1983

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Recommended Citation

Finn, Anne M., "The Use of Whole-Rolled Sunflower Seeds with or without Additional Limestone in Lactating Dairy Cattle Rations" (1983). *Electronic Theses and Dissertations*. 1312.
https://openprairie.sdstate.edu/etd/1312

THE USE OF WHOLE-ROLLED SUNFLOWER SEEDS WITH OR WITHOUT ADDITIONAL LIMESTONE IN LACTATING DAIRY CATTLE RATIONS

BY

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A thesis submitted
in partial fulfillment of the requirements for the
degree of Master of Science
Major in Dairy Science
South Dakota State University
1983

THE USE OF WHOLE-ROLLED SUNFLOWER SEEDS WITH OR WITHOUT ADDITIONAL LIMESTONE IN LACTATING DAIRY CATTLE RATIONS

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

David J. Schingoethe Date
Thesis Advisor

John G. Parsons Date
Head, Dairy Science Dept.

DEDICATION

My thesis is dedicated to my father, mother, and my sister, Irene, for their support and encouragement throughout my college education.

ACKNOWLEDGMENTS

I would like to express my gratefulness to Dr. David J. Schingoethe for his guidance in the writing of this thesis, and to Dr. Andrew K. Clark for the planning of this research. The encouragement of Drs. John G. Parsons and Clarence Moore in all areas of my education are also appreciated.

A very special thanks to Dr. Tilahun Sahlu for his help throughout my study, and to Kari Wright who helped in the analysis of the research. Thanks goes to my fellow graduate students and members of the farm crew for their cooperation in the experiment. Special thanks to Laura Lardy for her friendship and excellent typing skills in preparing this thesis.

Finally, the unforgettable friends I've made here during my stay in South Dakota who have made me feel at home. Thanks Andy, Milly, Ray, Roxy, Zeno, and Chelle.

AMF

THE USE OF WHOLE-ROLLED SUNFLOWER SEEDS WITH OR WITHOUT ADDITIONAL LIMESTONE IN LACTATING DAIRY CATTLE RATIONS

ABSTRACT

Thirty Holstein cows were utilized in a lactation trial evaluating diets containing whole-rolled sunflower seeds. Cows were randomly assigned to treatments, grouped according to previous milk production and lactation number. The cows were fed one of three diets: 1) the control was a corn-soybean meal mix (CSM), 2) corn-soybean meal mix plus 22% whole-rolled sunflower seeds, and 3) a corn-soybean meal mix with 22% whole-rolled sunflower seeds with an additional 3.5% limestone over SCS (SCL). Additional limestone was added in an attempt to form fatty acid insoluble soaps. Rations were formulated to be isonitrogenous at 16% crude protein, fed as a complete mixed ration, and were fed ad libitum. Diets were formulated to contain 45% corn silage, 10% alfalfa hay, and 45% of the respective concentrate mix. Cows were fed the rations from 21 days postpartum and continued for 120 days. Milk production (32.2, 32.0, and 32.8 kg/day for CSM, SCS, and SCL, respectively) was similar for all three diets. However, 4% fat-corrected milk (30.2, 28.1, and 30.2 kg/day) was lower when fed SCS because of lower milk fat percentages (3.57, 3.19, and 3.51%). Milk protein (3.01, 2.97, and 2.90%) and total solids (11.99, 11.65, and 11.87%) were similar when fed all three diets. Dry matter intakes were 21.0, 18.4, and 20.0 kg/day for CSM, SCS, and SCL, respectively. No differences were observed in volatile fatty acids (molar %, concentration) and pH among the three treatments. Serum cholesterol values were higher for treatments containing 22% whole-rolled sunflower seeds, 166.0, 223.0, and 255.0 mg/dl for the respective diets.

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INTRODUCTION

Most dairy cows do not consume enough energy to meet their production needs during the first few months of lactation (76). Adding fat to the diet is often effective in increasing the energy density of the diet, which may help cows come closer to consuming sufficient energy (76).

Sunflower seeds could be an attractive fat supplement for dairy cattle since the sunflower seeds contain 40 to 45% fat (57). In recent years, sunflowers have become an established crop in Minnesota, North Dakota, South Dakota and Texas (46). Most growers raise sunflower seeds with the intent of selling them as a cash crop; however, in certain situations (e.g., insect or weather damage, surplus production) farmers may find it more profitable to feed sunflower seeds to cattle than to market the seeds as a cash crop.

Added dietary fat usually decreases fiber digestibility in the rumen; therefore, it may be advantageous to bypass the fat through the rumen. Adding calcium to high fat diets may increase fat utilization by forming insoluble calcium soaps of the fatty acids, thereby bypassing the rumen. The objective of this research was to evaluate sunflower seeds with and without additional calcium for lactating dairy cows during early lactation.

LITERATURE REVIEW

Practical Application of Feeding Fat

There are several reasons why the feeding of fat has increased in popularity: 1) The need to increase the energetic density of a ration. Feeding added fat has resulted in the maintenance or an increase in milk production (2, 6, 95), milk fat percent (5, 68), and an increase in the energetic efficiency of dairy cows (87, 96). 2) An attempt to eliminate the negative effects of high concentrate carbohydrate feeding such as acidosis, reduced feed intake, and overweight animals (9). 3) The increased availability of fats that are suitable for incorporation into animal rations has also encouraged the feeding of additional fat (34, 36, 46, 72). Several sources of fat have been used, including cottonseed oil (15), whole cottonseed (2), soybean oil (49), whole soybeans (59, 70, 73), tallow (15, 66), formaldehyde treated lipids (13, 26), and sunflower seeds (58, 72).

Studies indicate a varied response to unsaturated oils on milk fat percent. Earlier studies (64, 74, 86, 87) with protected unsaturated oil depressed milk fat percent. Recent research (58, 72) indicated a slight increase in maintenance of fat percent when unsaturated oils were fed.

Adding fat to the ration is very efficient in increasing the caloric density of the diet; however, there are some adverse affects to additional fat. Polyunsaturated fatty acids are apparently inhibitory to microbial growth. Davison et al. (23) observed that feeding dietary unsaturated fat reduced fermentation of fiber in the rumen and absorption of divalent cations from the small intestine. Macleod et al. (53) and Kronfeld et al. (47) found that feeding saturated dietary fat did not adversely affect digestion. This inhibition of polyunsaturated fatty acids on microbial growth may be reversed by the addition of fiber and/or calcium ions, both of which likely act by removing fatty acid from the microbial cell surface (67).

