of feed additives greatly alters the environment and microbial population of the host animal. Subsequently this can lead to maximizing the efficiency of feed utilization to further increase ruminant productivity and decrease unfavorable characteristics, such as greenhouse gas emissions and inefficiencies in feeding (Nagaraja, 2012).

Types of Feed Additives

There are three major categories of animal feed additives: antibiotics, ionophores, and probiotics (Vohra et al., 2016). Antibiotics have primarily been used in animal agriculture to improve growth rates, reduce mortality and morbidity, and improve reproductive performance (Cromwell, 2002). Subtherapeutic doses of antibiotics are mainly used to increase animal growth rates rather than prevent the spread of disease. However, levels of antibiotics in animal feed have been under immense scrutiny due to consumer concerns regarding the quality and safety of animal food products (Bauman and Capper, 2011). Excessive use of particular antibiotics in animal production systems has led to an increased risk of resistance genes to human pathogens, causing resistance among antimicrobial pathogens and impeding successful treatment. Such antibiotics used in both animals and humans include Penicillins, Cephalosporins, Quinolones, Fluoroquinolones, Tetracyclines, Macrolides, Sulfas, and Glycopeptides (Pyatt et al., 2016). Residue from the accumulated antibiotic in animal products is said to be harmful for human consumption, as it can hinder functions of beneficial microorganisms in the gastrointestinal tract (Cabello, 2006; Sorum, 2006; Yousefian and Amiri, 2009). Furthermore, recent changes by the U.S. Food and Drug Administration (FDA) to the Veterinary Feed Directive (**VFD**) have made it more difficult for a producer to readily supplement antibiotics in the feed of their animals (Pyatt et al., 2016). With increased regulations and consumer perceptions over antibiotics, producers and cattle nutritionists have been discovering new ways to positively alter production parameters and animal health.

Unlike antibiotics, other classes of animal or "non-medically important drugs", such as ionophores, do not require a VFD and can continue to be used as labeled (Pyatt et al., 2016). Ionophores (such as monensin, lasalocid, laidlomycin, salinomycin, and narasin) are over-the-counter antimicrobial compounds fed to ruminants in order to increase feed efficiency. These compounds target ruminal microbial communities of gram-positive bacteria and alter the ruminal ecology to retain more carbon and nitrogen, increasing overall production efficiency of the animal (Callaway et al., 2003). Due to the complexity of the gram-negative bacterial lipopolysaccharide cell wall layer, these populations are less impermeable to ionophores compared to gram-positives (McGuffey et al., 2001). Researchers have proved that ionophores can improve nitrogen efficiency, decrease the risk of ruminal acidosis of cattle fed high-grain diets, and decrease methane emissions (McGuffey et al., 2001; Ipharraguerre and Clark, 2003; Beauchemin et al., 2008). Ionophores act on the cell membrane of susceptible bacteria, killing them due to transportation of ion gradients which inhibit the bacteria to effectively grow. Successful infiltration causes an efflux of intracellular K⁺ from the cell and an influx of extra cellular protons (Na⁺ and H⁺). However, not all bacteria, such as gram-positives, are susceptible to ionophores and can develop mechanisms of resistance. Unlike antibiotics, ionophores have demonstrated a complex degree of specificity and do no contribute to the development of antibiotic resistance. Therefore, with favorable legislation and supportive science, ionophore use in animal production is likely to continue (Callaway et al., 2003).

Due to safety and health issues of resistance and consumer skepticism surrounding antibiotics and ionophores, a third category of probiotics has become a

popular choice among producers in the feed additive realm. The Food and Agriculture Organization of the United Nations (**FAO**) and World Health Organization (**WHO**) defined probiotics as 'live microorganisms which when administered in adequate amounts confer a health benefit on the host' (FAO, 2005). Supplementing probiotics, such as live yeast or yeast cultures, to animals is an effective, natural strategy towards improving their health and performance. Especially in ruminants where yeast are naturally occurring organisms present in the main fermentation site of the cow's digestive tract (the rumen). The most commonly fed and researched yeast is Saccharomyces cerevisiae because of its approved commercialized use for human consumption (Jakobsen and Narvhus, 1996; Lourens and Viljoen, 2001; Sargent and Wickens, 2004) and successful results in feeding adult ruminants (Fuller, 1999; Seo et al., 2010). Other strains of yeast demonstrating probiotic properties belong to the genus *Pichia*, Metschnikowia, Yarrowia, Candida, Debaryomyces, Isaatchenkia, and Kluyveromyces. Yeast supplementation has also demonstrated its importance in improving the nutritive quality of feed and feed utilization, thereby enhancing production (Wallace and Raleigh, 1960; Martin et al., 1989). Researchers have verified the single-celled eukaryotes' biologically valuable nutritive components, including proteins, vitamin B-complexes, and trace minerals, as well as its ability to produce extracellular enzymes, such as amylases, β-galactosidase, and phytases (Thayer et al., 1978; Moore et al., 1994; Vohra and Satyanarayana, 2001; Paryad and Mahmoudi, 2008). Probiotics have the ability to withstand the harsh acidic and high bile concentrated environment of the gastrointestinal tract (GIT), while demonstrating the ability to adhere, replicate, and colonize the GIT for a prolonged period of time (Ziemer and Gibson, 1998; Dunne et al., 1999; Mombelli and

Gismondo, 2000; Soccol et al., 2010). These aforementioned qualities of increased safety, health, and performance when fed to animals make yeast a viable and preferred option for producers and their herds.

Types of Yeast Products Fed

Most commercially available yeast supplementation products contain a mixture of varying proportions of live and dead Saccharomyces cerevisiae cells. Products containing live cells are sold as live yeast, and have been formulated to ensure optimal growth conditions for ruminal bacteria by preventing the accumulating of lactic acid within the rumen (Nocek, 1997). Other yeast products containing more dead cells and the growth medium are sold as yeast cultures (Newbold and Rode, 2006). Therefore, yeast culture products do not contain a guaranteed live yeast cell level, but rather yeast fermentation by-products, such as dried yeast fermentation solubles (B-vitamins and organic acids) and plant protein products (amino acids). Callaway and Martin (1997) have suggested that these by-products affect the growth of ruminal microbes by stimulating the bacterium Selenomonas ruminantium, which can alter rumen fermentation. Brewer's yeast is another yeast alternative derived from the by-product of breweries and is obtained from the brewing process once it is complete. Yeast cell removal of Saccharomyces cerevisiae from beer production is immediately inactivated by means of organic acids and is a viable feedstuff in the livestock industry (Crawshaw, 2004; Hertrampf and Piedad-Pascual, 2000). Brewer's yeast can be fed fresh (liquid form) or dried and subsequently ground (brewer's dried yeast) (Chauvel et al., 1988; Hertrampf and Piedad-Pascual, 2000) to provide a main source of protein, vitamins, and minerals (Stone, 2006).

Another type of product manufactured to maintain a specific number of live yeast cells (> 1.5×10^5 cfu/g of DM) is known as active dry yeast (AlZahal et al., 2014). It has been proposed that active dry yeast, mechanistically, are focused on optimizing fiber digestion within the rumen (Chaucheyras-Durand et al., 2008). Due to their inherent state, dry yeast can only survive for a short period of time within the rumen by utilizing traces of dissolved oxygen. A decrease in oxygen subsequently increases the population of ruminal microbes, particularly the cellulolytic digesting bacteria of Fibrobacter succinogenes, Ruminococcus albus, and Butyrivibrio fibrosolvens (Girard and Dawson, 1994; Callaway and Martin, 1997; Mosoni et al., 2007). This action aids directly in fiber digestion of diets containing high concentrations of forage feedstuffs, and can help to create the ideal anaerobic ruminal environment for bacterial growth and nutrient digestion (AlZahal et al., 2014). Some examples of commercial yeast include Yea-sacc (Alltech Inc.), Levucell SC-20 (Lallemand Animal Nutrition), and Diamond V Original (Diamond V, Mills Inc.). Other less common supplemented yeast products include enzymatically hydrolyzed yeast (Nocek et al., 2011) and Saccharomyces cerevisiae fermentation product (**SCFP**) (Mullins et al., 2013).

In general, these yeast products carry out their actions in the rumen by stimulating microbial communities, enhancing oxygen sequestration, and modulating ruminal pH. When *Saccharomyces cerevisiae* was supplemented Wallace and Newbold (1995) found a 50% increase in the viable ruminal bacteria population. In addition, increases in fiber-digesting bacteria, such as *Fibrobacter succinogens*, *Ruminococcus* spp., and *Butyrivibrio fibrosolvens* (Weidemeier et al., 1987) have been reported in the rumen when yeast products were supplemented. Together these microbes enhance fiber

digestion, and thus increase feed intake (Weidemeier et al., 1987; Chaucheyras-Durand et al., 2008). When supplemented to a ruminant, yeast has the ability to consume oxygen presented to the rumen through water intake, rumination, and salivation (Wallace and Newbold, 1995). This action helps inhibit the growth of obligate cellulolytic anaerobes and makes the rumen a more conducive anaerobic environment. Live yeast has been shown to increase rumen pH, thereby decreasing the variation of rumen pH in a number of studies with differing levels of starch in the diets (Guedes et al., 2008). Yeast is also able to modulate rumen pH by stimulating mechanisms of protozoa to engulf starch particles and prevent the fermentation of lactate. This engagement of protozoa helps to diffuse lactate accumulation and, thus, cause an increase in ruminal pH (Williams and Coleman, 1997; Brossard et al., 2006).

Feeding Yeast to Dairy Cows

Yeast cultures have been fed to dairy cattle for more than 60 years with varied responses (Schingoethe et al., 2004) and are marketed under a variety of trade names (Robinson and Erasmus, 2009). Recent studies (Kung et al., 1997; Erasmus et al., 2005) and meta-analyses (Desnoyers et al., 2009; Robinson and Erasmus, 2009; Poppy et al., 2012) have indicated that yeast supplementation in ruminant diets yield variable results. Many claims have been made about the impact yeast cultures have on ruminant performance, which include increased feed efficiency (Schingoethe et al., 2004), milk production (Poppy et al., 2012), rumen fiber fermentation (Wohlt et al., 1988; Guedes et al., 2008; Marden et al., 2008), rumen microbial protein synthesis, rumen pH and digestion (Hasunuma et al., 2016). These performance standards involve largely unknown and complex pathways (Russell, 2002) that encompass a variety of mechanisms

in the rumen, which play a vital role in ruminant nutrition practices (Van Soest, 1994). The main goal of feeding yeast is to increase nutrient utilization in the rumen, thereby manipulating its function to positively impact the cow's production status.

Effects on the Rumen Environment, Fermentation, and Digestibility

The rumen is the main site of fermentative and complex hydrolytic processes in the ruminant. The rumen is populated by numerous obligate or facultative anaerobic microbial communities of bacteria, archaeons, ciliate protozoa, flagellate protozoa, and anaerobic fungi. This microbial community plays a large role in ruminant nutrition by facilitating fermentation and digesting complex plant polymers, such as cellulose, hemicelluloses, starch, and protein. This population provides essential energetic components, detoxifies compounds that are introduced as toxic, stimulates the immune response, and inhibits the pathogenic microorganism from attaching and penetrating the host (Vohra et al., 2016). More specifically, yeast in the rumen function to remove oxygen from freshly consumed feed by the host (Newbold et al., 1996), creating a desired anaerobic environment that promotes growth and multiplication of anaerobic bacteria, thus improving the metabolic activity in the rumen (Jouany, 2001; Jouany, 2006; Chaucheyras-Durandand and Durand, 2010). This anaerobic environment allows for ideal conditions for those strictly anaerobic microbes, such as cellulolytic bacteria, and aids in stimulating attachment to forage particles and increases the rate of cellulolysis (Seo et al., 2010). These conditions have led to improvements in fiber digestion and other nutrient digestibilities when yeast products are supplemented in the diets of dairy cows (Wiedmeier et al., 1987; Guedes et al., 2008; Marden et al., 2008). Previous

research has also demonstrated that supplementation of yeast to poor quality forage and high grain diets can increase the nutritional value of the ration (Wallace and Raleigh, 1960).

Ferraretto et al. (2012) reported that cows fed a high-starch diet containing 2 g/hd/d of yeast supplementation digested more dry matter (**DM**), organic matter (**OM**), and starch than those fed a high-starch diet with no yeast supplementation. In the same study, a high starch-diet containing 4 g/hd/d of yeast supplementation tended to have a greater organic matter digestibility, dry matter digestibility, and non-detergent fiber (**NDF**) digestibility than the high-starch diet without yeast (P < 0.09, P < 0.06, P < 0.01,respectively; Ferraretto et al., 2012). However, Ferraretto et al. (2012) determined that low-starch diets without yeast supplementation were greater in DM digestibility, OM digestibility, NDF digestibility, and starch digestibility than high-starch diets. The authors suggested the greater total-tract digestibilities may be related to a theory posed by Firkins (1997), which implies a reduced negative association of starch on ruminal fermentation (Ferraretto et al., 2012). Marden et al. (2008) and Bitencourt et al. (2011) reported that by feeding a live cell yeast (1 x 10¹⁰ cfu/g of S. cerevisiae) to cows at a rate of 5 g/hd/d and 1 g/hd/d, respectively, increased the NDF digestibility (P = 0.03 and P =0.08, respectively). In the meta-analysis report of Desnoyers et al. (2009), Saccharomyces cerevisiae supplementation and dosage linearly increased OM digestibility (P = 0.004). Desnoyers et al. (2009) found that yeast supplementation on OM digestibility was decreased by the proportion of concentrate in the diet (P = 0.020)and increased by dietary NDF content (P < 0.001), crude protein (**CP**) content (P =0.013), and dry matter intake (**DMI**; P = 0.002). Similar analysis of OM digestibility was reported in other meta-analyses by Lescoat et al. (2000) and Sauvant et al. (2004). In contrast, Leicester et al. (2016) reported findings of decreased whole tract OM (P = 0.08, P = 0.02) and CP (P = 0.05, P < 0.01) digestibility in cows fed two diets supplemented with two yeast products (Diamond V XPC, 14 g/hd/d; Yeasture DFM, 10 g/hd/d, respectively). It was suggested by the researchers that more OM and CP likely entered the small intestine in endogenous secretions, suggesting improvements in the intestinal health of cows fed yeast products (Leicester et al., 2016). Hristov et al. (2010) reported no total-tract digestibility effects of a yeast product supplemented at 56 g/hd/d. However, some studies have also reported no effect on NDF and OM digestibility in dairy cows fed live yeasts, but rather demonstrate increased digestibility for CP and acid detergent fiber (ADF) (Erasmus et al., 1992; Kristensen et al., 2014).

The yeast specie of *Saccharomyces cerevisiae* is able to compete with other amylolytic bacteria for fermentation of starch (Lynch and Martin, 2002). This characteristic of yeast decreases the likelihood of lactate accumulation in the rumen, and increases the response of ruminal growth factors, such as organic acids or vitamins, stimulating populations of cellulolytic bacteria and lactate-utilizing bacteria (Chaucheyras et al., 1995). Lactate, a common product of carbohydrate fermentation, has a lower acid dissociation constant and is not readily absorbed from the rumen, causing it to decrease ruminal pH much more rapidly than volatile fatty acids (**VFA**) if present in high concentrations (Longuski et al., 2009). However, yeast has demonstrated its ability to increase ruminal end product concentrations (Lescoat, 2000), improve ruminal microbial protein and digestibility, while reducing ruminal lactic acid (Robinson, 2002; Desnoyers et al., 2009). Robinson (2002) showed an average increase in pH (1.6%), an

overall increase in rumen VFA concentrations (5.4%), and an overall decrease in lactate concentration (5.4%) in cow's supplemented yeast. In addition, Desnoyers et al. (2009) demonstrated that both rumen pH (P < 0.01) and VFA concentration (P < 0.05) were increased, while lactic acid demonstrated a tendency to be reduced (P < 0.10), indicating the ability of yeast products to decrease rumen pH that is usually linked to an increase in VFA.

