Performance of Lactating Dairy Cows Fed Whole Cottonseed with Elevated Concentrations of Free Fatty Acids in the Oil

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ABSTRACT

Twenty-four multiparous cows were used in a 10-wk randomized block design trial to evaluate the effects of feeding whole cottonseed (WCS) containing increasing concentrations of free fatty acids (FFA) in the oil on nutrient intake and digestibility, milk yield and composition, and select plasma metabolites. Two lots of WCS containing either 3 or 12% FFA were blended to provide WCS with 3, 6, 9, and 12% FFA. Cottonseeds were included in the wheat silage-based total mixed ration at 12.5% of dry matter (DM). There was no difference in intakes of DM, crude protein (CP), or neutral detergent fiber; yield of milk; or percentages of milk protein, lactose, or SNF. Milk fat percentage was lowest for the diet containing WCS with 6% FFA. Concentrations of individual milk fatty acids C6:0 decreased and C16:1 increased linearly as FFA in WCS increased. A cubic response was observed for concentrations of C8:0, C10:0, and C12:0 because of higher concentrations when diets contained WCS with 6% FFA than 3 and 12% FFA, which were higher than 9% FFA. Intake and apparent total tract digestibility of acid detergent fiber increased linearly as FFA concentration in WCS increased. Apparent NDF digestibility was highest for diets containing WCS with 3 and 6% FFA; CP digestibility was highest with WCS containing 3 and 9% FFA. Differences in milk fatty acid concentration and nutrient digestibility suggest minor changes in rumen fermentation; however, feeding WCS with up to 12% FFA did not negatively impact nutrient intake and digestibility or milk yield or composition.

(Key words: whole cottonseed, free fatty acids, milk yield, nutrient digestibility)

Abbreviation key: EE = ether extract, FA = fatty acids, WCS = whole cottonseed.

INTRODUCTION

Whole cottonseed (WCS) is commonly included in the diet of lactating dairy cattle as a source of fat, protein, and fiber. The supplemental fat from WCS increases the energy density of the diet in support of higher milk yield and provides a source of effective fiber. High levels of unsaturated fatty acids (FA) in the diet are toxic to certain rumen microbes and can coat feed particles, preventing fibrolytic microbes from attaching to fiber and depressing fiber digestion (MacLeod and Buchanan-Smith, 1972; Eastridge and Firkins, 1991). In contrast to other fat sources, WCS can be fed at rates up to 15% of the diet without drastically disturbing fiber digestion because the FA contained in the seed are released slowly in the rumen and biohydrogenated; therefore, they are not toxic (Moore et al., 1986).

According to rules published by the National Cottonseed Products Association (NCPA, 1997), WCS used for oil production cannot contain more than 1.8% FFA in the oil. Typically, WCS that is considered undesirable for oil production is often sold as livestock feed. Previous research suggested that the concentration of unsaturated FFA may be responsible for disruption of normal rumen fermentation (Jenkins, 1993). Because the oil in WCS is 70% unsaturated and is completely hydrolyzed to FA in the rumen (Keele et al., 1989), high levels of FFA in WCS may have a negative effect on fiber digestion (Martinez et al., 1991; DePeters and Cant, 1992).

The ability of lipid sources to alter milk FA profiles is well documented (Hermansen, 1995). Various lipid sources, especially those containing polyunsaturated fat, have been implicated in spontaneous oxidation and off-flavors in milk (Timmons et al., 2001). Because of the high level of polyunsaturated FA in WCS and the possibility of reduced biohydrogenation of FFA in the rumen, WCS containing elevated concentrations of FFA could be detrimental to milk quality. The objectives of this study were to determine the effects of feeding WCS with elevated concentrations of FFA in the oil on nutrient intake and digestibility, milk yield and composition, and selected blood metabolites of lactating dairy cows.

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Table 1. Ingredient and chemical composition of experimental diets containing whole cottonseed (WCS) with increasing concentrations of free fatty acids (FFA) in the oil.