Brooks et al. (14), in 1954, first reported that alfalfa ash reversed inhibitory effects of added corn oil on fiber digestibility. It was later demonstrated that the effect was due to calcium in the ash (37, 97). Palmquist et al. (67) theorized that calcium improves fiber digestibility by forming insoluble soaps which remove the fatty acids from solution so they are no longer available to bind to rumen microbiota (67). The insoluble soaps which are formed are bypassed to the lower gut for absorption.

Another means of bypassing the rumen is with the use of protected feeds (35, 67, 77, 95, 96). Polyunsaturated oils are coated with a protein and then treated with formaldehyde. This protects the oils from microbial attack by making the oils insoluble in the rumen. Protected lipids then pass thru the rumen but are hydrolyzed after being solubilized under the more acidic conditions in the abomasum. The oils are then solubilized, hydrolyzed, absorbed, and can be incorporated into milk or body fat.

There are two major properties of fat which may influence their effect on digestion in the rumen: unsaturation and esterification (67). Researchers (30, 67) theorized that large quantities of unesterified acids, particularly polyunsaturates, are inhibitory to microbial growth.

There are varying results of feeding added fat on the dry matter intake. Most studies indicate that feed intake is not affected by fat or oil supplementation (35, 48, 52, 72). However, Palmquist and Conrad (65) reported that dry matter intake was increased when cows were fed a high fat ration. Macleod et al. (53) found that intakes of total dry matter from concentrates were decreased, dry matter from forage increased, and energy intake was unchanged by feeding protected fat. Similar results were reported by Smith et al. (79).

Rumen Metabolism of Lipids

In herbivora, 20% or more of the ration may consist of substances that can be digested only by the action of microorganisms (53). Thus, the factors which may govern this activity are obviously of great importance. The rumen microbial population promptly hydrolyzes the triglycerides from cereal grains and the galactolipids from forages, releasing free fatty acids (FFA) and allowing the glycerol and galactose to be fermented to volatile fatty acids (VFA).

Since the ruminal environment is highly reduced, the unsaturated fatty acids are rapidly hydrogenated to a saturated form with stearic acid predominating (55). Biohydrogenation is a multi step process. Viviani et al. (17) and Hawke et al. (17) observed that biohydrogenation of unsaturated fatty acids proceeded to completion only in the presence of the food particle fraction of rumen contents. Removal of the food particles greatly reduced the capacity of rumen contents to biohydrogenate. Harfoot et al. (17) investigated the adsorption of unesterified fatty acids to rumen bacteria and found that saturated fatty acids were much more tightly bound to the bacterial cells than were unsaturated acids of the same chain length. Increasing unsaturation resulted in decreased adsorption and in all cases the addition to the incubation mixture of either food particles from the rumen or inert solids such as silica created reduced the extent to which fatty acids became adsorbed to bacterial cells. It was also noted by Henderson et al. (17) that long chain fatty acids became inhibitory to the growth of rumen bacteria when their concentrations were greater than could be adsorbed onto the bacterial cell.

Recent emphasis has been placed upon avoiding triglyceride hydrolysis in the rumen through "protecting" lipids from the rumen microbes by treating associated proteins with formalin or heat, or by feeding whole oilseeds. The feeding of protected tallow tended to improve digestibility of dry matter, energy, and protein, but to diminish digestibility of fiber, calcium, and magnesium (48). Lucas et al. (50) reported similar results when protected tallow, vegetable oils, and fatty acids were fed to cows.

Fiber Digestibility

Studies indicated that feeding fat usually depressed fiber digestibility in sheep (14, 24) and cattle (96, 97). Few data are available on effects of fat on digestibility of rations in lactating cows (58, 65, 72). Digestibility studies with lactating cows may not show negative effects of fat on fiber digestibility because of greater intake and rate of passage (91), as well as possible compensatory increases in digestion by the lower digestive tract (12).

There are four theories, as summarized by Devendra and Lewis (24), to explain reduced fiber digestibility often observed with added fat. 1) Physical coating of the fiber with fat, preventing microbial attack. Studies (67) indicated that fatty acids inhibit rumen bacteria in pure culture, possibly by binding to the microbial cells. This binding can be reduced by adding fiber. 2) A modification of rumen microbial population from possible toxic effects of fat on certain microorganisms. Fatty acids are known to be inhibitory to the cellulolytic and methanogenic bacteria (29, 41). 3) Inhibition of microbial activity from surface-active effects of fatty acids on cell membranes. Fatty acids inhibit amino acid uptake and energy metabolism by the cellulolytic and methanogenic bacteria (31). 4) Reduced cation availability from formation of insoluble complexes with long chain fatty acids. This last effect could be directly on availability of cations for microbial formation or indirectly affecting rumen pH (67).

Calcium Addition

The addition of calcium to high fat rations has reversed depressed fiber digestibility in vitro (44) and in vivo with steers (45) and sheep (24, 28). Other divalent ions such as magnesium, barium, berryllium, strontium, and iron were ineffective in reversing inhibition in bacterial cultures (30).

Researchers (67) postulated that calcium improves fiber digestibility by forming insoluble soaps which remove the fatty acids from solution so they are no longer available to bind to rumen microbiota. Experiments (23, 37) confirmed that calcium improved digestion in fat-supplemented rations, but there was also an increase in the amount of fecal soaps excreted when fed those diets. Jenkins and Palmquist (42) measured a 30% increase in insoluble soaps in the rumen of cows fed tallow and 2% limestone. Fiber digestibility increased when tallow fatty acids were added as calcium soaps (43). Later research showed that the likelihood of forming insoluble soaps with calcium increased as chain length and saturation of the fatty acids increased (44).

Volatile Fatty Acids

In vitro research conducted by Varman et al. (93) demonstrated that the addition of unsaturated fatty acids of 18 carbon chain length to rumen fluid increased the production of propionate and lactate from pyruvate and glucose. The mechanism suggested was a direct toxic effect of unsaturated fatty acids upon methanogenic bacteria as well as a shift in hydrogen utilization from methane production toward propionic acid production. Czerkawski et al. (21) found a decrease in methane production in vivo following infusion of unsaturated fatty acids in the rumen of sheep. Research (2, 12, 57, 58) indicated that feeding fat usually increased the concentration of propionate, resulting in a narrower ratio of acetate to propionate. This was due to the depression of methanogenic and cellulolytic organisms in the rumen, causing a shift towards increased propionate production at the expense of acetate and butyrate production (93).

Bauman et al. (10) fed either a control or high grain, low fiber diet and reported that one of the major factors affecting ruminant propionate production was energy intake in reference to needs of the animals. When digestible energy intake was restricted to near maintenance requirements, propionate production per kilocalorie of digestible energy intake was higher than when animals were fed ad libitum.