Altering the ruminal environment and stabilizing its pH is another valuable, proven characteristic of yeast product supplementation in cows. Regulation of rumen pH can be essential in situations where there is a microbial imbalance, such as during the transition period (approximately 21 d before and after calving) where diet composition can change from a high forage-based to a high concentrate-based ration. Concentrated feeds are a source of rapidly fermentable carbohydrates, which when digested can lead to an increase in rumen VFA, thereby decreasing the pH. If the rumen buffering system is unable to neutralize this decrease in pH over prolonged periods of time, then potential health concerns can occur. In addition, decreases in productivity, microbial metabolism, and nutrient degradation have been issues to plague cows with suboptimal rumen pH (Chaucheyras-Durand et al., 2012). Health concerns, such as acidosis, inflammation, laminitis, diarrhea, and milk fat depression as well as decreased productivity due to decreased feed intake have been associated with a reduced pH. In various in vivo experiments, ruminal pH lowering effects of live yeast supplementation has been observed (Michalet-Doreau and Morand, 1996; Bach et al., 2007; Marden et al., 2008). In addition, increases in rumen pH have also been observed in non-acidotic, yeast supplemented cows (Guedes et al., 2008). Modifications of total rumen VFA molar

proportions and stabilization of ruminal pH when probiotic yeast was supplemented in the diets have been demonstrated in numerous studies (Lila et al., 2004; Hucko et al., 2009; Helal and Abdel-Rahman, 2010). In contrast, some studies have been consistent in demonstrating a lack of ruminal pH effects when yeast cultures are supplemented to lactating dairy cows (Wiedmeier et al., 1987; Erasmus et al., 1992; Yoon and Stern, 1996). Putnam et al. (1997), Robinson and Garrett (1999), and Hristov et al. (2010) have yielded similar results. Longuski et al. (2009) also did not see an effect on ruminal pH for yeast culture supplemented cows during a fermentable starch challenge where diets containing a dry corn treatment were replaced with a high-moisture corn treatment for the final 2 d of each period. It was speculated that due a to large ruminal buffering capacity combined with low lactate concentrations, that yeast culture effects could not be detected (Longuski et al., 2009).

Correlations between yeast, protozoa population, and ruminal pH have been suggested by Galip (2006) and Brossard et al. (2006). Acidotic animals supplemented *Saccharomyces cerevisiae* tended to have an increased protozoal population and ruminal pH. Chaucheyras-Durand and Fonty (2002) found the enhanced growth of particular rumen ciliate Entodiniomorphid protozoal communities when yeast was supplemented in the diet of ruminants. Protozoa have previously demonstrated their ability to increase rumen pH by engulfing starch granules (Bonhomme, 1990; Williams and Coleman, 1988), enhance large populations of rumen bacteria (Williams and Coleman, 1997; Brossard et al., 2006), and prevent lactate accumulation by outcompeting lactate-producing bacteria, such as *Streptococcus bovis* (Newbold et al., 1987; Nagaraja, 2012).

Effects on Preventing Potential Health Risks and Milk Fat Depression

As previously mentioned, one of the primary reasons for supplementing yeast is to alleviate potential health concerns related to metabolic diseases and during times of stress. Subacute ruminal acidosis, more commonly known as SARA, is a common metabolic disorder that occurs in dairy cows fed an excess of rapidly fermentable carbohydrates (AlZahal et al., 2014). Subacute ruminal acidosis is characterized when the pH of the rumen is below 5.6 for approximately 300 min/d (AlZahal et al., 2007). Common clinical symptoms of SARA include a decrease in dry matter intake (DMI), resulting in poor body condition and reduced production (Plaizier et al., 2008), and may predispose cows to milk fat depression (AlZahal et al., 2009, 2010). Milk fat depression is caused by specific fatty acid (FA) intermediates of biohydrogenation of dietary polyunsaturated fat acids (**PUFA**) to saturated fatty acids (**SFA**) (Jenkins et al., 2008). Disruption of key lipogenic enzymes are downregulated for their gene expression and lead to the disruption in milk fat production (Harvatine and Bauman, 2006). Bauman and Griinari (2003) found that these specific FA are only produced during periods when ruminal fermentation has been altered and are referred to as conjugated linoleic acids (CLA). The specific CLA isomers responsible for a diet-induced milk fat depression are trans-10, cis-12 CLA (Baumgard et al., 2000), trans-9, cis-11 CLA (rumenic acid) and cis-10, trans-12 CLA (Bauman et al., 2008). Longuski et al. (2009) reported findings that indicate a supplementation of yeast culture may help in preventing milk fat depression when cows are transitioning to a diet with highly fermentable starch. However, the mechanism by which this occurs remains unknown. Although no treatment effects were observed for FA less than C16 as a percentage of total milk FA, an increase

in C16:0 (28.4 versus 27.7%, P < 0.01) and decrease in total C18:0 FA (37.8 versus 38.9%, P = 0.10) was detected for high-moisture corn compared with dry corn (Longuski et al., 2009). Hristov et al. (2012) reported that overall milk FA composition was not altered by yeast supplementation (Diamond V Mills Inc., Original XP, fed at 56 g/hd/d). A significant, but numerically small reduction (P = 0.03) of C16:0 content and an increase in C18:0 were observed in the yeast supplemented treatment (Hristov et al., 2010). Bayat et al. (2015) results were in agreement with Hristov et al. (2010) and demonstrated little influence on milk FA composition when two treatments of highly concentrated, live yeast were supplemented (0.5 g/hd/d at 10¹⁰ cfu/d) in the diet of lactating dairy cows. One yeast treatment lowered (P < 0.05) milk cis-10 C16:1 concentration, while neither yeast treatment had an effect (P > 0.05) on the distribution of milk fat C18:1 isomers, C18:2 or CLA isomer concentrations, and odd- and branchedchain FA concentrations. Instead, these treatments yielded similar results to the control diet fed, suggesting that live yeast strains fed had no major influence on ruminal lipolysis, biohydrogenation, or microbial lipid synthesis (Bayat et al., 2015).

Effects of Feeding Yeast During Heat Stress

Potential benefits in feeding yeast cultures during times of heat stress have been examined in lactating dairy cows (Schingoethe et al., 2004; Shwartz et al., 2009; Salvati et al., 2015). Temperature, relative humidity, solar radiation, air movement, and precipitation are some of the environmental factors that can cause heat stress in dairy cattle (Bohmanova et al., 2007). Roenfeldt (1998) described the 'thermoneutral' zone of a lactating dairy cow to be ambient temperatures of 5 to 25°C. When temperatures

exceed 26°C farm management practices should be altered to allow the cow to adequately lower her body temperature (Berman et al., 1985). Schingoethe et al. (2004) demonstrated an improved feed efficiency (P = 0.04) for cows fed 60 g of yeast culture (Diamond V XP yeast culture, Cedar Rapids, IA) to the total mixed ration (TMR) daily during a 12-wk period of high daytime temperatures (average = 33°C). Other areas of production, such as overall milk yield and component yields have also been shown to increase in dairy cows under heat stress (Shwartz et al., 2009; Salvati et al., 2015). Shwartz et al. (2009) attributed the improved lactation performance to regulation of body homeothermia, rather than improved digestibility. Whereas, Salvati et al. (2015) reported improved lactation performance without changing intake or body tissue deposition. A treatment effect (P = 0.03) was detected for milk yield where cows supplemented yeast produced 1.3 kg/d more than cows without yeast. In addition, increases in energycorrected milk (ECM) and 4% fat-corrected milk (FCM) were observed in response to yeast, and protein and fat secretions also tended to respond positively (Salvati et al., 2015). Salvati et al. (2015) also noted that yeast supplementation facilitated heat dissipation because similar body temperatures were observed at lower respiratory frequencies.

However, other studies show that yeast supplementation does not always produce favorable lactation performance results during times of heat-stress. Shwartz et al. (2009) reported a decrease of DMI by 29%, milk yield, and milk component yield of protein (7%; P < 0.01) and lactose (5%; P < 0.01) in cows that were supplemented with yeast cultures and heat-stressed challenged in climatic chambers (18°C, 20% humidity). Schingoethe et al. (2004) found no statistical significance for cows fed control and yeast

culture diets, respectively, for milk production (34.9 and 35.4 kg/d; P = 0.75), 4% FCM (31.2 and 32.0 kg/d; P = 0.64), ECM (33.4 and 34.2 kg/d; P = 0.62), and DMI (23.1 and 22.1 kg/d; P = 0.38). Moallem et al. (2009) also demonstrated no significant differences in the milk fat and protein percentages.

Yeast cultures have also been shown to impact other parameters measured in the lactating dairy cow during periods of heat stress, such as blood metabolites, rumen fermentation, and digestibility. Salvati et al. (2015) demonstrated a treatment effect for plasma urea nitrogen (**PUN**) with yeast values having an increased value versus cows treated without yeast (16.7 and 14.8 mg/dL; P = 0.05) during heat-stressed conditions. However, Shwartz et al. (2009) demonstrated heat stressed yeast culture-fed cows tended (P < 0.10) to have lower PUN concentrations than control-fed cows (15.5 and 14.2) mg/dL). In addition, a glucose tendency (P = 0.09) was evaluated by Salvati et al. (2015) where cows supplemented yeast established a greater concentrations than those fed a control ration (62.9 and 57.3 mg/dL). In addition, rumen fermentation data of lactating dairy cows fed yeast during heat-stress was analyzed by Salvati et al. (2015). Positive effects of yeast supplementation on rumen function (Wallace, 1994) and fiber digestibility (Bitencourt et al., 2011; Ferraretto et al., 2012) were expected in Salvati et al. (2015), however, this mode of action did not mediate animal responses in this experiment. Total-tract digestibility of nutrients and rumen microbial yield did not respond to yeast, although the rumen fermentation profile was modified. Ruminal lactate (P = 0.02) and butyrate (P = 0.05) as proportions of ruminal organic acids were decreased by yeast, but no effects on other organic acids, ruminal pH, or protozoa content were detected (Salvati et al., 2015). Such parameters lack consistency when yeast is supplemented in the diet of a lactating dairy cow.

Effects on Performance and Production

A meta-analysis of 61 studies conducted by Poppy et al. (2012) found that under normal environmental conditions, milk production outcomes were improved by yeast product supplementation. Treatment with yeast culture to lactating dairy cow diets increased milk yield by 1.8 kg/d, 3.5% FCM by 1.61 kg/d, and ECM by 1.65 kg/d. Significant treatment effects were shown for milk fat yield (P = 0.009) and milk protein yield (P = 0.026) with 0.06 kg/d and 0.03 kg/d, respectively, being produced. Furthermore, an increase in DMI (0.62 kg/d; P = 0.003) during early lactation and a decrease in DMI (0.78 kg/d; P = 0.001) during late lactation was demonstrated for cows supplemented a yeast culture. An increase in DMI during early lactation provides a tool for producers to utilize in a group that normally struggles with health issues due to the innate nature of the transition period and stress burden posed on the animal. Erasmus et al. (1992) also demonstrated an increase in DMI by 1.4 kg/d, however results did not affect milk production yield and milk composition in cow's supplemented yeast. Other studies showed improvements in milk yield (P < 0.01), whereas yeast culture supplemented diets produced 1.2 kg more milk than those diets not containing yeast (Piva et al., 1993), and in milk components (Piva et al., 1993; Bitencourt et al., 2011). Other individual studies (Arambel and Kent, 1990), yielded no effects on DMI, milk yield or components when cows were supplemented with a yeast culture.

Difficulties in evaluating the effects of yeast supplementation have been identified to be strain-specific and dosage dependent (Desnoyers et al., 2009; Vohra et al., 2016). However, another potential factor affecting yeast supplementation results is the experimental methodologies and environmental conditions that vary greatly from one experiment to another (Desnoyers et al., 2009; Robinson et al., 2009). Although metaanalyses attempt to explain this variation through detailed analysis of quantifying experimental factors, these methods often do not account for interactions in the model and discard data that is deemed as outliers (Desnoyers et al., 2009). Therefore, assumptions are made about the reported experiments to justify inclusion in the model (Robinson and Erasmus, 2009). However, differences in design and length of studies upon which conclusions are made do exist. Ferraretto et al. (2012) described a 12 wk, completely randomized design in a continuous lactation trial with four treatments including varying levels of starch and yeast supplementation. No effect of dry matter intake (**DMI**) and milk production parameters were deemed significant. Erasmus et al. (1992) used a 75 d crossover design with 6 cows and two treatments (control and yeast) and found a DMI effect where cows supplemented with a yeast culture consumed more feed than those who were not. Other literature cites yeast supplementation findings during times of heat stress where lengths of trials differ substantially (Schingoethe et al., 2004; Shwartz et al., 2009; Salvati et al., 2015). Schingoethe et al. (2004) cited a feed efficiency effect for cow's supplemented yeast in 12 wk, randomized complete block design study. Whereas Shwartz et al. (2009) demonstrated no such effect for cows in a 28 d study period. No published literature of such parameters of study length included in a meta-analysis model exist, to the knowledge of the authors. Such findings would be of

interest to investigate a potential relationship among length of study and effect of yeast supplementation.

Summary of Literature and Research Justification

Feed additives, such as yeast (*Saccharomyces cerevisiae*), can be used to manipulate rumen function (Wiedmeier et al., 1987; Guedes et al., 2008; Marden et al., 2008), increase animal efficiency and performance (Schingoethe et al., 2004; Poppy et al., 2012), and minimize issues related to animal health (AlZahal et al., 2014) and the environment. By altering the ruminal environment and microbial population of the host animal, fermentation can be shifted to maximize the efficiency of feed utilization to further increase ruminant productivity while decreasing unfavorable characteristics, such as greenhouse gas emissions and inefficiencies in feeding (Nagaraja, 2012).

Improvements in fiber digestion, ruminal pH regulation, and microbial communities' ability to scavenge oxygen in the rumen lead to improved areas of production, seen in increased milk and component yields (Desnoyers et al., 2009; Bitencourt et al., 2011).

However, results have been inconsistent across studies and through numerous meta-analyses (Lescoat et al., 2000; Sauvant et al., 2004; Desnoyers et al., 2009). A variety of yeast products with contrasting effects are present in the marketplace due to the strain of yeast used, the concentration of the dose supplemented, and the manner in which the yeast is delivered in the diet (Vohra et al., 2016). Therefore, particular inspection through scientific research practices must be thoroughly investigated specific to the type and amount of yeast fed in a lactating dairy cow's diet.

The goal of this research is to evaluate supplementing a concentrated brewer's yeast (*Saccharomyces cerevisiae*) product in the diet of a lactating dairy cow compared to

a control ration with no yeast and a common, concentrated commercially available yeast supplement. This will be evaluated by examining effects on milk yield and composition, feed intake and efficiency, blood metabolites, rumen fermentation concentrations, and total tract digestibility. Additional objectives are to determine the effect of yeast supplementation on milk fatty acid concentration profiles. This is important because changes in fatty acid composition will give an indication to how yeast supplements are altering rumen fermentation, biohydrogenation, and thus milk composition production. It was hypothesized that diets containing a concentrated brewer's yeast supplement would increase milk and component yields, improve feed efficiency and nutrient utilization, and benefit rumen fermentation to yield a more productive and efficient dairy cow.

CHAPTER 2: EVALUATION OF SUPPLEMENTING BREWER'S YEAST TO LACTATING DAIRY COWS

INTRODUCTION

Yeast (Saccharomyces cerevisiae) has been fed to dairy cattle with varied responses on milk production (Desnoyers et al., 2009), rumen fermentation (Erasmus et al., 1992), and feed efficiency (Schingoethe et al., 2004). Yeast and other fungi are naturally present in the rumen of the cow's digestive tract, and are therefore, viewed as a more favorable option as a feed additive compared to antibiotics and ionophores (Yirga, 2015). Yeast product supplementation can be beneficial in the diet of lactating dairy cows by increasing feed efficiency (Schingoethe et al., 2004), alleviating disease (i.e. subacute ruminal acidosis – SARA; AlZahal et al., 2014), increasing energy-corrected milk (ECM) and milk fat yield (Poppy et al., 2012), and improving production performance under heat stress (Salvati et al., 2015). The ability to increase milk production, milk quality, and feed efficiency can provide environmental and economic benefits to the producer and consumer. In particular, dried brewer's yeast has been used as a viable by-product feedstuff in livestock systems to mimic the actions of naturally occurring ruminal yeast. Brewer's yeast advantages over other feed additives include its high palatability, low demand in the marketplace, and relatively inexpensive costs to feed as a by-product supplement (Hertrampf and Piedad-Pascual, 2000). However, to the knowledge of the authors, there is minimal literature specifically on the feeding of a concentrated, dried brewer's yeast product to lactating dairy cows.

Therefore, the main objective of this research was to evaluate supplementing a concentrated brewer's yeast (*Saccharomyces cerevisiae*) in the ration of lactating dairy

cows compared to a control ration with no yeast, and a common, concentrated commercially available yeast supplement. This will be evaluated by examining effects on milk yield and composition, feed intake and efficiency, blood metabolites, rumen fermentation concentrations, and total tract digestibility. Additional objectives are to determine the affect of yeast supplementation on milk fatty acid concentration profiles. It was hypothesized that diets containing a concentrated brewer's yeast supplement will increase milk and component yields, improve feed efficiency and nutrient utilization, and benefit rumen fermentation to yield a more productive dairy cow. We also hypothesized that since component yields will increase due to yeast supplementation that beneficial milk fatty acids will improve as well.

MATERIALS AND METHODS

All procedures and animal use were approved prior to the start of the trial by the South Dakota Institutional Animal Care and Use Committee.

Experimental Design

Thirty-six lactating Holstein cows (24 multiparous and 12 primiparous; days in milk (**DIM**) = 71.17 ± 16.42) were used in a randomized complete block design feeding study with three treatment diets. Two groups of 18 cows were used in the study due to availability of lactating cows. Cows were blocked in groups of three based on prior 7 d milk yield averages (kg/d), DIM, and lactation number. Cows were then randomly assigned to treatment within blocks. Cows spent two weeks prior (door training and covariate) to the 8 wk feeding period in order to adjust to the barn and Calan gate feeding system.

Treatment diets were: 1) control diet with no yeast (CON), 2) concentrated brewer's yeast product 1 (Y1), 3) concentrated commercial yeast product 2 (Y2). Yeast supplements were fed at a rate of 14.2 g/hd/d. Diets were formulated using AMTS. v 4.1.4.0. to meet a target milk yield of 41.0 kg/d with a 3.70% milk fat and a 3.15% milk protein, and a predicted DMI of 25.9 kg/d. The target milk yield and composition, and predicted DMI were used to support high producing lactating dairy cows. The amount of each ration offered was adjusted weekly using DM analysis of feedstuffs.

Animal Care and Feeding

The farm study was conducted and all cows were housed at the South Dakota State University Dairy Research and Training Facility (SDSU DRTF) in Brookings, South Dakota. The study was completed from March 29 – August 23, 2016 to accommodate available animals and finish the study with 18 cows in both groups. Cows were observed daily for health problems and were treated according to standard SDSU DRTF management practices.

Cows were housed in a group pen within a barn containing freestalls with rubber mattresses that were bedded daily with chopped straw. The pen was scraped and cleaned during each milking period, according to SDSU DRTF management practices. The pen was provided with water ad libitum, and had sprinklers and fans for cooling cows. Cows were fed using the Calan Broadbent gates and box system (American Calan Inc., Northwood, NH) to monitor and determine daily individual intakes. Diets were fed as a total mixed ration (TMR) and were fed once daily at 0800 h using a Calan Data Ranger (American Calan Inc., Northwood, NH) in amounts to allow for ad libitum consumption. Individual weighbacks (orts) were collected daily and were used to determine the amount

of individual TMR fed to each cow, targeting a 10% refusal rate. Treatment diet ingredient composition is shown in Table 1. A forage mix of corn silage, alfalfa haylage, and whole cottonseed was combined in a vertical mixer wagon (Patz 1200 Series Trailer TMR Vertical Mixer, Patz Corporation, WI) and a grain mix, mixed at the South Dakota State University Feed Mill, was added to the mixer. The grain mix contained ground corn, soybean meal, dried distillers grains with solubles (**DDGS**), ground soy hulls, Energy Booster 100, salt, calcium carbonate, sodium bicarbonate, magnesium oxide, urea 281%, trace mineral and vitamin premixes, and soybean oil. Treatment mixes contained: 1) DDGS for the CON diet, 2) DDGS with 14.2 g/hd/d of Y1 treatment yeast for the Y1 diet, and 3) DDGS with 14.2 g/hd/d of Y2 treatment yeast for the Y2 diet. Treatment mixes were blended at the South Dakota State University Feed Mill, where DDGS were used in combination as a carrier with the yeast supplements. A basal diet was formulated (Table 2) with the forage and grain mixes, and specific treatment mixes were individually weighed, added to the TMR, and mixed by a Calan Data Ranger. No recombinant bovine somatotropin (**rBST**) or ionophore supplementation, such as Rumensin, were used.

Animal Sampling

Feed intakes and orts for individual cows were recorded once daily at 0830 h.

Dry matter concentration of corn silage, alfalfa haylage, and whole cottonseed was determined weekly by drying samples for 24 h at 105°C. Diets were promptly adjusted to maintain a constant forage to concentrate ratio throughout the experiment. Samples of basal TMR, corn silage, alfalfa haylage, whole cottonseed, grain mix, CON treatment mix, Y1 treatment mix, and Y2 treatment mix were collected weekly during the study and

stored at -20°C until further analysis. At the end of the study, individual feed samples were composited equally by month and period to create six total samples per feedstuff for nutrient analysis. Additional TMR samples were obtained once per week to determine particle size and using the Penn State Particle Separator (Kononoff and Heinrichs, 2003).

Body measurements of body weight (**BW**) and body condition score (**BCS**) were taken weekly. During covariate and wk 8, BW and BCS were collected on two consecutive days. Three trained individuals assessed the BCS of cows based on the scale described by Wildman et al. (1982), where 1=emaciated and 5=obese. Cows were milked twice per day in a double-8 parallel parlor at 530 and 1730 h, and daily milk weights were electronically recorded (ALPROTM, DeLaval, Sweden). Milk from individual cows was sampled weekly at each milking, except during covariate and wk 8 where milk samples were collected for two consecutive days. Samples were taken for component analysis by Dairy Herd Improvement Association (DHIA; MQT Lab Services, Kansas City, MO) and fatty acid composition determination. The milk samples for fatty acid analysis were stored at -20°C until further analysis.

Rumen fluid was sampled from each cow on 2 d during wk 7 and 8, approximately 4 h post-feeding via esophageal tubing. The initial 200 ml of fluid collected was discarded due to concerns of contamination by the water-bleach cleaning solution used to rinse the pump and saliva. Thereafter, 50 mL of rumen fluid was collected and immediately measured for pH using a handheld pH meter (Waterproof pH Testr 30, Oakton Instruments, Vernon Hills, IL), and 2 aliquots (10 mL each) were acidified with either 200 µL of 50% (volume/volume) sulfuric acid or 2 mL of 25% (weight/volume) meta-phosphoric acid. Vials of acidified rumen fluid samples were

stored at -20°C until later analyses of ammonia N (NH₃-N) and volatile fatty acid (VFA) analysis.

Blood samples were collected from each cow on 2 d during wk 7 and 8, approximately 4 hour post-feeding by venipuncture of the coccygeal artery. Blood was drawn into 10-mL vacutainer tubes containing a serum separator tube (BD Vacutainer® SSTTM Gel Separator Tube; Becton, Dickson, and Co.) for plasma urea nitrogen (**PUN**), glucose, and cholesterol determination. Blood samples were centrifuged at 1000 x *g* for 20 min at 5°C (CR412 centrifuge; Jouan Inc., Winchester, VA), and plasma samples were stored at -20°C for further analysis.

Fecal grab samples were collected from each cow on 2 d during wk 7 and 8, approximately 4 h post-feeding via fecal grab samples to determine total tract digestibility. Acid detergent insoluble ash (**ADIA**) was used as an internal digestibility marker. Samples were placed in bags (9.5 x 17.8 cm; Nasco Whirl-Pak® Standard Bag, WI) and were stored at -20°C until further processing and analysis.

Laboratory Analysis

Forage (corn silage, alfalfa haylage, whole cottonseed), concentrate (grain mix, CON treatment mix, Y1 treatment mix, and Y2 treatment mix), and TMR samples were each composited by period (1 or 2) and month (C=covariate, 1=wk 1-4, and 2=wk 5-8). Samples were then dried for 48 h at 55°C in a Despatch oven (Style V-23, Despatch Oven Co., Minneapolis, MN), and were ground to a 4 mm particle size using a Wiley Mill (model 3; Arthur H. Thomas Co., Philadelphia, PA). Further grinding to a 1 mm particle size was done using an ultracentrifuge mill (Brinkman Instruments Co., Westbury, NY). To correct analysis to 100% DM, aliquots of feed samples were dried for 4 h in a 105°C

muffle furnace. Ash content was determined by incinerating a 1 g sample for 8 h at 450°C in a muffle furnace (AOAC 17th ed., method 942.05; 2002). Organic matter (**OM**) was calculated as OM = (100 - % Ash). Samples were analyzed for nitrogen content via Dumas combustion analysis (AOAC 2002, method 968.06), on a Rapid N Cube (Elementar Analysensysteme, GmbH, Hanau, Germany). Nitrogen content was then multiplied by 6.25 to calculate crude protein (**CP**). Neutral detergent fiber (**NDF**; Van Soest et al., 1991) and acid detergent fiber (ADF; Robertson and Van Soest, 1981) were analyzed sequentially using the Ankom 200 fiber analysis system (Ankom Technology Corp., Fairport, NY). For NDF, heat-stable bacterial α-amylase, sodium sulfite, and a neutral detergent solution were used. For ADF, an acid detergent solution was used. Lignin was also determined sequentially on the ADF residue (Van Soest, 1963). Ether extract (EE) was determined using the Ankom Extraction System (Ankom Technology Corp., Fairport, NY) with petroleum ether as the solvent (AOAC 2002, method 920.39). Non-fibrous carbohydrate was calculated as % NFC = 100 – (% Ash + % CP + % NDF + % EE), according to the NRC (2001).

Dried and ground samples of corn silage, alfalfa haylage, whole cottonseed, grain mix, CON treatment mix, Y1 treatment mix, and Y2 treatment mix were further composited into wk 1-8 composites by group and sent, along with original TMR composites, to a commercial laboratory (Dairyland Laboratories, Inc., Arcadia, WI) for analysis of starch, minerals (Ca, Cl, Mg, P, K, Na, S, Fe, Mn, Mo, and Zn), and dietary cation-anion difference (**DCAD**). Starch was determined by a modified procedure analyzing glucose using YSI Biochemistry Analyzer (YSI Inc., Yellow Springs, OH; Bach Knudsen, 1997). Mineral content, excluding chloride, was determined using

inductively coupled plasma spectroscopy (AOAC International, 1995). Chloride content was determined using a direct reading chloride analyzer (Corning 926, Corning Inc., Corning, NY). The DCAD equation used was DCAD = $(Na^+ + K^+) - (Cl^- + S^{2-})$ (Ender et al., 1962; Block, 1984).

Total mixed ration composites were analyzed for feed fatty acid composition by modification of the Sukhija and Palmquist (1988) methods as first described by Abdelquader et al. (2009). Composited and ground TMR samples were weighed such that 20 to 25 mg of lipid from the feed was contained in 750 µL n-butanol. An internal standard of C19:1 (~20 mg/mL) was used for ease of peak identification for each sample. Acetyl chloride was added to the reaction mixture while vortexing to ensure separation of the fatty acid chain from the glycerol molecule. Nitrogen gas was used to prevent oxidation and samples were placed on a 60°C heating block for 90 m. A 6% potassium carbonate solution and hexane were added to each sample once removed and cooled from the heating block. Potassium carbonate was used as an alkaline buffer to stop the acid hydrolysis. Hexane was used to contain the butyl esters of the fatty acids. A series of washings and centrifugation for 20 m at 2000 rpm yielded a layer of hexane and fatty acids that were pipetted into gas chromatography vials for analysis. An automated gas chromatography (model 6890; Hewlett-Packard Co., Palo Alto, CA) was used to analyze the fatty acids of the TMR according to carbon numbers and unsaturated bonds. All prepared fatty acid samples were analyzed via GC (Hewlett Packard 6890, Palo Alto, CA) as described by Abdelquader et al. (2009).

Milk samples were sent weekly to Heart of America DHIA Laboratory (Kansas City, MO) for composition analysis. During covariate and wk 8, milk samples were

collected on two consecutive days and sent to DHIA (MQT Lab Services, Kansas City, MO). Fat, protein, and lactose were analyzed via mid-infrared spectroscopy (AOAC, 2006; Bentley 2000 Infrared Milk Analyzer, Bentley Instruments, Chaska, MN). Milk urea nitrogen (MUN) was determined using a modified Berthelot reaction (ChemSpec 150 Analyzer, Bentley Instruments, Chaska, MN). Somatic cell count (SCC) was analyzed using laser technology (Soma Count 500, Bentley Instruments, Chaska, MN). Milk fatty acid samples were thawed and composited by day in accordance with individual cow daily milk weight concentrations. A modification of Sukhija and Palmquist (1988) methods were used in a butylation and separation of fatty acids by gas chromatography (model 6890; Hewlett-Packard Co., Palo Alto, CA) as described by Abdelqader et al. (2009). Weekly milk samples were composited by cow and week into 10 mL vials based on the weighed volume of the day's milking. Samples were prepared as previously described for feed fatty acids, except the internal standard of C13:1 (~20 mg/mL) was used.

Rumen fluid samples preserved with sulfuric acid were thawed and centrifuged at $30,000 \times g$ for 20 minutes at 4°C (Centrifuge: Eppendorf 5403, Eppendorf North America, Hauppauge, NY), and were analyzed for ammonia N using a colorimetric assay performed on a micro-plate spectrophotometer (Cary 50, Varian Inc., Walnut Creek, CA), according to Chaney and Marbach (1962). The rumen fluid samples that were preserved with meta-phosphoric acid were thawed and centrifuged at $30,000 \times g$ for 20 min at 4°C and, were analyzed for the following VFA concentrations: acetate, propionate, butyrate, isobutyrate, isovalerate, and valerate. Concentrations were measured using an automated GC (model 6890; Hewlett-Packard Co., Palo Alto, CA) using a flame-ionization detector.

Volatile fatty acids were separated on a capillary column (15 m \times 0.25 mm i.d.; Nukol, 17926-01C; Supelco Inc., Bellefonte, PA) using 2-ethylbutyrate as an internal standard. The split ratio of 100:1 in the injector port was at a temperature of 250°C with flow rate of 1.3 mL/min of helium. The column and detector temperature were maintained at 140°C and 250°C, respectively.

Plasma samples were thawed, vortexed, and analyzed for PUN, glucose, and cholesterol concentrations using commercially available enzymatic or colorimetric assay kits on a microplate spectrophotometer (Cary 50, Varian Inc., Walnut Creek, CA). Diacteyl monoxime was used to analyze PUN (procedure 0580, Stanbio Laboratory, Boerne, TX). Glucose was determined by the glucose oxidase reaction (Trinder, 1969), using a glucose kit (Catalog #: G7521, Pointe Scientific, Inc., Canton, MI). Cholesterol concentrations were analyzed using a cholesterol esterase reagent (Catalog #: C7510, Pointe Scientific, Inc., Canton, MI).

Fecal samples for each cow were composited by week and period on an as-is basis by volume. Samples were dried and ground, as previously described for feed samples. Fecal samples were analyzed in the same manner as described for feeds for DM, ash, CP, NDF, ADF, and lignin. Acid detergent insoluble ash (ADIA) was used as an internal marker and analysis was conducted on all fecal samples and only month 2 composites of periods 1 and 2 TMR feed samples. The method for ADIA analysis consists of analyzing the sample for ADF content (Robertson and Van Soest, 1981) and then determining the ash content using a modified procedure of the AOAC 17th ed., method 935.29 (2002). Digestibility calculations were determined according to Merchen (1988).

Statistical Analysis

All data were analyzed using SAS version 9.4 (SAS Institute Inc., Cary, NC). Analyzed feedstuffs, including feed fatty acids, and particle distribution using the PSPS were evaluated using the PROC MEANS in SAS procedure to calculate the mean and standard error of the mean for individual nutrient contents. Lactation performance data and milk fatty acid data were analyzed as a randomized complete block design experiment with week as the repeated measure and cow (block) as the subject using the MIXED procedures of SAS (Littell et al., 2006). The model included treatment, week, group, and the interactions of all terms. Energy-corrected milk (ECM) were calculated by using the following equation: ECM = [(0.327 x kg milk) + (12.95 x kg fat) + (7.2 x kg)]protein)] (Orth, 1992). Feed efficiency was calculated by ECM/DMI, where DMI=dry matter intake. Akaike's criterion was used to determine the most suitable covariance structure in repeated measures for each parameter. Covariance structures tested were compound symmetry, first-order autoregressive, Toeplitz, and unstructured. Compound symmetry resulted in the least absolute Akaike's values, and was used for the final model. Blood metabolite data, rumen fermentation parameters, and total tract digestibility analysis used the MIXED procedures of SAS. The model included treatment, group, and the interaction of both terms.

Environmental temperature conditions were evaluated and data was downloaded from a weather station located in northeast Brookings, S.D. (High Plains RCC CLIMOD).