When using tallow and cottonseed oil as sources of fat, Brown et al. (15) found that the influence on volatile fatty acid (VFA) production was more marked on low roughage than on high roughage diets. Mattos et al. (54) reported that the production of acetic acid in the rumen of cows fed the unprotected fat diets was less than in those fed the protected fat diet.

Intestinal Absorption

Digestibilities of unsaturated fatty acids are lower for ruminant's than for nonruminant's, whereas, the opposite occurs with saturated fatty acids (67). This lower digestibility in ruminants is due to the difference between ruminants and nonruminants lipid metabolism. Lipid in ruminant feed is largely present in the form of thin layers of free fatty acids on the surface of the feed particles. Due to the low bicarbonate content of pancreatic secretions, the pH of the proximal one-half of the ruminant intestine remains relatively acid (3). This low pH increases solubility of fatty and bile acids, and may also solubilize calcium soaps (94). This would allow higher absorption of both fatty acid and calcium than would be possible at neutral or alkaline pH (67). Active micelle formation of the fatty acids occurs in the upper tract under the influence of bile salts and ingesta phospholipids. The larger percentage of lipid absorption is in the lower three quarters of the jejunum where the pancreatic phospholipase has had a chance to hydrolyze lecithin into a fatty acid and the highly polar lysolicithin, which further enhances micelle formation (55).

Fatty acids enter the intestine nonionically bound to particulate matter. In the acid milieu of the duodenum, taurocholic acid, phosphatidyl choline, and phosphatidyl ethanolamine emulsify the fatty acids followed by dispersion into micelles (67). The mixed micelles enter the microvilli (brush border) and the apical pole of the absorption cells by simple diffusion. The conjugated bile salts are not absorbed during lipid absorption but move along the tract and are absorbed in the lower region of the ileum. The absorption of the conjugated bile salts involves an energy-requiring active tranport (89).

Serum Cholesterol

Most evidence indicates that concentrations of all plasma lipids, with the possible exception of triglycerides which turn over rapidly, are elevated by feeding dietary supplements of unprotected polyunsaturated fatty acids to ruminants for comparatively short feeding periods (2 to 3 wk), although possibly not over longer feeding periods (18, 52, 85, 93). Similarly, concentrations of all lipid classes in plasma may be elevated by feeding protected polyunsaturated fatty acids (6). In particular, free cholesterol concentrations were raised two to three fold in lactating cows (6, 13, 97) and steers (32), although the effect was less pronounced in calves (96) and lambs (25). This appeared to be a consequence of increased cholesterol synthesis in the intestine and in lactating animals of decreased removal of cholesterol from the plasma by the mammary gland.

Nestle et al. (61) found that supplementing the diet of sheep and goats with fat significantly enhanced sterolgenesis in the small intestine in vivo. The hypercholesterolemia that developed in ruminants fed fat appeared to be primarily due to an increased intestinal biosynthesis of cholesterol, but may also have been partly due to a decreased fecal excretion of bile acids.

Milk Production and Composition

Most conventional ruminant rations contain sufficient fat to maintain optimum production (67). Earlier work reported by Banks et al. (6) reported that a low fat (.7% of total ration dry matter) ration limited milk production, and that fat supplemented diets increased milk yields by 25 to 40% and fat yields by 44 to 69% compared to fat deficient diet. Kronfeld et al. (47) found that mammary uptake of glucose appears to be a major determinant of the rate of milk secretion.

Results are not conclusive on the effect added fat has on milk production. Experiments conducted from the late 1920's to the mid 1940's consistently demonstrated increased milk production (range 2 to 10%) by cows fed concentrates containing 4 to 7% fat as compared to control diets containing 1 to 3% fat (67). Other research (79) showed no effect on milk yield when feeding protected tallow to dairy cows.

Nevens (62), in 1926, attempted to increase the fat content of the ration by the addition of fat or feeds high in fat. He concluded that the percentage of fat in milk can be materially increased by the feeding of rations high in oil compared with rations low in oil, but furnishing an equal amount of total digestible nutrients. The increased fat content was due to the oil itself and not a stimulation due to a larger excess of energy in the rations.

The length of the feeding period appeared to be important. In most comparatively long term (4 to 6 wk periods on each treatment) feeding trials (1, 15, 36, 69) in which vegetable oil supplements were added to the diets, there was either no significant effect on milk fat yield or it was depressed relative to that of control animals receiving no such dietary supplement. On the other hand, in some experiments in which these supplements were fed for shorter periods (2 to 3 wk), milk fat increased slightly (7, 54, 93). Steele and Moore (81) found that when dietary supplements of cottonseed oil (10% of the concentrate) were fed to lactating cows, the milk fat yield increased during the first 8 days of the 4 wk treatment period, but was markedly depressed during the last 4 days. Possibly the rumen microbial populations changed with time under the influence of comparatively large amounts of dietary unsaturated fatty acids, which led to changes in the availability of essential metabolites to the host animals (83).

The physical state in which the oil is supplied may also be important. In one experiment (83), soybean oil itself depressed milk fat yield considerably although the yield was actually increased when an equivalent amount of oil was fed in the form of intact soybeans. Adding polyunsaturated oils to normal rations for dairy cows depressed milk fat percentages, but increased the proportion of unsaturated fatty acids in the milk fat (26, 36, 60, 62, 78, 83, 93).

There have been several theories proposed throughout the years in an attempt to explain milk fat depression when diets are supplemented with polyunsaturated oils. These theories include: 1) a shift in VFA production (78), 2) configuration changes in the dietary fatty acids by rumen microorganisms (60), 3) depressed de novo synthesis of fatty acids in the mammary gland (67), and 4) reduced uptake of precursors of fatty acids by the mammary gland (78). However, fat depression could be a combination of several factors.

Shaw et al. (78) theorized that feeding unsaturated oils caused a shortage of acetate in rumen fermentation. Davis (22) later reported acetate production rates were unaffected by the diet. Shaw and Ensor (78) reported an increase in propionate production, which probably accounted for the narrowed acetate to propionate ratio. This narrowed ratio could induce a glucogenic response of adipose tissue which causes it to: 1) compete with mammary gland for lipogenic substrate, reducing the amount of acetate available to the mammary gland;

2) take up and esterify increased amounts of dietary long chain fatty acids; and 3) decrease mobilization of adipose long chain fatty acids for milk fat synthesis (67).