All data are presented as least square means with the highest standard error of the mean (**SEM**) among the values. Significant differences among treatments were declared at $P \le 0.05$ and tendencies were declared at $0.05 < P \le 0.10$.

RESULTS AND DISCUSSION

Feed Analysis

The analyzed nutrient composition of the individual ingredients is presented in Table 3. Values are comparable to those listed in the NRC (2001) for the same feedstuffs. Since treatment mixes were purchased in one batch at the beginning of the study, there was little variation in nutrient composition over the duration of the study. In addition, there was very little variation seen in the grain mix and forages. The formulated basal TMR diet was similar to the analyzed nutrient composition of the ration fed (Table 4). Concentrations of ash, OM, ether extract, lignin, hemicellulose, NFC, and starch were similar and had a very small standard errors (SE). The analyzed DM of the basal TMR (45.3%) was numerically less than the formulated concentration of 50.3%. The CP value analyzed was less than that formulated, however, levels still met and exceeded lactating dairy cow feeding requirements (NRC, 2001). Both concentrations of analyzed fiber, ADF and NDF, in the basal TMR were greater than the parameters in the formulated diet. Differences in the mineral composition of diets formulated versus analyzed were minimal, except for concentrations of Mn, Zn, and Fe, however, these differences are unlikely to have significant effects on lactation performance.

Particle size of the CON TMR measured with the Penn State Particle Separator (**PSPS**) demonstrated little variation overall (Table 5). However, percentages of particle

sizes analyzed do not adhere to the PSPS guidelines of 2-8%, 30-50%, 30-50%, and <20% for the upper sieve (19 mm), middle sieve (8 mm), lower sieve (1.18 mm), and the bottom pan, respectively (Kononoff and Heinrichs, 2003). The most particles were retained on the 1.18 mm screen and bottom pan, measuring 33.5% and 30.7%, respectively. However, the upper sieve retained a greater amount of feed than recommended guidelines suggest with 26.8%. Mertens (1997) describes physically effective fiber (**peNDF**) as the dietary fiber source which effectively stimulates rumination and salivation, and has been suggested to be determined by feed particles retained on the 1.18 mm screen. Particles retained on the 1.18 mm sieve pass out of the rumen slower than those which are not (Poppi et al., 1985). The increased percentages retained for the upper sieve and bottom pan may indicate that the NDF in the diet was not adequately utilized to support an increased rumen activity by the yeast additive due to an increased passage rate allowing the yeast to be quickly washed out of the rumen (Kung, et al., 1997).

Lactation Performance

Lactation production responses to supplementation of yeast in the diets are presented in Table 6. No significant treatment effects were observed for any of the animal performance measures. In addition, no treatment by group interactions were found. Week effects were determined for every production parameter, due to changes observed in a normal lactation curve, except SCC (P = 0.14). A treatment by week interaction was observed for DMI (P = 0.01) with cows fed Y1 consuming the most feed (24.6 kg/d), compared to CON and Y2 cows (24.2 kg/d and 24.1 kg/d, respectively). In Figure 1, DMI is presented during the 8 wk feeding period. Although statistically

significant, DMI between the three treatments were numerically similar, indicating that week-by-week changes were occurring during the trial. This increase in DMI of 0.4 kg/d has been reported in similar studies analyzing the potential benefits of adding yeast cultures to dairy cattle diets (Williams et al., 1991; Erasmus et al., 1992; Bittencourt et al., 2011). A review by Yoon and Stern (1995), indicated significant increases in DMI in 2 of 10 studies where Saccharomyces cerevisiae was supplemented. Dann et al. (2000) and Wohlt et al. (1991) observed increases in DMI during early lactation, which aided in greater milk yields through wk 18 of lactation. This increase in DMI could be due to the modes of action demonstrated by yeast cultures in the rumen (Yoon and Stern, 1996; Newbold et al., 1996). Yeast added to rumen fluid *in vitro* demonstrated an ability to increase the rate of oxygen removal (Newbold et al., 1996). A removal of oxygen aids in the anaerobic ruminal process of fermentation, thus increasing the efficiency of (Jouany, 2001) and the viable count of bacterial communities (Newbold et al., 1996). Some of the bacteria that benefitted from this environment include those that are strictly anaerobic, such as cellulolytic bacteria (Seo et al., 2010). Therefore, cellulolytic digesting bacteria are utilized at a greater capacity, and can potentially cause a greater DMI for cows consuming yeast products.

Cows fed the CON diet demonstrated an increased milk production of 1.1 kg/d compared to diets supplemented with yeast with a treatment by week interaction (P = 0.04). Figure 2 shows milk production by week during the feeding period. Fluctuations of milk yield for all treatments demonstrated during wk 2 through 4 are thought to be due to temperature changes experienced by the two groups, which may have caused heat stress. Daily maximum and minimum temperatures during group 1 and group 2 trial

periods are illustrated in Figure 3 and Figure 4, respectively. As demonstrated in Figure 3, more temperature variation of both highs and lows are shown to have occurred throughout the feeding period of group 1 compared to group 2. However, group 2 spent the entirety of its trial period occurring during the summer months, subjecting the cows to an extended period of time in constant heat. Cows in group 2 were subjected to greater maximum and minimum temperatures of 27.0°C and 15.5 °C, respectively, when compared to group 1. Temperatures exceeding 26°C for prolonged periods of time, in addition to increased humidity, lack of air movement, and other factors, have been demonstrated to cause heat stress in dairy cattle (Berman et al., 1985; Bohmanova et al., 2007). Therefore, it is possible to deduce that cows in group 2 were subjected to heat stress conditions. Although increases in lactation performance of dairy cows under heat stress due to yeast supplementation have been shown (Salvati et al., 2015), such results were not demonstrated in this study. However, this potential impact of temperature led to examining group effects and treatment by group interactions in the remainder of the data analyzed. Figure 5 presents overall milk production by treatment for groups 1 and 2. A group effect for milk production (P = 0.03) is detected where group 1 produced 2.5 kg/d more milk than group 2 (35.0 kg/d versus 32.5 kg/d). Since greater milk yield and environmental temperatures were reported for group 1 and group 2, respectively, it is plausible to reason that group 2 cows were experiencing heat stress, and thus supplementation of yeast could have aided in a more efficient production and provided benefits in body homeostatic conditions. Treatment by group interactions were not statistically significant. Therefore, we cannot definitively suggest that feeding yeast to

lactating dairy cows during times of heat stress can improve or inhibit overall milk production. Rather, these probiotic products maintain milk yield performance.

There were no significant treatment differences found for the milk components. There were treatment by week effects for protein yield (kg/d), lactose yield (kg/d), and solids-not-fat (SNF; kg/d) (P < 0.05). This effect could be due the relationship of the three parameters and the presence of lactose, caseins, and whey proteins, along with minerals (ash) found in milk SNF. Group tendencies for milk fat percentage, protein (kg/d), and lactose (kg/d) were detected (P < 0.10), as well as group effects for SNF percentage and kg/d (P < 0.01). Milk urea nitrogen (MUN) and SCC were not significantly different among treatments or groups.

Energy-corrected milk (**ECM**) was greater for cows fed the CON diet compared with those fed Y1 and Y2. However, there were no differences among treatments (P > 0.10). Figure 6 shows feed efficiency for the three diets fed. Due to differences in DMI, statistically significant effects of treatment and week were found for feed efficiency (P < 0.01). Cows fed both the CON and Y2 diets demonstrated slightly increased feed efficiency (1.51) when compared to the Y1 diet (1.36). Schingoethe et al. (2004) reported an increased feed efficiency for heat stressed cows fed a yeast culture. However, other heat stress related research has not concluded such findings (Salvati et al., 2015).

Body weights demonstrated a group (P = 0.03) effect and in all treatments showed cows gaining weight throughout the trial from initial to final. Body condition scores (**BCS**) did not have any statistically significant differences.

The fatty acid (FA) profile of the basal TMR fed is shown in Table 7. The basal TMR contained more long chain fatty acids (C17:0 to C22:6) compared to medium and short chained FA. Contributing to the concentration of the long chain fatty acids were C16:0 and C18:2 cis-9, 12, which were the greatest amount of FA found in the feed with and 16.1 mg/100 mg FA and 33.1 mg/100 mg FA, respectively. These FA profiles are consistent with those found by the United States Department of Agriculture (USDA, 1998), demonstrating the high amount of C16:0 and C18:2 found in "vegetable oil" related sources supplemented in the experimental ration, such as corn, cottonseed, and soybean. Milk FA concentrations of the cows fed the CON, Y1, and Y2 treatment diets are presented in Table 8. Most milk FA were unaffected by treatment, group, and the interaction of the terms. No effect of treatment was observed for FA less than C16:0, but a group effect (P = 0.05) was demonstrated for C16:0. This is in agreement with Longuski et al. (2009) and Hristov et al. (2010) for milk FA profiles of cows' supplemented yeast. In addition, group effects (P < 0.05) were found for C8:0, C10:0, C16:1 trans-9, C18:3 n-6, and conjugated linoleic acid (CLA) trans-10, cis-12, and are thought to be due to variation in temperature. A treatment effect (P = 0.05) for CLA cis-9, trans-11 was detected where the Y1 diet showed a greater concentration of FA compared to CON and Y2. The main trans C18:1 isomer is vaccenic acid (C18:1 trans-11) and it serves as the main precursor for CLA cis-9, trans-11 (Månsson, 2008). No significant effects were found for vaccenic acid and no indications of milk fat depression were determined in total milk fat concentrations, indicating that the amount of CLA in the milk was not high enough disrupt milk production. Numerous week effects were determined for milk FA concentrations, and could be attributed to various factors related

to the state of lactation, genetics (breed), presence of mastitic infection, ruminal fermentation, or feed consumed (Palmquist et al., 1993; Jensen, 2002). However, no FA had a significant treatment by week interaction.

Plasma Metabolite Profile

Concentrations of plasma metabolites are presented in Table 9. Tendencies for glucose were detected for treatment (P = 0.07) and group (P = 0.09). In addition, a strong treatment by group interaction effect (P < 0.01) was observed for cows on yeast supplementation, which demonstrated a greater plasma concentration of glucose compared to the CON diet. Figure 7 shows the glucose concentrations for groups 1 and 2 by treatments. Control diets demonstrated similar plasma glucose concentrations between groups, while differences in the yeast treatments by groups were noted. An increase in plasma glucose concentration has been reported when dairy cows under heat stress were supplemented with live yeast (Dehghan-Banadaky et al., 2013). Others stated no differences in glucose concentrations (Piva et al., 1993; Putnam et al., 1997). However, Rhoads et al. (2009) demonstrated the cows' reliance on glucose as an essential energy source when experiencing heat stressed conditions by providing increased glucose availability to the mammary gland for milk production. Plasma urea nitrogen (PUN) values exhibited a treatment effect (P < 0.01) and a treatment by group tendency (P =0.10). Cows fed both Y1 and Y2 diets demonstrated slightly decreased PUN concentrations (14.7 mg/dL and 16.0 mg/dL, respectively) compared to CON fed cows (17.2 mg/dL). This is consistent with results reported by Bitencourt et al. (2011), where PUN concentrations sampled 2 h post-feeding were numerically less than a control diet with no yeast. Salvati et al. (2015) found that inclusion of yeast increased PUN (P =

0.05), rather than decreasing as we saw. However, this relationship is based on a weak statistical threshold to be considered as a repeatable effect, and therefore, is not considered highly significant to the authors. There were no differences in concentrations of cholesterol, which is a steroid hormone precursor.

Rumen Fermentation

Rumen fermentation characteristics are presented in Table 10. There was a group (P = 0.02) and treatment by group (P = 0.03) effect for ammonia with Y1 treatment cows having the greatest concentration. Treatments did not influence rumen pH and rumen ammonia N (NH3-N), which is in agreement with Piva et al. (1993), Putnam et al. (1997), and Erasmus et al. (2005). A meta-analysis by Desnoyers et al. (2009), demonstrated a positive effect of yeast supplementation on rumen pH which was due to increases of DMI in diets that had greater proportions of concentrates.

Group effects (*P* < 0.05) were detected for molar concentrations of acetate, propionate, and total volatile fatty acids (**VFA**), where yeast supplemented diets presented greater concentrations than the CON diet, as well as percentage of valerate. Several studies are in agreement with this finding and have demonstrated an increase in production of acetate, propionate, and total VFA when dairy cows were supplemented with *Saccharomyces cerevisiae* (Nisbet and Martin, 1991; Piva et al., 1993; Miller-Webster et al., 2002). Lynch and Martin (2002) have also shown similar results in a 48 h *in vitro* study. A greater propionic acid production would be expected to have a positive effect on milk yield for cows fed yeast products (Cakiroglu et al., 2010), which could help explain the group effect seen in milk production. Propionate is used by the cow to make glucose, which is a necessary precursor for lactose production and milk yields

(Cakiroglu et al., 2010). A greater rumen VFA concentration can be due to a greater DMI in cows supplemented with yeast (Desnoyers et al., 2009). In addition, a group tendency and effect for butyrate (P = 0.07) and valerate (P = 0.01) percentages, respectively, are present along with treatment by group interaction effects (P < 0.05) for molar concentrations and percentage of valerate. Changes in valerate concentration with yeast supplementation is difficult to explain and has thought to have little biological significance (Hristov et al., 2010). No differences were detected among treatment for pH and acetate:proprionate ratio. A non-significant acetate:propionate ratio could indicate that although molar concentrations of propionate were significant enough for a group effect, they were not large enough relative to the production of acetate to yield an acetate:propionate ratio difference.

Total Tract Nutrient Digestion

Total tract nutrient digestibility is presented in Table 11. Digestibility values are high compared to other published literature (Wohlt et al., 1991). The method of one-time per day fecal grab sampling approximately 4 h post-feeding could have affected the validity and plausibility of these results. There were no differences in digestibility among treatments or treatment by group for any of the nutrients measured. This is consistent with both *in vitro* (Arambel and Kent, 1990; Wohlt et al., 1991; Bitencourt et al., 2011) and *in situ* (Doreau and Jouany, 1998) work. Other sources of literature suggest that *Saccharomyces cerevisiae* supplementation and dosage do increase digestibilities, such as organic matter (Desnoyers et al., 2009; Ferraretto et al., 2012; Leicester et al., 2016) and crude protein (Wohlt et al., 1998; Leicester et al., 2016). While Williams and Newbold (1990) suggests that yeast supplementation may alter the site of nutrient digestion,

leading to inaccuracies in total-tract digestibility determination. However, there were group effects (P < 0.05) detected for all nutrient digestibilities measured. For all nutrients analyzed, group 1 cows had greater digestibilities than group 2. Greater total-tract nutrient digestibilities for group 1 may have been related to the higher temperatures and the challenges associated with potential heat stress experienced by group 2 cows (Kadzere et al., 2002). However, Moallem et al. (2009) and Salvati et al. (2015) found no statistical treatment differences for total-tract digestibility of nutrients in heat stressed cows.

CONCLUSION

Yeast products maintained performance, rather than improving production as originally hypothesized. Milk yield fluctuations by group and week are thought to be due to temperature variability experienced by the two groups during times of potential heat stress, especially for group 2 cows where average maximum temperatures exceeded the cow's thermoneutral zone. Due to effects of DMI increases and milk yield decreases by cows supplemented yeast products, statistically significant effects of treatment by week were found for feed efficiency. There were no differences in milk components, BCS, and body weight. The milk fatty acid profile was not influenced by treatment or group, however, numerous week effects were shown and are thought to be associated with factors related to stage of lactation, or adjustment to ingredients in the basal diet. A shift in the metabolic profile was demonstrated, but cows maintained production and it was not enough to alter performance. Group effects of increases in propionate production are thought to influence the improvement of glucose concentrations demonstrated by yeast

supplemented diets. This research demonstrates that producers can supplement yeast to lactating dairy cows without any adverse effects.

Figure 1. Dry matter intakes (kg/d) for cows fed no yeast (**CON**), a concentrated brewer's yeast product (**Y1**), and a concentrated commercial yeast product (**Y2**).