McCullough (56) showed that there was an excellent correlation between rumen propionate concentrations and degress of milk fat depression. Elevated propionate levels might simply diminish the availability of β-hydroxybutyrate for fatty acid synthesis (92). A more likely explanation (17) is that increased propionate concentrations in the rumen lead to increased lactate and glucose production. That in turn stimulates insulin production, reducing the rate of release of free fatty acids from adipose tissue. Accordingly, there would be a diminished availability of preformed long chain fatty acids for milk fat synthesis.

Intravenous infusion of polyunsaturated oils resulted in a higher proportion of unsaturated fatty acids in milk fat and an increase in the content and yield of fat in milk (68). Depression of milk fat also did not occur when oils were infused into the abomasum (75) or when fats protected from ruminal biohydrogenation were fed (5, 54, 97).

Unsaturated oils fed either protected or unprotected suppressed de novo synthesis of fatty acids in the mammary gland (67). Toussant et al. (90) showed that polyunsaturated fats suppressed fatty acid synthetase activity. Moore and Steele (60) reported that with increased uptake of 18-carbon fatty acids by both unprotected and protected oil fed cows, there was inhibition of the key enzymes of fatty acid biosynthesis. This increased uptake of 18-carbon fatty acids was at the expense of the amount of short and medium fatty acids synthesized (67).

All this evidence appears to indicate that if the rumen could be bypassed, or if the process of metabolism in the rumen modified, the adverse effects on milk fat content when feeding cows a higher amount of polyunsaturated oil may be avoided. In addition, the oil may be more efficiently utilized for milk synthesis.

Fatty Acid Composition

The fatty acids that are present in milk fat of the cow are derived from two metabolic processes. The first, acetate and β-hydroxybutyrate, are taken up from the blood and are than utilized as precursors for the de novo synthesis of fatty acids in the mammary gland (82). In the second process, triglycerides in the chylomicrons and low density lipoproteins of the blood are taken up by the mammary gland. These triglycerides are partially or completely hydrolyzed and the resulting fatty acids are incorporated into new triglycerides by the mammary gland. Under normal dietary conditions, the major fatty acids present in blood triglycerides of the cow are 16:0, 18:0, and 18:1, and these are taken up by the mammary gland from the blood.

In virtually all experiments (7, 15, 19, 36, 54, 69, 71, 81, 84, 96) involving the feeding of dietary unsaturated fatty acids, the relative proportions of 18:0 and 18:1 fatty acids in milk increased markedly, whereas those of the medium chain-length fatty acids tended to decrease relative to the corresponding values from control animals. There were rarely any changes in the proportions of linoleic and linolenic acids. Inhibition of fatty acid synthesis in the mammary gland may have been due in part to reduced availability of acetate as substrate, but in greater measure to inhibition of one of the enzymes of the fatty acid synthetase, probably acetyl CoA carboxylase (17).

The increased amount of 18:1 acids in milk when fed high unprotected oil diets was mainly due to an increase in the trans isomers of 18:1 (60). In animals fed a basal ration, the C18:1 of milk fat was Cis-9-octadecenoate and was produced in part by desaturation of stearic acid. Trans-octadecenoates are normal intermediates in the hydrogenation process of rumen microbes (39), and when increased fatty acids are fed, more trans-acids will reach the mammary gland.

Protein Composition

Protein composition of milk is influenced by supplementing the ration with fat. Concentration of milk protein has been reduced (6, 26), particularly with feeding protected lipid (53). While other researchers (35, 68, 97) found that feeding protected polyunsaturated fats held or increased protein percent. A cause for reduced protein percent could be that significant quantities of dietary energy are unavailable for rumen fermentation and microbial growth, potentially decreasing availability of microbial protein and glucose precursors to the animal (79).

MATERIALS AND METHODS

Thirty lactating Holstein cows were utilized in a lactation trial evaluating diets containing whole sunflower seeds or soybean meal as a protein source. Cows were assigned to one of three treatments in a completely random design trial. Cows were placed on the trial 3 wk postpartum and continued on the trial for 120 days.

Treatments were designated corn-soybean meal mix (CSM), sunflower-corn soybean meal mix (SCS), and mix SCS plus 3.5% additional limestone (SCL) (Table 1). Diets were formulated to be isonitrogenous at 16% crude protein and to contain 45% corn silage, 10% alfalfa hay, and 45% concentrate mix on a dry matter basis. However, due to the underestimated dry matter of the silage, actual dry matter coming from the three feed sources was 47.0, 9.2, and 43.8%, respectively.

Cows were housed in a free stall barn and individually fed ad libitum. Amounts of feed fed and weighbacks were recorded daily. Cows were weighed 3 consecutive days prior to going on the trial, twice monthly, and three times at the end of the trial.

Cows were milked twice daily throughout the trial. Two 24 h milk samples were collected during the week before cows went on the trial and then collected every 2 wk throughout the trial, and analyzed for protein by Kjeldahl (4), milk fat by Milk-O-Tester[1], and total solids by Mojonnier method (63). Milk samples were lyophilized and the fat extracted for determination of fatty acid composition by the

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[1] MK-II, N. Foss Electric, Hillerod, Denmark.

TABLE 1. Composition of concentrate mixes[a,b].

Ingredients	Diets CSM	 SCS	 SCL
	(%, dry matter basis)		
Ground corn	60	45	40
Soybean meal, 44% CP	38	31	32.5
Rolled sunflower seeds	--	22	22
Limestone	1	1	4.5
Trace mineral salt	1	1	1

[a] CSM = corn-soybean meal (Control); SCS = corn-soybean meal-sunflower seeds; SCL = SCS plus additional limestone.

[b] Mixes also contained 8800 IU added vitamin A and 2200 IU added vitamin D/kg.

Roese-Gottleib procedure (63). Analysis of fat was by gas-liquid chromatography using a stainless steel Supelco SP 2330 Cyanosilicone Column[1]. A .5 ml sample was injected onto the column which was maintained at a temperature of 200°C and a flow rate of 20 ml/min.

Rumen fluid was sampled monthly, 2 to 4 h after AM feeding, by esophageal tube and suction strainer into 300 ml bottles containing .5 ml saturated mercuric chloride. Samples were measured immediately for pH, and prepared for later analysis of ammonia (16) and volatile fatty acids (VFA) (11). Volatile fatty acids were determined by gas-liquid chromatography with a neopentyl succinate column as described by Baumgardt (11). Jugular blood samples were collected from each cow when rumen samples were taken and serum samples analyzed for total cholesterol[2].

Feeds were sampled weekly, and samples were composited monthly for analyses. Samples were oven dried at 57°C for 48 h and ground through a 2 mm screen in a Wiley Mill. All analyses were conducted on dried samples. Samples were analyzed for crude protein, ether extract, and ash (4); neutral detergent fiber (NDF), and acid detergent fiber (ADF) (35). Soluble nitrogen in feeds was measured using 10% Burroughs' mineral buffer as described by Crooker et al. (20). Gross energy was determined in feeds using a Parr Adiabatic Bomb calorimeter.

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[1] Supelco, Inc., Bellefonte, Pennsylvania.

[2] Accu-zyme II Cholesterol Reagent, Coulter Electronics, Hialea, FL.

Data were analyzed by analysis of variance by General Linear Models procedure (SAS Institute, Cary, NC) (8). Milk production and composition data were adjusted for pretreatment production and composition by covariate analysis, using production the 3rd wk postpartum and pretreatment milk composition as covariates. Differences among treatment means were evaluated using the Waller-Duncan range test (80).

RESULTS AND DISCUSSION

Feed Composition

Chemical composition of the ration components appears in Table 2. Concentrate mixes were formulated to be isonitrogenous, but the sunflower seed containing mixes had slightly less crude protein than the CSM mix, due to the underestimated crude protein content of the sunflower seeds. Total ration crude protein (% of DM) was 16.6, 16.3, and 16.1 for rations CSM, SCS, and SCL, respectively. Ether extract values were higher for the sunflower seed rations (2.5, 6.1, and 5.7% for total ration DM, respectively) due to the increased fat content from sunflower seeds. Concentrate mix SCL had a slightly lower ether extract value than SCS mix, possibly because the added calcium as limestone may bind with the fat in the feed and cause incomplete fat extraction by ether. Ash content increased with the addition of the sunflower seeds in the ration and was the highest for the SCL ration. Fiber components, acid detergent fiber (19.5, 22.7, and 22.4% of total ration DM, respectively), and neutral detergent fiber (36.8, 39.0, and 38.9% of total ration DM, respectively) were higher in the sunflower seed rations, reflecting the greater fiber content of sunflower seeds. Soluble N was similar for all three rations (36.3, 37.7, and 36.9% of total ration DM, respectively). Gross energy values of the concentrate mixes reflected the addition of fat, 4.3, 4.6, and 4.5 kcal/g, respectively, for the concentrate mixes.

TABLE 2. Chemical composition of ration components.

Component	Alfalfa hay	Corn silage	Concentrate mix		
			CSM	SCS	SCL
			(%)		
Dry matter (DM)	88.0	41.6	89.4	90.1	91.0
			(% of DM)		
Crude protein	15.9	8.8	25.1	24.5	24.0
Ether extract	1.3	2.5	2.8	11.0	10.0
Ash	10.4	4.5	5.4	6.0	8.2
Acid detergent fiber	44.6	25.6	8.5	15.0	14.4
Neutral detergent fiber	58.1	45.4	23.2	28.1	27.9
			(% of N)		
Soluble nitrogen	43.0	54.7	15.1	18.4	16.6
			(kcal/g)		
Gross energy	3.9	3.9	4.3	4.6	4.5

Yield and Composition of Milk

Average yield and composition of milk for the experiment appear in Table 3 while milk production and composition by weeks is illustrated in Figures 1 to 4. Milk production was similar among the three treatment groups. Similar results were reported by other studies (15, 67). Brown et al. (15) reported that 6% added tallow or cottonseed oil had no effect on milk production. Cows fed the control diet (CSM) attained peak milk sooner than cows fed sunflower seeds (Figure 1). This possibly indicated slower adaptation to sunflower seed diets. However, cows fed SCS and SCL rations were more persistent in maintaining milk production.

Cows fed SCS produced less ($P<.01$) 4% fat-corrected milk (Table 3, Figure 2) because of lower ($P<.01$) milk fat percentages. Depression in milk fat with the SCS ration did not occur until 6 wk after the trial started (Figure 3). This depression could be due to several factors. One factor is possibly the rumen microbial populations changed with time under the influence of comparatively large amounts of dietary fatty acids (83). The addition of limestone to sunflower seeds apparently prevented the milk fat depression experienced with the SCS ration. Milk fat percentages for cows fed SCL were similar to those for cows fed CSM; however, the cows fed SCL had a slight decrease in fat percent after about 8 wk on the experiment (Figure 3).

McGuffey and Schingoethe (58) observed no decrease in fat percentage when they fed sunflower seeds. However, their trial was short in

TABLE 3. Milk production and milk composition for cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone[a].

Components	Diets			SE^d
	CSM	SCS	SCL	
Milk production (kg/day)	32.20	31.96	32.84	.35
4% Fat corrected milk (kg/day)	30.21^b	28.06^c	30.34^b	.37
Fat (%)	3.57^b	3.18^c	3.51^b	.145
Protein (%)	3.01	2.97	2.90	.064
Total solids (%)	11.99^b	11.65^c	11.87^b	.171
Solids-not-fat (%)	8.41	8.46	8.37	.143

[a] Values covariate adjusted using pretreatment milk production and composition.

[b,c] Means with different superscripts are different ($P<.01$).

[d] Standard error of mean.

Figure 1. Milk production of cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

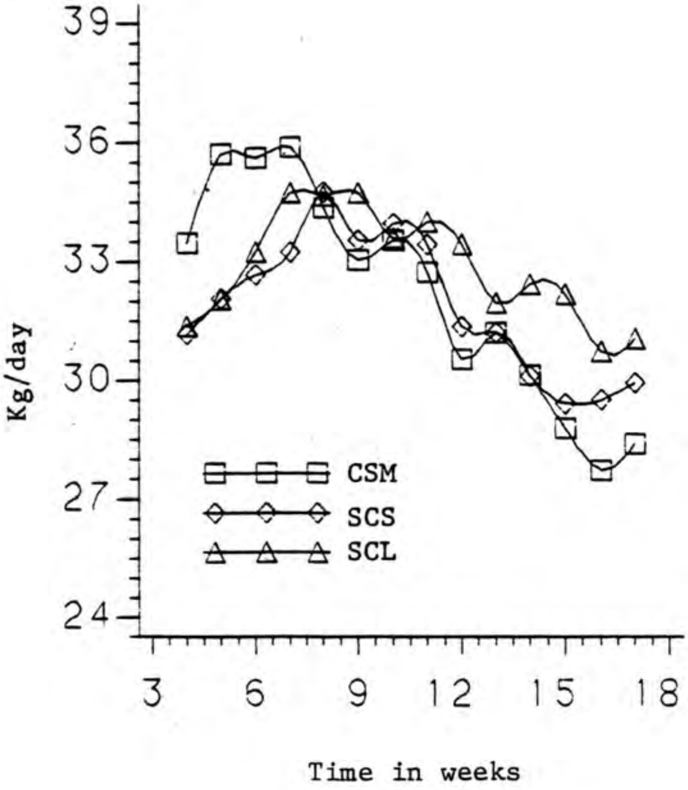

39
36
33
30
27
24
Kg/day
3
6
9
12
15
18
Time in weeks
CSM
SCS
SCL

Figure 2. Fat corrected milk production of cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