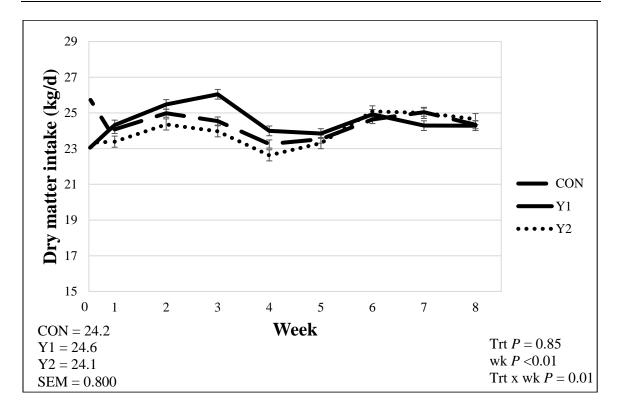
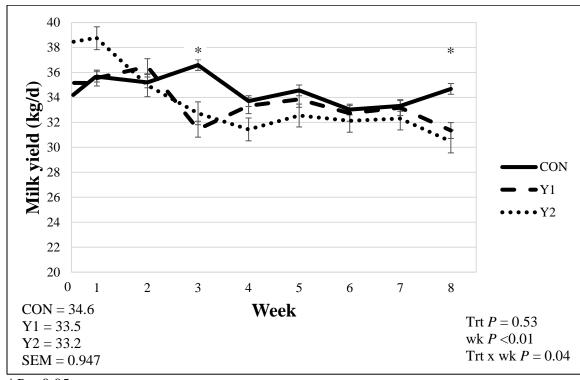


Figure 2. Milk production (kg/d) for cows fed no yeast (**CON**), a concentrated brewer's yeast product (**Y1**), and a concentrated commercial yeast product (**Y2**).



**P* < 0.05

40 35 30 25 Temperature, °C 20 15 Max Temp 10 • Min Temp 5 0 8 11 14 17 20 23 26 29 32 35 38 41 44 47 50 53 -5 -10 **Days of Trial**

Figure 3. Daily maximum¹ and minimum² temperatures during group 1 feeding.³

¹Maximum average temperature = 20.3°C

 $^{^{2}}$ Minimum average temperature = 8.1 $^{\circ}$ C

³Dates of group 1 feeding period: April 12, 2016 – June 13, 2016.

40 35 30 Temperature, °C 20 15 Max Temp 10 •• Min Temp 5 5 8 11 14 17 20 23 26 29 32 35 38 41 44 47 50 53 -5 -10 **Days of Trial**

Figure 4. Daily maximum¹ and minimum² temperatures during group 2 feeding.³

 $^{^{1}}$ Maximum average temperature = 27.0°C

²Minimum average temperature = 15.5°C

³Dates of group 2 feeding period: June 21, 2016 – August 22, 2016.

Figure 5. Milk production (kg/d) for group 1 and group 2 cows fed no yeast (**CON**), a concentrated brewer's yeast product (**Y1**), and a concentrated commercial yeast product (**Y2**).

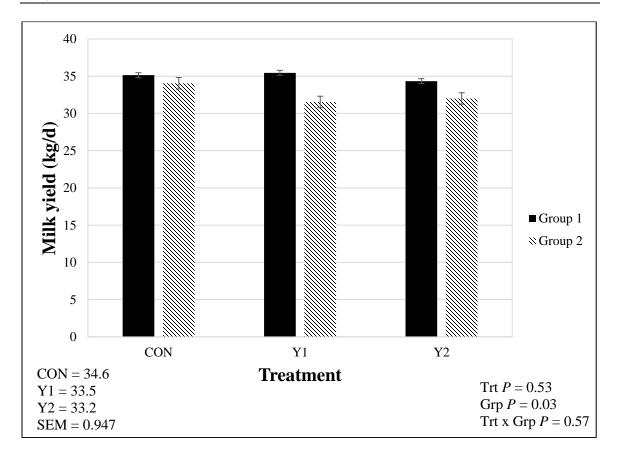
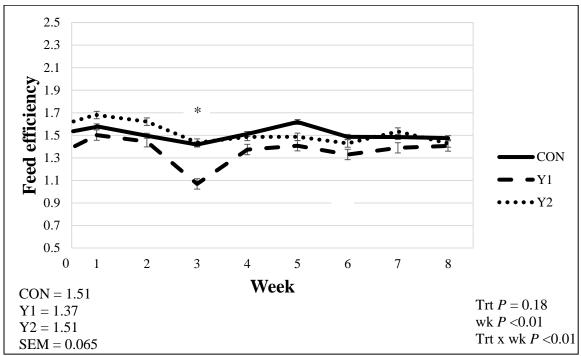


Figure 6. Feed efficiency 1 for cows fed no yeast (**CON**), a concentrated brewer's yeast product (**Y1**), and a concentrated commercial yeast product (**Y2**).



¹Feed efficiency = ECM/DMI.

^{*}*P* < 0.05

Figure 7. Serum glucose concentrations for group 1 and group 2 cows fed no yeast (**CON**), a concentrated brewer's yeast product (**Y1**), and a concentrated commercial yeast product (**Y2**).

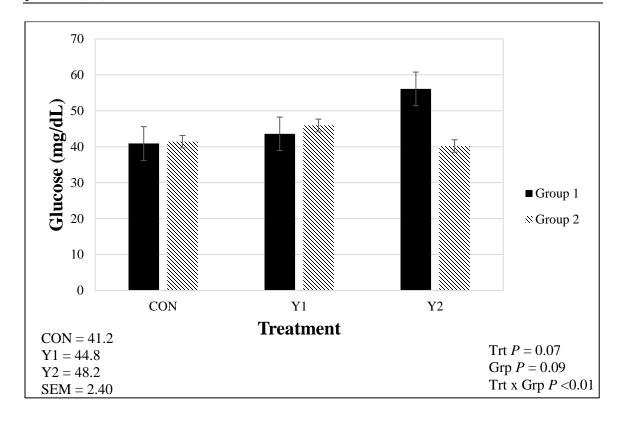


Table 1. Ingredient composition for the CON, Y1, and Y2 treatment diets fed to lactating dairy cows.

		Treatment ¹	
Ingredient ² , % DM	CON	Y1	Y2
Corn silage	27.5	27.5	27.5
Alfalfa haylage	19.2	19.2	19.2
Whole cottonseed	5.7	5.7	5.7
Corn, ground	17.9	17.9	17.9
Soybean meal	3.8	3.8	3.8
Soybean hulls, ground	6.78	6.78	6.78
DDGS	5.2	4.6	4.6
Y1 treatment mix	0.0	0.06	0.0
Y2 treatment mix	0.0	0.0	0.06
Energy Booster 100 ³	0.50	0.50	0.50
Salt	0.60	0.60	0.60
Calcium carbonate	1.00	1.00	1.00
Sodium bicarbonate	0.40	0.40	0.40
Magnesium oxide	0.16	0.16	0.16
Urea, 281%	0.40	0.40	0.40
JPW Dairy TM premix ⁴	0.16	0.16	0.16
JPW Dairy Vitamin premix ⁵	0.16	0.16	0.16
Soybean oil ⁶	0.20	0.20	0.20

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product (**Y2**).

²Formulated using AMTS. v 4.1.4.0.

³Contained: 2.5% C12:0 Myristic, 28.0% C16:0 Palmitic, 45.0% C18:0 Stearic, 8.3% C18:1 Oleic, 1.5% C18:2 Linoleic, and 0.1% C18:3 Linolenic (Milk Specialties Global, Eden Prairie, MN).

⁴Contained: 11.7 % Ca (DM basis), 1.96 % S, 10,527 mg/kg Fe, 63,158 mg/kg Zn, 12,632 mg/kg Cu, 63,158 mg/kg Mn, 325 mg/kg Se, 632 mg/kg Co, and 1,053 mg/kg I (JPW Nutrition, Sioux Falls, SD).

⁵Contained: 25.8 % Ca (DM basis) 1,545 IU/kg Vitamin A, 387 IU/kg Vitamin D, and 4,826 IU/kg Vitamin E (JPW Nutrition, Sioux Falls, SD).

⁶Added for dust control.

Table 2. Formulated¹ nutrient composition of the basal total mixed² ration.

	a mumem con	iposition of the basar total mixed fation.
Item ³ , % DM	Basal TMR	
DM, %	50.3	
Ash	7.46	
OM	92.5	
CP	17.1	
NDF	30.2	
ADF	20.2	
Ether extract	4.70	
Lignin	3.61	
Hemicellulose	10.0	
Cellulose	16.6	
NFC^4	41.2	
Starch	26.2	
Ca	0.93	
P	0.36	
Mg	0.35	
K	1.38	
S	0.21	
Na	0.36	
Cl	0.62	
Mn, mg/kg	111	
Zn, mg/kg	114	
Cu, mg/kg	25.8	
Fe, mg/kg	181	
Mo, mg/kg	-	
DCAD, mEq/100g	20.0	

¹Formulated using AMTS. v 4.1.4.0. ²14.2 g/hd/d of treatment was added to make Y1 and Y2 diets. ³% DM, unless otherwise indicated. ⁴NFC = 100 - (% NDF + % CP + % EE + % Ash) (NRC, 2001).

Table 3. Analyzed nutrient composition of major ingredients used in the CON, Y1, and Y2 treatment diets.

		-	rages ¹		Concentrates ²					
Item ³ , % DM	CS	SE^4	AH	SE^4	WCS	SE^4	GM	SE^4	DDGS	SE^4
DM, %	30.3	0.234	42.7	0.620	91.2	0.032	86.2	0.355	87.0	0.395
Ash	4.55	0.020	10.0	0.300	4.45	0.026	8.88	0.015	4.90	0.130
OM	95.5	0.020	90.0	0.300	95.6	0.026	91.1	0.015	95.1	0.130
CP	8.61	0.095	20.8	0.375	20.9	0.012	17.7	0.026	30.0	0.147
NDF	40.7	0.121	42.5	0.665	51.7	0.254	21.1	0.043	33.5	0.248
ADF	25.0	0.035	32.5	0.135	39.8	0.141	11.5	0.046	12.0	0.214
Ether extract	2.87	0.029	2.95	0.200	19.3	0.087	4.35	0.017	8.34	0.115
Lignin	2.82	0.118	8.69	0.010	11.4	0.121	0.95	0.038	2.23	0.085
Hemicellulose	15.7	0.087	10.0	0.800	11.9	0.110	9.62	0.003	21.5	0.090
Cellulose	21.6	0.075	29.9	0.120	38.6	0.130	10.3	0.032	10.8	0.222
NFC ⁵	43.4	0.180	23.4	0.790	3.69	0.325	48.0	0.065	23.2	0.132
Starch	33.3	0.979	0.24	0.055	0.44	0.101	39.3	0.098	4.90	0.179
Ca	0.37	0.009	1.64	0.030	0.18	0.0	1.71	0.009	0.07	0.0
P	0.27	0.0	0.33	0.0	0.79	0.017	0.35	0.0	1.01	0.006
Mg	0.24	0.006	0.47	0.010	0.39	0.003	0.45	0.009	0.36	0.004
K	1.19	0.003	3.13	0.185	1.34	0.015	0.99	0.012	1.28	0.018
S	0.11	0.003	0.35	0.010	0.25	0.006	0.22	0.0	0.64	0.007
Na	0.01	0.0	0.12	0.005	0.01	0.0	1.22	0.0	0.22	0.002
Cl	0.32	0.003	0.80	0.035	0.08	0.003	1.18	0.017	0.20	0.0
Mn, mg/kg	38.5	0.289	56.3	2.25	18.5	0.289	261	2.31	19.3	0.31
Zn, mg/kg	16.5	8.37	30.5	1.50	42.5	1.44	367	0.866	67.5	2.50
Cu, mg/kg	11.5	0.289	16.5	0.50	5.50	0.289	63.5	2.02	1.67	0.401
Fe, mg/kg	170	6.35	166	16.5	55.0	0.577	288	1.44	90.9	1.60
Mo, mg/kg	0.56	0.072	3.20	0.18	0.35	0.113	0.82	0.029	1.04	0.112
DCAD, mEq/100g	15.4	0.170	40.8	3.02	16.7	0.938	31.2	0.318	-3.59	0.182

¹CS = corn silage, AH = alfalfa haylage. ²WCS = whole cottonseed, GM = grain mix, DDGS = Dried distillers grains with solubles carrier of CON, Y1, and Y2 diets.

³% DM, unless otherwise indicated.

⁴Standard error.

 $^{{}^{5}}NFC = 100 - (\% NDF + \% CP + \% EE + \% Ash) (NRC, 2001).$

Table 4. Analyzed nutrient composition of the basal total mixed ration¹ fed.

Item ² , % DM	Basal TMR	SE ³
DM, %	45.3	0.482
Ash	7.05	0.006
OM	93.0	0.006
CP	15.7	0.075
NDF	35.8	0.176
ADF	24.5	0.268
EE	4.13	0.075
Lignin	4.48	0.055
Hemicellulose	11.4	0.095
Cellulose	22.2	0.260
NFC^4	37.3	0.173
Starch	22.6	0.222
Ca	0.92	0.035
P	0.35	0.0
Mg	0.37	0.003
K	1.64	0.064
S	0.23	0.012
Na	0.41	0.006
Cl	0.68	0.012
Mn, mg/kg	123.5	1.16
Zn, mg/kg	136	2.89
Cu, mg/kg	36.8	0.433
Fe, mg/kg	375	8.08
Mo, mg/kg	1.34	0.098
DCAD, mEq/100g	26.0	0.361

^{114.2} g/hd/d of treatment was added to make Y1 and Y2 diets.
2% DM, unless otherwise indicated.

³Standard error of analyzed basal TMR composites.

⁴NFC = 100 - (% NDF + % CP + % EE + % Ash) (NRC, 2001).

Table 5. Particle distribution and physically effective fiber using the Penn State Particle Separator of the basal total mixed ration.

Item	Basal TMR ¹	
Screen ²	Percentage retained on each sieve	SE^3
Upper (19 mm)	26.8	1.47
Middle (8 mm)	33.5	1.14
Lower (1.18 mm)	8.94	0.18
Bottom Pan	30.7	0.44

¹CON TMR = CON treatment total mixed ration; 45.4% DM, 7.05% Ash, 15.7% CP, 35.8 % NDF, 24.5 % ADF, 37.3 % NFC, 4.13 % EE, and 22.6% Starch.

²Particle size distribution of diets was measured using the Penn State Particle Separator (Kononoff and Heinrichs, 2003).

³Standard error.

Table 6. Dry matter intake, milk yield and composition, efficiency calculations, and body characteristics for cows fed CON, Y1, and Y2 diets.

	Treatment ¹					<i>P</i> -value ²			
Item	CON	Y1	Y2	SEM	Trt	Wk	Trt x Wk	Grp	Trt x Grp
DMI, kg/d	24.2	24.6	24.1	0.800	0.85	< 0.01	0.01	0.38	0.15
Milk, kg/d	34.6	33.5	33.2	0.947	0.53	< 0.01	0.04	0.03	0.57
Fat, %	4.08	3.92	3.77	0.152	0.32	< 0.01	0.73	0.09	0.63
Fat, kg/d	1.33	1.29	1.30	0.068	0.91	< 0.01	0.78	0.16	0.62
Protein, %	2.86	2.89	2.86	0.033	0.88	< 0.01	0.71	0.53	0.84
Protein, kg/d	0.98	0.96	0.94	0.032	0.69	< 0.01	0.04	0.10	0.87
Lactose, %	4.90	4.93	4.94	0.021	0.42	0.03	1.00	0.01	0.50
Lactose, kg/d	0.98	0.96	0.94	0.032	0.69	< 0.01	0.04	0.10	0.87
SNF, %	9.02	9.09	9.04	0.045	0.46	< 0.01	0.98	< 0.01	0.96
SNF, kg/d	3.08	3.00	2.97	0.091	0.66	< 0.01	0.04	< 0.01	0.69
MUN, mg/dL	14.2	13.7	13.6	0.402	0.51	< 0.01	0.38	0.65	0.56
SCC (1000/mL)	114.9	218.8	249.2	123.6	0.69	0.14	0.38	0.15	0.70
ECM,3 kg/d	35.5	34.5	34.3	1.41	0.79	< 0.01	0.45	0.27	0.91
Feed efficiency ⁴	1.51	1.37	1.51	0.065	0.18	< 0.01	< 0.01	0.97	0.64
Body weight, kg	671.7	673.7	667.1	6.37	0.72	< 0.01	0.97	0.03	0.72
Initial, kg	641.9	658.0	678.3	24.58	< 0.01	-	-	-	-
Final, kg	656.8	673.9	689.6	19.68	0.44	-	-	-	-
BCS ⁵	2.43	2.49	2.44	0.030	0.34	0.03	0.71	0.45	0.16
Initial	2.44	2.52	2.48	0.016	< 0.01	-	-	-	-
Final	2.46	2.56	2.50	0.150	0.24	-	-	-	-

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product (**Y2**).

²Trt = Treatment; Wk = Week; Grp = Group.