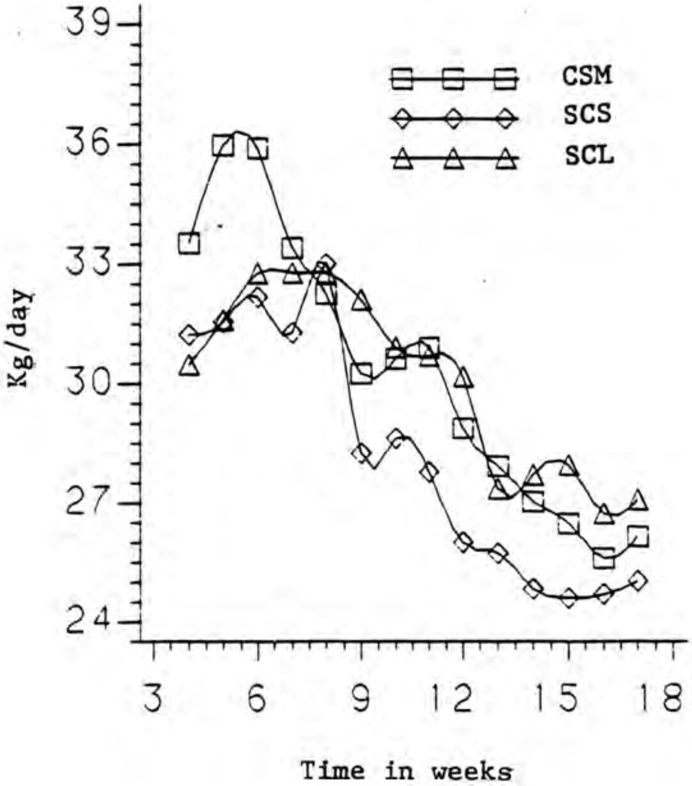
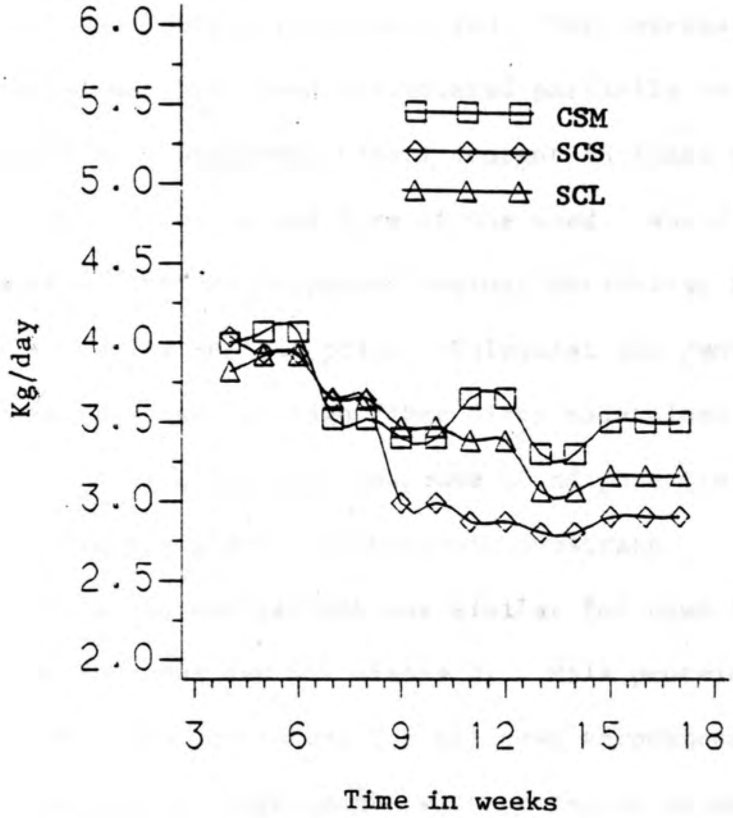

CSM
SCS
SCL
Kg/day
39
36
33
30
27
24
3
6
9
12
15
18
Time in weeks

Figure 3. Fat percent of milk from cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

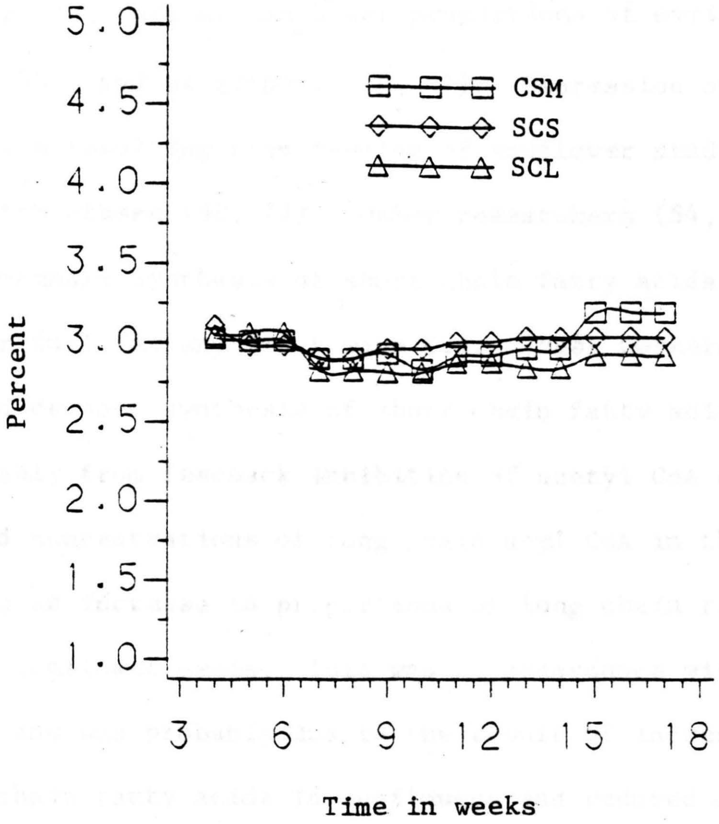

6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
Kg/day
CSM
SCS
SCL
3
6
9
12
15
18
Time in weeks

duration (28 day periods) which was not long enough in duration to detect the difference observed in this experiment. Rafalowski and Park (72) also did not depress fat percentages when 10, 20, and 30% sunflower seed concentrates were fed. Fat depressing effects of dietary oil may have been compensated partially by the relatively high fiber (26% acid detergent fiber) content in these high sunflower seed diets (72), or due to the form of the seed. Whole sunflower seeds may have more effectively bypassed ruminal metabolism than rolled sunflower seeds fed in this trial. Palmquist and Jenkins (67) suggested that cows fed high fat-high fiber diets maintained milk fat test by prevention of a glucogenic response of adipose tissue which competes with the mammary gland for lypogenic substrate.

Milk protein percent was similar for cows fed CSM and SCS, but was lower for cows fed SCL (Table 3). Milk protein percent, unlike fat percent, was consistent for all cows throughout the experimental trial (Figure 4). Experiments with protected animal or vegetable fats have increased (5) or decreased (53, 77) protein content of milk. McGuffey and Schingoethe (58) and Rafalowski and Park (72) did not report any effect on milk protein percent. Results imply that milk protein content may be influenced by type, amount, and degree of fatty acid saturation of lipid supplements.

Total solids content of milk was similar when fed diets CSM and SCL, but lower when fed SCS. This effect was mainly a reflection of fat and protein percentages. Rafalowski and Park (72) found a significant depression of total solids in milk from cows receiving 20 and 30% sunflower seeds. Researchers (72) concluded that this

Figure 4. Protein percent of milk from cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.


5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
Percent
CSM
SCS
SCL
3
6
9
12
15
18
Time in weeks

indicates that synthesis of nonfat components, presumably lactose, by the mammary gland was altered by the diet.

Fatty acid composition of milk fat was markedly different between cows fed sunflower seed rations and the CSM ration (Table 4), however, similar for cows fed SCS and SCL diets. Milk fat from sunflower seed rations had lower proportions of myristic, palmitic ($P<.05$), and palmitoleic ($P<.01$). Depression of shorter chain fatty acids resulting from feeding of sunflower seed supplement agreed with others (58, 72). Other researchers (54, 87) found decreased mammary synthesis of short chain fatty acids when protected tallow or full fat soy flour were fed. These workers suggested that decreased de novo synthesis of short chain fatty acis (C_6 to C_{16}) was probably from feedback inhibition of acetyl CoA carboxylase by increased concentrations of long chain acyl CoA in the mammary gland. There was an increase in proportions of long chain fatty acids ($P<.01$) when fed sunflower seeds. This was in accordance with other studies (58, 72) and was probably due to the result of increased consumption of long chain fatty acids in sunflowers and reduced de novo fatty acid synthesis by the mammary gland in the presence of high dietary fatty acids (58).

Feed Intake and Body Weight Changes

Dry matter intakes (Table 5 and Figure 5) were similar ($P>.05$) for cows fed the three diets, although cows fed the sunflower seed rations consumed slightly less dry matter than cows fed the control, CSM ration. There was a similar trend for the dry matter intake as

TABLE 4. Fatty acid composition of milk fat for cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

Fatty Acid	Diets CSM	 SCS	 SCL	SE[e]
	(wt %)			
Capric (10:0)	2.75	1.81	2.46	.28
Lauric (12:0)	2.80	1.94	2.38	.24
Myristic (14:0)	10.57[c]	8.07[d]	9.29[c,d]	.52
Palmitic (16:0)	31.55[a]	26.33[b]	25.94[b]	1.40
Palmitoleic (16:1)	3.27[c]	2.02[d]	2.21[d]	.21
Stearic (18:0)	13.20[c]	16.93[d]	15.71[d]	.80
Oleic (18:1)	34.29	39.07	37.31	1.59
Linoleic (18:2)	2.52[b]	4.26[a]	3.27[a,b]	.41
Linolenic (18:3)	Trace	Trace	Trace	---

[a,b] Means with different superscripts are different ($P<.05$).

[c,d] Means with different superscripts are different ($P<.01$).

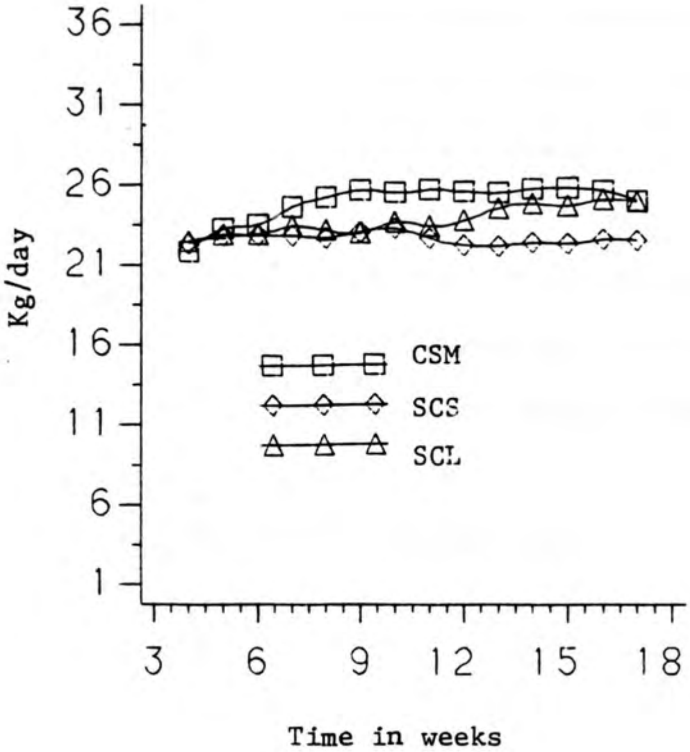
[e] Standard error of the mean.

TABLE 5. Feed intakes and body weight changes for cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

Parameter	Total Ration[a]		
	CSM	SCS	SCL
Dry matter intake, kg/day	21.0	18.4	20.0
Dry matter intake, % of body wt	3.6	3.2	3.4
	—— (wt change, kg) ——		
Weeks 4 - 7	-20.75	-11.82	-19.01
Weeks 8 - 11	12.84	1.40	8.15
Weeks 12 - 17	4.80	3.08	14.37
Average daily wt changes	-.10	-.02	+.09

[a] Contained (DM basis) 47% corn silage, 9.2% alfalfa hay, and 43.8% of respective concentrate mix (Table 1).

Figure 5. Dry matter intake for cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.