 $^{^{3}}ECM = [(0.327 \text{ x kg milk}) + (12.95 \text{ x kg fat}) + (7.2 \text{ x kg protein})] \text{ (Orth, 1992)}.$

⁴Feed efficiency = ECM/DMI.

 $^{^{5}}$ Body condition score with 1 = emaciated and 5 = obese (Wildman et al., 1982).

Table 7. Feed fatty acid (**FA**) concentration of the basal total mixed ration¹.

	Basal TMR ¹							
Item ²	g/kg of DM of TMR	SE ³	mg/100 mg FA	SE ³				
C4:0	0.09	0.015	0.37	0.066				
C6:0	0.01	0.0	0.05	0.002				
C8:0	0.04	0.004	0.17	0.021				
C10:0	0.36	0.040	1.50	0.135				
C12:0	0.25	0.108	1.02	0.426				
C13:0	0.19	0.083	0.77	0.334				
C14:0	1.00	0.073	4.25	0.413				
C15:1	0.01	0.005	0.06	0.022				
C16:0	3.80	0.123	16.1	0.120				
C16:1 <i>cis-</i> 9	0.07	0.001	0.29	0.003				
C16:1 trans-9	0.03	0.006	0.13	0.022				
C18:0	1.29	0.022	5.45	0.142				
C18:1 trans-6	0.0	0.004	0.02	0.018				
C18:1 trans-10	0.02	0.006	0.07	0.026				
C18:1 <i>cis-</i> 9	3.09	0.088	13.1	0.174				
C18:1 <i>cis</i> -11	0.32	0.023	1.34	0.063				
C18:2 cis-9,12	7.82	0.244	33.1	0.242				
C18:3 n-3	0.49	0.013	2.08	0.030				
C18:3 n-6	0.37	0.072	1.56	0.270				
C20:0	1.32	0.015	5.59	0.197				
Total ⁴	23.6	0.614	100	0.0				
Others ⁵	3.95	0.048	16.7	0.631				
Short ⁶	0.30	0.017	1.28	0.074				
Medium ⁷	7.94	0.159	33.6	0.374				
Long ⁸	16.3	0.500	68.8	0.435				
Saturated	9.34	0.306	36.5	0.350				
Monounsaturated	5.69	0.080	24.1	0.816				
Polyunsaturated	9.31	0.310	39.4	0.365				
Total Unsaturated ⁹	15.0	0.310	63.5	0.634				

¹CON TMR = CON treatment total mixed ration; 45.4% DM, 7.05% Ash, 15.7% CP, 35.8 % NDF, 24.5 % ADF, 37.3 % NFC, 4.13 % EE, and 22.6% Starch.³

²Represented as number of carbons:number of double bonds.

³Standard error.

⁴Total fatty acids.

⁵Others = C5:0, C9:0, C11, C11:1, C12:1, C14:1, C15:0, C20:1,5, C20:1 *cis*, C20:2-11,14, C20:3 Homo Gamma, C20:4, C22:0, C22:2, C22:3, C22:4, C22:5 n-6, C22:6, C23:0, and C24:0.

 $^{^{6}}$ Short = C4:0 to C9:0.

 $^{^{7}}$ Medium = C10:0 to C16:1.

 $^{^{8}}$ Long = C17:0 to C22:6.

⁹Total unsaturated fatty acids = Monounsaturated + Polyunsaturated.

Table 8. Milk fatty acid (**FA**) concentrations from cows fed CON, Y1, and Y2 diets on weeks 2, 4, 6, and 8.

	Treatment ¹			, , ,	P-value ²				
Item ³ , mg/100 mg FA	CON	Y1	Y2	SEM	Trt	Wk	Trt x Wk	Grp	Trt x Grp
C4:0	2.68	2.56	2.65	0.053	0.22	0.22	0.92	0.56	0.69
C6:0	1.62	1.58	1.61	0.034	0.65	< 0.01	0.58	0.45	0.76
C8:0	1.22	1.21	1.19	0.023	0.66	< 0.01	0.31	0.01	0.66
C10:0	2.67	2.78	2.67	0.077	0.48	< 0.01	0.36	0.01	0.47
C12:0	2.95	2.98	2.89	0.120	0.84	0.13	0.99	0.09	0.70
C14:0	10.5	10.6	10.3	0.213	0.59	< 0.01	0.62	0.22	0.61
C15:1	0.65	0.67	0.64	0.034	0.75	< 0.01	0.53	0.36	0.81
C16:0	30.4	30.0	29.9	0.648	0.81	< 0.01	0.53	0.05	0.32
C16:1 <i>cis</i> -9	1.15	1.05	1.05	0.052	0.22	0.02	0.67	0.57	0.90
C16:1 trans-9	0.29	0.28	0.31	0.009	0.11	< 0.01	0.45	< 0.01	0.88
C18:0	14.1	14.4	14.2	0.717	0.93	< 0.01	0.17	0.18	0.44
C18:1 trans-6	0.19	0.19	0.16	0.022	0.42	0.02	0.23	0.47	0.67
C18:1 trans-9	0.06	0.05	0.05	0.015	0.90	< 0.01	0.81	0.54	0.96
C18:1 trans-10	0.71	0.67	0.64	0.040	0.41	< 0.01	0.98	0.63	0.53
C18:1 trans-11	0.30	0.31	0.31	0.020	0.82	< 0.01	0.97	0.64	0.40
C18:1 <i>cis</i> -6	0.37	0.35	0.37	0.035	0.91	0.01	0.93	0.85	0.78
C18:1 <i>cis</i> -9	18.4	18.9	18.4	0.675	0.82	< 0.01	0.82	0.41	0.60
C18:1 <i>cis</i> -11	0.65	0.81	1.26	0.294	0.27	< 0.01	0.77	0.65	0.48
C18:2 trans-9,12	0.14	0.13	0.14	0.012	0.67	< 0.01	0.89	0.40	0.35
C18:2 <i>cis</i> -9,12	6.54	6.69	7.16	0.262	0.12	< 0.01	0.72	0.19	0.39
C18:3 n-3	0.35	0.36	0.37	0.024	0.87	0.07	0.67	0.14	0.75
C18:3 n-6	0.20	0.20	0.18	0.013	0.49	< 0.01	0.30	0.05	0.47
C20:0	0.45	0.48	0.52	0.035	0.29	< 0.01	0.51	0.80	0.24
CLA ⁴ (trans-10, cis-12)	0.06	0.05	0.05	0.007	0.94	0.02	0.30	0.02	0.28
CLA ⁴ (<i>cis-9</i> , <i>trans-11</i>)	0.01	0.04	0.01	0.011	0.05	0.01	0.23	0.60	0.65
Others ⁵	3.21	3.49	3.20	0.326	0.75	0.22	0.27	0.33	0.26
Short ⁶	5.64	5.50	5.64	0.097	0.51	< 0.01	0.71	0.79	0.74

Medium ⁷	50.4	49.9	49.3	0.948	0.65	< 0.01	0.42	0.76	0.28
Long ⁸	44.0	45.2	45.1	1.07	0.67	< 0.01	0.43	0.65	0.20
Saturated	68.9	68.8	68.0	0.814	0.58	< 0.01	0.60	0.16	0.87
Monounsaturated	23.8	24.2	24.2	0.603	0.78	< 0.01	0.75	0.26	0.49
Polyunsaturated	7.27	7.55	7.93	0.296	0.18	< 0.01	0.78	0.03	0.28
Total Unsaturated ⁹	31.1	31.7	32.1	0.679	0.53	< 0.01	0.79	0.16	0.88

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product (**Y2**).

²Trt = Treatment; Wk = Week; Grp = Group.

³Represented as number of carbons:number of double bonds.

⁴CLA = Conjugated linoleic acid.

⁵Others = C5:0, C7:0, C9:0, C11, C11:1, C12:1, C14:1, C15:0, C17:0, C17:1, C19:0, C20:1,5, C20:1,8, C20:1 *cis*, C20:2 *cis*-11,14, C20:3 Homo Gamma, C20:3 *cis*-11,14, C20:4, C20:5, C22:0, C22:1, C22:2, C22:3, C22:4, C22:5 n-3, C22:5 n-6, C22:6, C23:0, and C24:0.

 $^{^{6}}$ Short = C4:0 to C9:0.

 $^{^{7}}$ Medium = C10:0 to C16:1.

 $^{^{8}}$ Long = C17:0 to C22:6.

⁹Total unsaturated fatty acids = Monounsaturated + Polyunsaturated.

Table 9. Plasma metabolite concentrations during weeks 7 and 8 of cows fed CON, Y1, and Y2 diets.

		Γreatmen	t^1	_		ıe ²	
Plasma metabolite	CON	Y1	Y2	SEM	Trt	Grp	Trt x Grp
Glucose, mg/dL	41.2	44.8	48.2	2.40	0.07	0.09	< 0.01
PUN ³ , mg/dL	17.2	14.7	16.0	0.484	< 0.01	0.42	0.10
Cholesterol, mg/dL	122.3	128.7	126.9	5.22	0.63	0.61	0.63

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product (**Y2**).

²Trt = Treatment; Grp = Group.

³Plasma urea nitrogen.

Table 10. Ruminal pH, NH₃, and VFA concentrations of cows fed CON, Y1, and Y2 diets.

dicts.							
		reatmen	t^{l}	<i>P</i> -value ²			
Rumen measure	CON	Y1	Y2	SEM	Trt	Grp	Trt x Grp
pН	6.81	6.85	6.80	0.046	0.74	0.53	0.87
NH ₃ -N, mg/dL	18.5	18.8	17.2	0.673	0.18	0.02	0.03
Acetate, mM	49.5	51.6	52.1	1.60	0.45	< 0.01	0.44
Propionate, mM	16.6	17.8	17.8	0.675	0.32	< 0.01	0.42
Isobutyrate, mM	1.51	1.39	1.45	0.082	0.61	0.51	0.30
Butyrate, mM	8.75	8.43	8.93	0.391	0.65	0.16	0.71
Isovalerate, mM	1.95	2.00	1.92	0.127	0.89	0.40	0.22
Valerate, mM	1.71	1.58	1.60	0.083	0.48	0.89	0.04
Total VFA, mM	80.0	82.8	83.8	2.58	0.53	< 0.01	0.40
Acetate ³	61.7	62.3	62.2	0.610	0.76	0.17	0.52
Propionate ³	20.8	21.4	21.2	0.435	0.64	0.85	0.94
Isobutyrate ³	1.87	1.68	1.73	0.071	0.15	0.12	0.30
Butyrate ³	11.0	10.3	10.7	0.268	0.14	0.07	0.48
Isovalerate ³	2.47	2.44	2.30	0.126	0.57	0.28	0.39
Valerate ³	2.16	1.94	1.93	0.088	0.12	0.01	0.03
Acetate:Propionate	3.01	2.94	2.95	0.086	0.81	0.76	0.84
1	11						

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product (**Y2**).

²Trt = Treatment; Grp = Group.

 $^{^{3}}$ mM/100 mM.

Table 11. Total tract digestibility of nutrients by cows fed a CON, Y1, and Y2 diets.

		Treatmen	t^1		<i>P</i> -value ²			
Item, % digested	CON	Y1	Y2	SEM	Trt	Grp	Trt x Grp	
DM	89.7	89.3	88.5	0.59	0.22	< 0.01	0.29	
OM	64.1	61.7	61.2	1.86	0.42	< 0.01	0.45	
CP	65.6	65.0	63.3	2.06	0.62	< 0.01	0.44	
NDF	75.3	72.9	72.9	1.39	0.30	< 0.01	0.29	
ADF	74.8	72.5	72.3	1.44	0.31	< 0.01	0.25	
Lignin	83.3	82.0	81.2	1.30	0.42	0.05	0.45	

¹No yeast (**CON**); concentrated brewer's yeast product (**Y1**); concentrated commercial yeast product ($\mathbf{Y2}$). ²Trt = Treatment; Grp = Group.

OVERALL CONCLUSION

Research findings presented helped to further our understanding of the main objective of evaluating supplementation of a concentrated brewer's yeast (Saccharomyces cerevisiae) in the ration of lactating dairy cows. Similar to published literature and meta-analyses on yeast fed to cattle, our results demonstrated inconsistent results (Lescoat et al., 2000; Sauvant et al., 2004; Desnoyers et al., 2009). Overall milk production and milk fat yields did not demonstrate significant results in our study, as previously demonstrated in numerous studies (Arambel and Kent, 1990; Piva et al., 1993; Wang et al., 2001). However, we speculate that heat stress and temperature variability contributed to our findings as shown by Schingoethe et al. (2004), Shwartz et al. (2009), and Salvati et al. (2015). We did see an increase in dry matter intake (**DMI**) for cows fed the Y1 treatment, indicating that yeast products are a viable option for cows prone to decreased intake, such as during the transition period. In addition, a treatment by week effect for feed efficiency (P < 0.05) was detected for CON and Y2 diets, but not Y1. Similar to findings by Longuski et al. (2009) and Hristov (2010), milk fatty acid composition was not largely altered by yeast supplementation. Rumen microbial fermentation was altered by diets fed yeast, but not enough to statistically increase milk production by treatments. Therefore, the results from this research demonstrate that yeast products can maintain performance without causing detriment to the cow or its production status.

ACKNOWLEDGEMENTS

Funding for this research was provided by The F. L. Emmert Company (Cincinnati, OH) with support provided from the South Dakota State University Agricultural Experiment Station (Brookings, SD). The authors also thank fellow graduate students and farm personnel in the Food and Dairy Science Department at South Dakota State University (Brookings, SD) at the time this study was conducted for their help with sampling and animal care.

LITERATURE CITED

- Abdelqader, M. M., A. R. Hippen, D. J. Schingoethe, K. F. Kalscheur, K. Karges, and M. L. Gibson. 2009. Evaluation of corn germ from ethanol production as an alternative fat source in dairy cow diets. J. Dairy Sci. 92:1023-1037.
- Alugongo, G. M., J. X. Xiao, Y. H. Chung, S. Z. Dong, S. L. Li, I. Yoon, Z. H. Wu, and Z. J. Cao. 2016. Effects of Saccharomyces cerevisiae fermentation products on dairy calves: performance and health. J. Dairy Sci. 100:1189–1199.
- AlZahal, O., E. Kebreab, J. France, and B. W. McBride. 2007. A mathematical approach to predicting biological values from ruminal pH measurements. J. Dairy Sci. 90:3777-3785.
- AlZahal, O., L. Dionissopoulos, A. H. Laarman, N. Walker, and B. W. McBride. 2014.

 Active dry Saccharomyces cerevisiae can alleviate the effect of subacute ruminal acidosis in lactating dairy cows. J. Dairy Sci. 97:7751-7763.
- AlZahal, O., M. M. Or-Rashid, S. L. Greenwood, M. S. Douglas, and B. W. McBride.

 2009. The effect of dietary fiber level on milk fat concentration and fatty acid
 profile of cows fed diets containing low levels of polyunsaturated fatty acids. J.

 Dairy Sci. 92:1108-1116.
- AlZahal, O., M. M. Or-Rashid, S. L. Greenwood, and B. W. McBride. 2010. Effect of subacute ruminal acidosis on milk fat concentration, yield and fatty acid profile of dairy cows receiving soybean oil. J. Dairy Res. 77:376-384.
- AOAC International. 1995. Official Methods of Analysis. 16th ed. AOAC International, Arlington, VA.

- AOAC International. 2002. Official Methods of Analysis. 17th ed. Association of Official Analytical Chemists. Gaithersburg, MD.
- Arambel, M. J. and B. A. Kent. 1990. Effect of yeast culture on nutrient digestibility and milk yield response in early to midlactation dairy cows. J. Dairy Sci. 73:1560-1563.
- Bach, A., C. Iglesias, and M. Devant. 2007. Daily rumen pH pattern of loose-housed dairy cattle as affected by feeding pattern and live yeast supplementation. Anim. Feed Sci. Technol. 136:146–153.
- Bach Knudsen, K. E. 1997. Carbohydrate and lignin contents of plant materials used in animal feeding. Anim. Feed Sci. Technol. 67:319-338.
- Bauman, D. E., and J. L. Capper. 2011. Sustainability and dairy production: challenges and opportunities. Proc. Cornell Nutr. Conf. Feed Manuf.: 136-153.
- Bauman, D. E., and J. M. Griinari. 2003. Nutritional regulation of milk fat synthesis.