36
31
26
21
16
11
6
1
Kg/day
3
6
9
12
15
18
Time in weeks
CSM
SCS
SCL

percent of body weight (Table 3). Dry matter intake was in accordance with other studies (35, 48, 52, 72) in that it was not affected by fat supplementation. Gross energy intake was similar for all three diets, 85.8, 88.5, and 87.6 mcal/day, respectively. Milk yield per kg dry matter intake was slightly higher for the sunflower seed fed diets (1.53, 1.73, and 1.64 kg milk/kg DM intake, respectively).

Average weight changes appear in Table 5. The trial was divided into three periods: period 1 - weeks 4 to 7 postpartum; period 2 - weeks 8 to 11; and period 3 - weeks 12 to 17. During the first period, all cows lost weight, while all cows were gaining weight in the second and third periods. There were no significant differences among treatments; however, average daily weight change was the highest on the SCL ration (90 g/day). Smith et al. (79) found no significant difference in body weight change when cows were fed supplemented fat.

Rumen Fluid Composition

Rumen pH, NH_3, VFA, and serum cholesterol values appear in Table 6. There was no significant difference in rumen pH among treatment groups. In cows fed sunflower seeds, there was slightly higher rumen NH_3 and ruminal concentrations of VFA were similar for all treatments.

Concentrations of serum cholesterol (Table 6) were increased ($P< .05$) when cows were fed sunflower seeds. This response was apparently due to the increased demand for intestinal cholesterol for absorption and transport of fat (61). Serum cholesterol was slightly

TABLE 6. Rumen pH, NH_3, serum cholesterol, and volatile fatty acid composition for cows fed soybean meal (CSM) or sunflower seed rations without (SCS) or with (SCL) additional limestone.

	Diet			
Component	CSM	SCS	SCL	SE[c]
Rumen pH	6.4	6.6	6.5	.05
Rumen NH_3 (mg/100·ml)	13.1	16.0	17.2	1.30
Serum cholesterol (mg/100 ml)	166.0[a]	223.0[b]	255.0[b]	14.00
	(mole %)			
Acid				
Acetate	49.4	49.3	49.4	.67
Propionate	26.2	26.2	25.4	.69
Isobutyrate	1.17	1.16	1.00	.99
Butyrate	17.7	17.7	18.9	.44
Isovalerate	3.0	3.3	2.8	.15
Valerate	2.6	2.5	2.4	.11
Total, μm/ml	83.0	77.0	79.9	3.63

[a,b] Means with different superscripts are different ($P<.05$).

[c] Standard error of the mean.

higher for the SCL ration (P>.05) than for the SCS ration, possibly indicating that the calcium formed insoluble soaps with the fat which then bypassed the rumen into the lower gut for absorption.

SUMMARY

Results indicate that milk production and fat percent were not adversely affected when sunflower seeds plus additional calcium were fed, but fat percent was reduced when cows were fed sunflower seeds without additional calcium. Milk from cows fed diets containing the sunflower seeds contained more unsaturated fatty acids, more stearic acid, and more linoleic acid than control fed animals. Milk protein percent was similar for cows fed the control and sunflower seeds without added calcium, but slightly lower when fed the sunflower seeds with added calcium. Serum cholesterol concentrations were higher for cows fed diets containing sunflower seeds.

It appears that formation of insoluble soaps by the addition of calcium may be an effective and simple method of increasing ruminal bypass of fat. This may allow the fat to be better used as an energy source by early lactation cows, and prevent any depression in milk fat percentages that may occur without the additional dietary calcium.

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APPENDIX

APPENDIX TABLE 1. Sum of squares for milk production (kg)[a].

Source of variation	Degrees of freedom	Milk production	Fat corrected milk
Total	419	586100.2	696098.9
Treatment	2	3446.9	18829.0
Wk	13	61857.5	157399.0
Trt x wk	26	19265.0	16139.3
Error	377	309713.7	359059.8

[a] Covariate adjusted.

APPENDIX TABLE 2. Sum of squares for milk composition (%)[a].

Source of variation	Degrees of freedom	Components			
		Fat %	Protein %	Solids %	SNF
Total	209	89.40	17.51	120.58	58.03
Treatment	2	3.70	.326	4.26	1.32
Wk	6	24.61	1.15	21.20	1.78
Trt x wk	12	4.36	.582	4.44	2.02
Error	188	39.82	7.92	54.92	38.03

[a] Covariate adjusted.

APPENDIX TABLE 3. Sum of squares for fatty acid composition of milk fat (%).

Source of variation	Degrees of freedom	Fatty acids								
		C10:0	C12:0	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3
Total	29	26.41	19.63	105.72	727.34	21.43	244.99	807.49	60.85	0
Treatment	2	4.62	3.40	31.28	196.06	9.13	72.19	116.90	15.23	0
Error	27	21.79	16.22	74.44	531.27	12.29	172.80	690.59	45.62	0

APPENDIX TABLE 4. Sum of squares for serum cholesterol (mg/100 ml).

Source of variation	Degrees of freedom	Cholesterol
Total	71	445333.8
Treatment	2	97488.0
Period	2	49593.7
Trt x Per	4	14965.7
Error	63	283286.3

APPENDIX TABLE 5. Sum of squares of rumen NH_3 (mg/100 ml) and volatile fatty acid composition (molar %).

Source of variation	Degree of freedom	RNA_3	Fatty acids					
			C_2	C_3	IC_4	C_4	IC_5	C_5
Total	89	4762.04	1206.57	1415.79	2532.58	533.19	65.55	35.65
Treatment	2	264.68	.241	13.28	55.07	24.79	2.87	.326
Period	2	186.62	15.37	33.77	18.99	9.07	3.44	.433
Trt x Per	4	185.50	80.15	185.04	44.62	18.36	2.80	.679
Error	81	4125.23	1110.80	1183.69	2413.89	480.95	56.43	34.21

APPENDIX TABLE 6. Sum of squares for volatile fatty acid composition (µm/ml).

Source of varition	Degrees of freedom	Fatty acids						Total
		C_2	C_3	IC_4	C_4	IC_5	C_5	
Total	89	8529.90	4254.40	1650.70	1972.99	51.35	55.57	36056.61
Treatment	2	89.65	55.31	43.36	4.47	1.84	1.16	540.86
Period	2	400.99	115.24	24.22	19.69	1.98	.315	1762.70
Trt x Per	4	233.98	336.19	46.01	71.97	.458	2.16	1703.49
Error	81	7805.26	3747.66	1537.10	1876.83	47.06	51.92	32049.54