 Annu. Rev. Nutr. 23:203-227.
- Bauman, D. E., J. W. Perfield II, K. J. Harvatine, and L. H. Baumgard. 2008. Regulation of fat synthesis by conjugated linoleic acid: Lactation and the ruminant model. J. Nutr. 138:403-409.
- Baumgard, L. H., B. A. Corl, D. A. Dwyer, A. Saebo, and D. E. Bauman. 2000.
 Identification of the conjugated linoleic acid isomer that inhibits milk fat
 synthesis. Am. J. Physiol. Regul. Integr. Comp. Physiol. 278:R179–R184.
- Bayat, A. R., P. Kairenius, T. Stefanski, H. Leskinen, S. Comtet-Marre, E. Forano, F. Chaucheyras-Durand, and K. J. Shingfield. 2015. Effect of camelina oil or live yeasts (Saccharomyces cerevisiae) on ruminal methane production, rumen

- fermentation, and milk fatty acid composition in lactating cows fed grass silage diets. J. Dairy Sci. 98:3166-3181.
- Beauchemin, K. A., M. Kreuzer, F. O. O'Mara, and T. A. McAllister. 2008. Nutritional management for enteric methane abatement: A review. Aust. J. Exp. Agric. 48:21-27.
- Berman, A., Y. Folman, M. Kaim, M. Mamen, Z. Herz, D. Wolfenson, A. Arieli, and Y. Graber. 1985. Upper critical temperatures forced ventilation effects for high yielding dairy cows in a subtropical climate. J. Dairy Sci. 68:1488–1495.
- Bitencourt, L. L., J. R. M. Silva, B. M. L. Oliveira, G. S. Dias Júnior, F. Lopes, S. Siécola Júnior, O. F. Zacaroni, and M. N. Pereira. 2011. Diet digestibility and performance of dairy cows supplemented with live yeast. Sci. Agric. 68:301-307.
- Block, E. 1984. Manipulating dietary anions and cations for prepartum dairy cows to reduce incidence of milk fever. J. Dairy Sci. 67:2939–2948.
- Bohmanova, J., I. Misztal, and J. B. Cole. 2007. Temperature-humidity indices as indicators of milk production losses due to heat stress. J. Dairy Sci. 90:1947-1956.
- Bonhomme, A. 1990. Rumen ciliates: their metabolism and relationships with bacteria and their hosts. Anim. Feed Sci. Technol. 30:203–266.
- Brewer, M. T., K. L. Anderson, I. Yoon, M. F. Scott, and S. A. Carlson. 2014.

 Amelioration of salmonellosis in pre-weaned dairy calves fed Saccharomyces cerevisiae fermentation products in feed and milk replacer. Vet. Microbiol. 172:248-255.

- Brossard, L., F. Chaucheyras-Durand, B. Michalet-Doreau, and C. Martin. 2006. Dose effect of live yeasts on rumen microbial communities and fermentations during butyric latent acidosis in sheep: new type of interaction. Anim. Sci. 82:829–836.
- Cabello, F. C. 2006. Heavy use of prophylactic antibiotics in aquaculture: a growing problem for human and animal health and for the environment. Environ.

 Microbiol. 8:1137-1144.
- Cakiroglu, D., Y. Meral, D. Pekmezci, and F. Akdag. 2010. Effects of live yeast culture (Saccharomyces cerevisiae) on milk production and blood lipid levels of Jersey cows in early lactation. J. Anim. Vet. Adv. 9:1370-1374.
- Callaway, T. R., T. S. Edrington, J. L. Rychlik, K. J. Genovese, T. L. Poole, Y. S. Jung,
 K. M. Bischoff, R. C. Anderson, and D. J. Nisbet. 2003. Ionophores: Their use as ruminant growth promotants and impact on food safety. Curr. Issues Intest.
 Microbiol. 4:43–51.
- Callaway, E. S., and S. A. Martin. 1997. Effects of a *Saccharomyces cerevisiae* culture on ruminal bacteria that utilize lactate and digest cellulose. J. Dairy Sci. 80:2035-2044.
- Capper, J. L., R. A. Cady, and D. E. Bauman. 2009. The environmental impact of dairy production: 1944 compared with 2007. J. Anim. Sci. 87:2160-2167.
- Chaney, A. L., and E. P. Marbach. 1962. Modified reagents for determination of urea and ammonia. Clin. Chem. 8:130-132.
- Chaucheyras-Durand F., E. Chevaux, C. Martin, and E. Forano. 2012. Use of yeast probiotics in ruminants: effects and mechanisms of action on rumen pH, fiber

- degradation, and microbiota according to the diet in: probiotic in animals. Chapter 7.
- Chaucheyras-Durand, F., and H. Durand. 2010. Probiotics in animal nutrition and health.

 Benef. Microb. 1:3–9.
- Chaucheyras-Durand, F., and G. Fonty. 2002. Influence of a probiotic yeast

 (Saccharomyces cerevisiae CNC I-1077) on microbial colonization and
 fermentation in the rumen of newborn lambs. Microb. Ecol. Health Dis. 14:30–36.
- Chaucheyras F., G. Fonty, G. Bertin, J. M. Salmon, and P. Gouet. 1995. Effects of a strain of Saccharomyces cerevisiae (Levucell SC), a microbial additive for ruminants, on lactate metabolism in vitro. Can. J. Microbio. 42:927-933.
- Chaucheyras-Durand, F., N. D. Walker, and A. Bach. 2008. Effects of active dry yeasts on the rumen microbial ecosystem: Past, present and future. Anim. Feed Sci. Technol. 145:5–26.
- Chauvel, J., and R. Granier. 1988. Utilisation de la levure de bière liquide pour le porc charcutier. Journées Rech. Porc. France, 20:369-374.
- Crawshaw, R., 2004. Co-product feeds: animal feeds from the food and drinks industries.

 Nothingham University Press.
- Cromwell, G. L. 2002. Why and how antibiotics are used in swine production. Anim. Biotech. 13:7-27.
- Dann, H. M., J. K. Drackley, G. C. McCoy, M. F. Hutjens, and J. E. Garrett. 2000.

 Effects of yeast culture (Saccharomyces cerevisiae) on prepartum and postpartum intake and milk production of Jersey cows. J. Dairy Sci. 83:123-127.

- Dehghan-Banadaky, M., M. Ebrahimi, R. Motameny, and S. R. Heidaria. 2013. Effects of live yeast supplementation on mid-lactation dairy cows performances, milk composition, rumen digestion and plasma metabolites during hot season. J. Appl. Anim. Res. 41:137–142.
- Desnoyers, M., S. Giger-Reverdin, G. Bertin, C. Duvaux-Ponter, and D. Sauvant. 2009.

 Meta-analysis of the influence of Saccharomyces cerevisiae supplementation on ruminal parameters and milk production of ruminants. J. Dairy Sci. 92:1620-1632.
- Doreau, M., and J. P. Jouany. 1998. Effect of Saccharomyces cerevisiae culture on nutrient digestion in lactating dairy cows. J. Dairy Sci. 81:3214-3221.
- Dunne, C., L. Murphy, S. Flynn, L. O'Mahony, S. O'Halloran, M. Feeney, D. Morrissey,
 G. Thornton, G. Fitzgerald, F. Shanahan, and J. K. Collins. 1999. Probiotics: from myth to reality: demonstration of functionality in animal models of disease and in human clinical trials. Antonie Van Leeuwenhoek. 76:279-292.
- Ender, F., I. W. Dishington, and A. Helgebostad. 1962. Parturient paresis and related forms of hypocalcaemic disorders induced experimentally in dairy cows. Acta. Vet. Scand. 3(Suppl 1):5–52.
- Erasmus, L. J., P. M. Botha, and A. Kistner. 1992. Effect of yeast culture supplement on production, rumen fermentation, and duodenal nitrogen flow in dairy cows. J. Dairy Sci. 75:3056-3065.
- Erasmus, L. J., P. H. Robinson, A. Ahmadi, R. Hinders, and J. E. Garrett. 2005. Influence of prepartum and postpartum supplementation of a yeast culture and monensin, or

- both, on ruminal fermentation and performance of multiparous dairy cows. Anim. Feed Sci. Tech. 122:219-239.
- Ferraretto, L. F., R. D. Shaver, and S. J. Bertics. 2012. Effect of dietary supplementation with live-cell yeast at two dosages on lactation performance, ruminal fermentation, and total tract nutrient digestibility in dairy cows. J. Dairy Sci. 95:4017–4028.
- Firkins, J. L. 1997. Effects of feeding nonforage fiber sources on site of fiber digestion. J. Dairy Sci. 80:1426-1437.
- Food and Agriculture Organization of the United Nations and World Health Organization (FAO). 2005. Responsible use of antibiotics in aquaculture. In:Serrano, P.H. (Ed.), FAO Fisheries Technical Paper 469. FAO, Rome, Italy, 98.
- Fuller, R. 1999. Probiotics for farm animals. Probiotics: A Critical Rev. 15:15-22.
- Galip, N. 2006. Effect on supplemental yeast culture and sodium bicarbonate on ruminal fermentation and blood variables in rams. J. Anim. Physiol. Anim. Nutr. 90:446-452.
- Girard, I. D., and K. A. Dawson. 1994. Effects of a yeast culture on the growth characteristics of representative ruminal bacteria. J. Anim. Sci. 77: 300.
- Guedes, C. M., D. Goncalves, M. A. M. Rodrigues, and A. Dias-da-Silva. 2008. Effect of yeast Saccharomyces cerevisiae on ruminal fermentation and fiber degradation of maize silage in cows. Anim. Feed Sci. Technol. 145:27-40.
- Harvatine, K. J., and D. E. Bauman. 2006. SREBP1 and thyroid hormone responsive spot 14 (S14) are involved in the regulation of bovine mammary lipid synthesis during diet-induced milk fat depression and treatment with CLA. J. Nutr. 136:2468-2474.

- Hasunuma, T., Y. Uyeno, K. Akiyama, S. Hashimura, H. Yamamoto, H. Yokokawa, T.
 Yamaguchi, M. Itoh, H. Mizuguchi, S. Sato, M. Hirako, and S. Kushibiki. 2016.
 Consecutive reticular pH monitoring in dairy cows fed diets supplemented with active dry yeast during the transition and mid-lactation periods. Anim. Feed Sci.
 Tech. 221:215-225.
- Helal, F. I. S., and K. A. Abdel-Rahman. 2010. Productive performance of lactating ewes fed diets supplementing with dry yeast and/or bentonite as feed additives. World J. Agric. Sci. 6:489–498.
- Hertrampf, J. W., and F. Piedad-Pascual. 2000. Handbook on ingredients for aquaculture feeds. Kluwer Academic Publishers. pp. 624.
- Hristov, A. N., M. Vander Pol, M. Agle, S. Zaman, C. Schneider, P. Ndegwa, V. K. Vaddella, K. Johnson, K. J. Shingfield, and S. K. R. Karnati. 2009. Effect of lauric acid and coconut oil on ruminal fermentation, digestion, ammonia losses from manure, and milk fatty acid composition in lactating cows. J. Dairy Sci. 92:5561-5582.
- Hristov, A. N., G. Varga, T. Cassidy, M. Long, K. Heyler, S. K. Karnati, B. Corl, C. J. Hovde, and I. Yoon. 2010. Effect of Saccharomyces cerevisiae fermentation product on ruminal fermentation and nutrient utilization in dairy cows. J. Dairy Sci. 93:682-692.
- Hucko, B., V. A. Bampidis, A. Kodes, V. Christodoulou, Z. Mudrik, and V. Poláková Plachy. 2009. Rumen fermentation characteristics in pre-weaning calves receiving yeast culture supplements. Czech J. Anim. Sci. 54:435–442.

- Ipharraguerre, I. R., and J. H. Clark. 2003. Usefulness of ionophores for lactating dairy cows: A review. Anim. Feed Sci. Technol. 106:39–57.
- Jakobsen, M., and J. Narvhus. 1996. Yeasts and their possible beneficial and negative effects on the quality of dairy products. Int. Dairy. J. 6:755-768.
- Jenkins, T. C., R. J. Wallace, P. J. Moate, and E. E. Mosley. 2008. Board-invited review:

 Recent advances in biohydrogenation of unsaturated fatty acids within the rumen microbial ecosystem. J. Anim. Sci. 86:397-412.
- Jensen, R.G. 2002. The composition of bovine milk lipids: January 1995 to December 2000. J. Dairy Sci. 85:295–350.
- Jouany, J. P. 2001. A new look to at yeast culture as probiotics for ruminants. Feed Mix. 9:17–19.
- Jouany, J. P. 2006. Optimizing rumen functions in the close-up transition period and early lactation to drive dry matter intake and energy balance in cows. Anim. Reprod. Sci. 96:250–264.
- Kadzere, C. T., M. R. Murphy, N. Silanikove, and E. Maltz. 2002. Heat stress in lactating dairy cows: A review. Livest. Prod. Sci. 77:59-91.
- Kononoff, P. H., and A. J. Heinrichs. 2003. The effect of corn silage particle size and cottonseed hulls on cows in early lactation. J. Dairy Sci. 2003. 86:2438-2451.
- Kristensen, N. B., V. Duchwaider, and B. A. Røjen. 2014. Effects of dietary yeast supplementation on milk production, feed intake, fecal, and urinary variables in dairy cows under field conditions in Denmark. Knowledge Centre for Agriculture, Cattle, Agro Food Park 15, DK-8200 Aarhus N, Denmark.

- Kung, Jr., L., E. M. Kreck, R. S. Tung, A. Hession, A. C. Sheperd, M. A. Cohen, H. E. Swain, and J. Leedle. 1997. Effects of a live yeast culture and enzymes on in vitro ruminal fermentation and milk production of dairy cows. J. Dairy Sci. 80: 2045-2051.
- Leicester, H. C. v., P. H. Robinson, and L. J. Erasmus. 2016. Effects of two yeast based direct fed microbials on performance of high producing dairy cows. Anim. Feed Sci. Tech. 215:58-72.
- Lescoat, P., D. Ali-Haimoud-Lekhal, and C. Bayourthe. 2000. Effets de Saccharomyces cerevisiae et Aspergillus oryzae sur la digestion et le fonctionnement ruminal: Étude bibliographique. In: Rèmes Rencontres autour des Recherches sur les Ruminants, Paris, France. 7:199.
- Lila, Z. A., N. Mohammed, T. Yasui, Y. Kurokawa, S. Kanda, and H. Itabashi. 2004.

 Effects of a twin strain of Saccharomyces cerevisiae live cells on mixed ruminal microorganism fermentation in vitro. J. Anim. Sci. 82:1847–1854.
- Littell, R. C., W. W. Stroup, G. A. Milliken, R. D. Wolfinger., and O. Schabenberger. 2006. SAS for mixed models. SAS institute.
- Longuski, R. A., Y. Ying, and M. S. Allen. 2009. Yeast culture supplementation prevented milk fat depression by a short-term dietary challenge with fermentable starch. J. Dairy Sci. 92:160-167.
- Lourens, A., and B. C. Viljoen. 2001. Growth and survival of a probiotic yeast in dairy products. Food Res. Int. 34:791-796.

- Lynch, H. A. and S. A. Martin. 2002. Effects of Saccharomyces cerevisiae culture and Saccharomyces cerevisiae live cells on in vitro mixed ruminal microorganism fermentation. J. Dairy Sci. 85:2603-2608.
- Månsson, H. L. 2008. Fatty acids in bovine milk fat. Food and Nutr. Research. 52:1-3.
- Marden, J. P., C. Julien, V. Monteil, E. Auclair, R. Moncoulon, and C. Bayourthe, C. 2008. How does live yeast differ from sodium bicarbonate to stabilize ruminal pH in high yielding dairy cows. J. Dairy Sci. 91:3528-3535.
- Martin, S. A. 1998. Manipulation of ruminal fermentation with organic acids: A review.

 J. Anim. Sci. 76:3123-3132.
- Martin, S. A., S. F. Al-Khaldi, J. D. Evans, and H. M. Sullivan. 1999. Potential for manipulating gastrointestinal microflora: Alternatives to antibiotics and ionophores. Recent Res. Dev. Microbiol. 3:441-454.
- Martin, S. A., B. J. Nisbet, and R. G. Dean. 1989. Influence of a commercial yeast supplement on the in vitro ruminal fermentation. Nutr. Reprod. Int. 40:395-403.
- McGuffey, R. K., L. F. Richardson, and J. I. D. Wilkinson. 2001. Ionophores for dairy cattle: Current status and future outlook. J. Dairy Sci. 84(E. Suppl.):E194-E203.
- Merchen, N.R. 1988. Digestion, Absorption and Excretion in Ruminants. pages 182-189 in The Ruminant Animal: Digestive Physiology and Nutrition. D. C. Church, ed. Prentice Hall Inc., New Jersey.
- Mertens, D. R. 1997. Creating a system for meeting the fiber requirements of dairy cattle.

 J. Dairy Sci. 80:1463–1482.

- Michalet-Doreau, B., and D. Morand. 1996. Effect of yeast culture, Saccharomyces cerevisiae, on ruminal fermentation during adaptation to high-concentrate feeding.

 Ann. Zootech. 45:337.
- Miller-Webster, T. W. H. Hoover, M. Holt, and J. E. Nocek. 2002. Influence of yeast culture on ruminal microbial metabolism in continuous culture. J. Dairy Sci. 85:2009-2014.
- Moallem, U., H. Lehrer, L. Livshitz, M. Zachut, and S. Yakoby. 2009. The effects of live yeast supplementation to dairy cows during the hot season on production, feed efficiency, and digestibility. J. Dairy Sci. 92:343-351.
- Mombelli, B., and M. R. Gismondo. 2000. The use of probiotics in medicinal practice.

 Int. J. Antimicrob. Agents. 16:531-536.
- Moore, B.E., K. E. Newman, P. Spring, F. E. Chandler. 1994. The effect of yeast culture (Yea Sacc 1026) in microbial populations digestion in the cecum and colon of the equine. J. Anim. Sci. 72:1.
- Mosoni, P., F. Chaucheyras-Durand, C. Bera-Maillet, and E. Forano. 2007.

 Quantification by real-time PCR of cellulolytic bacteria in the rumen of sheep after supplementation of a forage diet with readily fermentable carbohydrates: effect of a yeast additive. J. Appl. Microbiol. 103:2676–2685.
- Moya, D., S. Calsamiglia, A. Ferret, M. Blanch, J. I. Fandino, L. Castillejos, and I. Yoon. 2009. Effects of dietary changes and yeast culture (Saccharomyces cerevisiae) on rumen microbial fermentation of Holstein heifers. J Anim Sci 87:2874-2881.
- Mullins, C. R., L. K. Mamedova, A. J. Carpenter, Y. Ying, M. S. Allen, I. Yoon, and B.J. Bradford. 2013. Analysis of rumen microbial populations in lactating dairy

- cattle fed diets varying in carbohydrate profiles and Saccharomyces cerevisiae fermentation product. J. Dairy Sci. 96:5872-5881.
- Murphy, S. P., and L. H. Allen. 2003. Nutritional importance of animal source foods. J. Nutr. 133:3932S-3935S.
- Nagaraja, T. 2012. A microbiologist's view on improving nutrient utilization in ruminants. In: 23rd Annual Ruminant Nutrition Symposium, University of Florida Dairy Extension, Gainesville, Florida, 149175.
- National Research Council. 2001. Nutrient Requirements of Dairy cattle. 7th Rev. Ed. Natl. Acad. Press, Washington, DC.
- Newbold, C. J. 2007. Microbial feed additives for ruminants. In: Wallace, R.J., Chesson, A. (Eds.). Biotechnology in Animal Feeds and Animal Feeding. Wiley \
 InterScience, pp. 259-278.
- Newbold, C. J., and L. M. Rode. 2006. Dietary additives to control methanogenesis in the rumen. Intl. Cong. Series 1293: 138–147.
- Newbold, C. J., R. J. Wallace and F. M. McIntosh. 1996. Mode of action of the yeast Saccharomyces cerevisiae as a feed additive for ruminants. Brit. J. Nutr. 76:249-261.
- Newbold, C. J., A. G. Williams, and D. G. Chamberlain. 1987. The in vitro metabolism of d, l-lactic acid by rumen microorganisms. J. Sci. Food Agric. 38:9–18.
- Nisbet, D. J., and S. A. Martin. 1991. Effect of a Saccharomyces cerevisiae culture on lactate utilization by the ruminal bacterium Selenomonas ruminantium. J. Anim. Sci. 69:4628-4633.

- Nocek, J. E. 1997. Bovine acidosis: Implications on laminitis. J. Dairy Sci. 80:1005-1028.
- Nocek, J. E., M. G. Holt, and J. Oppy. 2011. Effects of supplementation with yeast culture and enzymatically hydrolyzed yeast on performance of early lactation dairy cattle. J. Dairy Sci. 94:4046-4056.
- Orth, R. 1992. Sample day and lactation report, DHIA 200 Fact Sheet A-2, Mid-States DRPC, Ames, IA.
- Palmquist, D. L., A. D. Beaulieu, and D. M. Barbano. 1993. Feed and animal factors influencing milk fat composition. J Dairy Sci. 76:1753–1771.
- Paryad, A., and M. Mahmoudi. 2008. Effect of different levels of supplemental yeast (Saccharomyces cerevisiae) on performance: blood constituents and carcass characteristics of broiler chicks. Afr. J. Agric. Res. 3:835-842.
- Piva, G., S. Belladonna, G. Fusconi, and F. Sicbaldi. 1993. Effects of yeast on dairy cow performance, ruminal fermentation, blood components and milk manufacturing properties. J. Dairy Sci. 76:2717-2722.
- Plaizier, J. C., D. O. Krause, G. N. Gozho, and B. W. McBride. 2008. Subacute ruminal acidosis in dairy cows: The physiological causes, incidence and consequences.

 Vet. J. 176:21-31.
- Poppi, D. P., R. E. Hendrickson, and D. J. Minson. 1985. The relative resistance to escape of leaf and stem particles from the rumen of cattle. J. Agric. Sci. 105:9–14.
- Poppy, G. D., A. R. Rabiee, I. J. Lean, W. K. Sanchez, K. L. Dorton, and P. S. Morley. 2012. A meta-analysis of the effects of feeding yeast culture produced by

- anaerobic fermentation of Saccharomyces cerevisiae on milk production of lactating dairy cows. J. Dairy Sci. 95:6027-6041.
- Putnam, D. E., C. G. Schwab, M. T. Socha, N. L. Whitehouse, N. A. Kierstead, and B. D. Garthwaite. 1997. Effect of yeast culture in the diets of early lactation dairy cows on ruminal fermentation and passage of nitrogen fractions and amino acids to the small intestine. J. Dairy Sci. 80:374-384.
- Pyatt, N. A., G. D. Hufstedler, C. A. Guthrie, B. W. Hoffman, and K. Keffaber. 2016.
 391 Veterinary feed directive (VFD) and its practical considerations for the beef industry. J. Anim. Sci. 94(Suppl2):183-183.
- Quigley, J. D., L. B. Wallis, H. H. Dowlen, and R. N. Heitmann. 1992. Sodium bicarbonate and yeast culture effects on ruminal fermentation, growth, and intake in dairy calves. J. Dairy Sci. 75:3531-3538.
- Randolph, T. F., E. Schelling, D. Grace, C. F. Nicholson, J. L. Leroy, D. C. Cole, M. W. Demment, A. Omore, J. Zinsstag, and M. Ruel. 2007. Invited review: Role of livestock in human nutrition and health for poverty reduction in developing countries, J. Anim. Sci. 85:2788-2800.
- Rhoads, M. L., R. P. Rhoads, M. J. Van Baale, R. J. Collier, S. R. Sanders, W. J. Weber,
 B. A. Crooker, and L. H. Baumgard. 2009. Effects of heat stress and plane of nutrition on lactating Holstein cows: I. Production, metabolism, and aspects of circulating somatotropin. J. Dairy Sci. 92:1986–1997.
- Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analysis and its application to human foods. Pages 123-158 in The Analysis of Dietary Fiber in Food. W.P.T. James and O. Theander, eds. Marcel Dekker Inc., New York, NY.

- Robinson, P. H. 2002. Yeast products for growing and lactating dairy cattle: impacts on rumen fermentation and performance. In XII Int. Meet. Milk Meat Prod. Hot Clim., Mexicali, Mexico.
- Robinson, P. H. and L. J. Erasmus. 2009. Effects of analyzable diet components on responses of lactating dairy cows to Saccharomyces cerevisiae based yeast products: A systematic review of the literature. Anim. Feed Sci. Tech. 149:185-198.
- Robinson, P. H., and J. E. Garrett. 1999. Effect of yeast culture (Saccharomyces cerevisiae) on adaptation of cows to postpartum diets and on lactational performance. J. Anim. Sci. 77:988-999.
- Roenfeldt, S. 1998. You can't afford to ignore heat stress. Dairy Management. 35:6-12.
- Russell, J. B. 2002. Rumen Microbiology and Its Role in Ruminant Nutrition. Russell Publications, Ithaca, NY, USA.
- Salvati, G. G., N. N. Morais Junior, A. C. Melo, R. R. Vilela, F. F. Cardoso, M. Aronovich, R. A. Pereira, and M. N. Pereira. 2015. Response of lactating cows to live yeast supplementation during summer. J. Dairy Sci. 98:4062-4073.
- Sargent, G., and H. Wickens. 2004. Brewers' yeast in C: difficile infection: probiotic or B-group vitamins. Pharm. J. 273:230-231.
- Sauvant, D., S. Giger-Reverdin, and P. Schmidely. 2004. Rumen acidosis: Modeling ruminant response to yeast culture. Pages 221–229 in Alltech's 20th Annu. Symp. Biotechnol. Feed Ind. Reimagining the Feed Industry. T. P. Lyons and K. A. Jacques, ed. Nottingham University Press.

- Schingoethe, D. J., K. N. Linke, K. F. Kalschuer, A. R. Hippen, D. R. Rennich, and I. Yoon. 2004. Feed efficiency of mid-lactation dairy cows fed yeast culture during summer. J. Dairy Sci. 87:4178-4181.
- Seo J. K., S. Kim, M. H. Kim, S. D. Upadhaya, D. K. Kam, and J. K. Ha. 2010. Direct fed microbials for ruminant animals. Asian-Aust. J. Anim. Sci. 23:1657-1667.
- Shwartz, G., M. L. Rhoads, M. J. VanBaale, R. P. Rhoads, and L. H. Baumgard. 2009.

 Effects of a supplemental yeast culture on heat-stressed lactating Holstein cows. J.

 Dairy Sci. 92:935-942.
- Soccol, C. R., L. P. de Souza Vandenberghe, M. R. Spier, A. B. P. Medeiros, C. T. Yamaguishi, J. D. Lindner, A. Pandey, and V. Thomaz-Soccol, 2010. The potential of probiotics: a review. Food Technol. Biotech. 48:413-434.
- Sorum, H. 2006. Antimicrobial drug resistance in fish pathogens. In: Aarestrup, F.M. (Ed.), Antimicrobial Resistance in Bacteria of Animal Origin. ASMPress, Washington DC, pp. 213-238.
- Steinfeld, H., P. Gerber, T. Wassenaar, V. Castel, M. Rosales, and C. de Haan. 2006.

 Livestock's Long Shadow-Environmental Issues and Options. FAO Agriculture

 Technical Paper, Rome.
- Stone, C. W. 2006. Yeast products in the feed industry: a practical guide for feed professionals. Engormix, the largest online agricultural community in the world.
- Sukhija, P. S., and D. L. Palmquist. 1988. Rapid method for determination of total fatty acid content and composition of feedstuffs and feces. J. Agric. Food Chem. 36:1202-1206.

- Thayer, R. H., R. F. Burkitt, R. D. Morrison, and E. E. Murray. 1978. Efficiency of utilization of dietary phosphorus by caged turkey breeder hens fed ration supplemented with live yeast culture. In: Animal Science Research Report, Oklahoma. Agricultural Experiment Station, Oklahoma State University, Stillwater, OK, pp. 173–181.
- Thornton, P. K. 2010. Livestock production: recent trends: future prospects. Philos. Trans. R. Soc. B. 365:2853-2867.
- Trinder, P. 1969. Determination of glucose in blood using a glucose oxidase with an alternative oxygen acceptor. Ann. Clin. Biochem. 6:24-27.
- USDA. 1998. Food Composition Standard Release 12.
- USDA. 2007. Dairy 2007, Part I: Reference of Dairy Cattle Health and Management Practices in the United States, 2007 USDA-APHIS-VS, Fort Collins, CO.
- Van Soest, P. J. 1963. Use of detergents in the analysis of fibrous feeds. II. A rapid method for determination of fiber and lignin. J. Assoc. Off. Agric. Chem. 46:829-835.
- Van Soest, P.J., 1994. Nutritional Ecology of the Ruminant. Cornell University: Press Cornell University.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and non-starch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74:3583-3597.
- Vohra, A., and T. Satyanarayana. 2001. Phytase production by the yeast Pichia anomala. Biotechnol. Lett. 23:551-554.

- Vohra, A., S. Poonam, and A. Madan. 2016. Review article: Probiotic yeasts in livestock sector. Anim. Feed Sci. Tech. 219:31-47.
- Wallace, J. D., and R. J. Raleigh. 1960. The influence of yeast in a high roughage wintering ration for Hereford calves as measured by digestibility and performance. Proc. Annu. Mtg. Western Sec. Am. Soc. Anim. Prod.: Vol. II, Utah State Univ., Coll. Agric., Logan, UT, June 23-24.
- Wallace, R. J. 1994. Ruminal microbiology, biotechnology, and ruminant nutrition: Progress and problems. J. Anim. Sci. 72:2992-3003.
- Wallace, R. J., and C. J. Newbold. 1995. Microbial feed additives for ruminants. In:
 Fuller, R., Heidt, P.J., Rusch, V., Der Waaij, D. (Eds.). Probiotics: Prospects of
 Use in Opportunistic Infections. Institute for Microbiology and Biochemistry,
 Herborn-Dill, Germany, pp. 101–125.
- Wang, Z., M. L. Eastridge, and X. Qiu. 2001. Effects of forage neutral detergent fiber and yeast culture on performance of cows during early lactation. J. Dairy Sci. 84:204-212.
- Wiedmeier, R. D., M. J. Arambel, and L. Walters. 1987. Effect of yeast culture and Aspergillus oryzae fermentation extract on ruminal characteristics and nutrient digestibility. J. Dairy Sci. 70:2063.
- Wildman, E. E., G. M. Jones, P. E. Wagner, R. L. Boman, H. F. Troutt Jr., and T. N. Lesch. 1982. A dairy cow body condition scoring system and its relationship to selected production characteristics. J. Dairy Sci. 65:495-501.

- Williams, A. G., and G. S. Coleman. 1988. The rumen protozoa. In: Hobson, P.N. (Ed.),

 The Rumen Microbial Ecosystem. Elsevier Science Publishing Company, New

 York, 77.
- Williams, A. G., and G. S. Coleman. 1997. The rumen protozoa. In: Hobson, P.N., Stewart, C.S. (Eds.), The Rumen Microbial Ecosystem. Blackie Academic and Professional, London, pp. 73–120.
- Williams, P. E. V., and J. C. Newbold. 1990. Rumen probiosis: the effects of novel micro organisms on rumen fermentation and rumen productivity. In:Haresign, W., Cole,D.J.A. (Eds.), Recent Advances in Animal Nutrition. Butterworths, London, UK.
- Williams, P. E. V., C. A. G. Tait, G. M. Innes, and C. J. Newbold. 1991. Effects of the inclusion of yeast culture (Saccharomyces cerevisiae plus growth medium) in the diet of dairy cows on milk yield and forage degradation and fermentation patterns in the rumen of steers. J. Anim. Sci. 69:3016-3026.
- Wohlt, J. E., T. T. Corcione, and P. K. Zajac. 1998. Effect of yeast on feed intake and performance of cows fed diets based on corn silage during early lactation. J. Dairy Sci. 8:1345-1352.
- Wohlt, J. E., A. D. Finkelstein, and C. H. Chung. 1991. Yeast culture to improve intake, nutrient digestibility, and performance by dairy cattle during early lactation. J. Dairy Sci. 74:1395-1400.
- Yirga, H. 2015. The use of probiotics in animal nutrition. J. Probiot. Health. 3:132.
- Yoon, I. K., and M. D. Stern. 1995. Influence of direct-fed microbials on ruminal microbial fermentation and performance of ruminants: A review. Austral. Asian. J. Anim. Sci. 8:533-555.

- Yoon, I. K., and M. D. Stern. 1996. Effects of Saccharomyces cerevisiae and Aspergillus oryzae cultures on ruminal fermentation in dairy cows. J. Dairy Sci. 79:411–417.
- Yousefian, M., and M. S. Amiri. 2009. A review of the use of prebiotic in aquaculture for fish and shrimp. Afr. J. Biotech. 8:7313-7318.
- Ziemer, C. J., and G. R. Gibson. 1998. An overview of probiotics: prebiotics and synbiotics in the functional food concept: perspectives and future strategies. Int. Dairy J. 8:473-479.