	FFA in oil of WCS			
	3%	6%	9%	12%
Ingredient	(% of DM)			
Wheat silage	44.9	44.9	44.9	44.9
Ground corn	24.5	24.5	24.5	24.5
3% FFA WCS	12.5	8.3	4.2	
12% FFA WCS		4.2	8.3	12.5
Soybean hulls	3.0	3.0	3.0	3.0
Soybean hulls (48% CP)	9.2	9.2	9.2	9.2
Protein supplement ¹	3.8	3.8	3.8	3.8
Mineral-vitamin premix ²	2.1	2.1	2.1	2.1
Chemical	(%)			
DM	57.5	57.4	57.8	57.8
	(% of DM)			
CP	19.8	18.8	19.6	19.0
NDF	46.5	47.2	46.8	47.8
ADF	29.8	29.1	28.4	29.6
EE ³	6.3	6.3	6.4	6.4
NE _L , ⁴ Mcal/kg	1.6	1.6	1.6	1.6

¹Protein supplement was composed of 60% Menhaden fish meal and 40% distillers grains with solubles.

²Premix contained 34% CP from urea, 24.50% Ca, 3.68% P, 1.27% Mg, 0.08% K, 3.03% Na, 4.60% Cl, 0.31% S, 11.67 ppm of Co, 665 ppm of Cu, 4622 ppm of Fe, 58 ppm of I, 2039 ppm of Mn, 14.69 ppm of Se, 1943 ppm of Zn, 60,360 IU/kg of vitamin A, 24,145 IU/kg vitamin D, and 300 IU/kg of vitamin E.

³EE = ether extract.

⁴Determined using NRC (2001) values.

MATERIALS AND METHODS

Two lots of WCS differing in FFA concentrations were obtained from warehouses in South Georgia and transported to The University of Georgia Dairy Center in Athens, GA. One lot of WCS contained 3% FFA, and the second lot contained 12% FFA as determined by the procedure outlined by NCPA (1997). Experimental diets, fed as TMR (Table 1), were formulated for a 670-kg lactating cow producing 32 kg/d of milk (3.5% milk fat and 3.3% protein) with a daily gain of 0.45 kg. Dietary treatments consisted of 4 concentrations of FFA in WCS and were achieved by stepwise substitution of the low FFA WCS with high FFA WCS.

Twenty-four multiparous lactating Holstein cows were used in a 10-wk randomized block design trial from March 28 through June 6, 2001. Cows were managed according to procedures approved by The University of Georgia Institutional Animal Care and Use Committee. Cows were fed the control diet for 14 d prior to the start of the 56-d trial period. Cows were housed in a free stall barn and individually fed twice daily behind Calan gates (American Calan, Inc., Northwood, NH) at 105% of the previous day's intake. Cows were assigned to 1 of 6 blocks according to milk yield ($\bar{x}$ = 37.8; range

32.7 to 49.9 kg/d). Cows within blocks were assigned randomly to 1 of 4 dietary treatments at the end of the 14-d preliminary period. The amount of feed offered and orts were recorded daily. Feed and orts were sampled daily and composited weekly by treatment. No significant sorting was observed among cows based on visual analysis of orts. Pooled samples were dried at 55°C for 72 h, ground to pass through a 2-mm screen using a Wiley mill screen (Arthur W. Thomas Co., Philadelphia, PA), and stored for analyses.

Milk yield was recorded at each milking (2 times daily). Milk samples were collected at each milking during the pretrial and at wk 2, 3, 4, 5, and 6 and pooled by cow within each week for analyses of milk fat and protein using infrared spectrometry (DHIA, Belleview, FL). Total solids were determined by lyophilizing duplicate 10-mL aliquots (Virtis Freezemobile 12; Virtis Co Inc., Gardiner, NY). The solids were then ashed for 3 h at 510°C to determine the mineral content. Lactose percentage was calculated by difference. During wk 5 and 7, milk fat was isolated from an aliquot by centrifugation at 21,000 × g for 15 min. Milk FA were converted to methyl esters, and FA profile was determined by the method of Kramer et al. (1997) using Hewlett Packard 5890A equipped with a 30- × 0.25-mm (i.d.) polyamino glycol capillary column (Spelco Inc., Bellefonte, PA). Cows were weighed on 3 consecutive d during the last week of the pretrial period and the last week of experimental period.

Fecal grab samples were collected on 3 consecutive d during wk 5 at 12-h intervals. Sampling schedule was advanced 2 h each day during collection period. Samples were dried, composited by cow, ground to pass through a 2-mm screen, and stored for analyses.

Nitrogen was determined using a Leco FP528 (Leco Corp., St. Joseph, MI), and CP was calculated (percentage of N × 6.25). Ether extract (EE) of feed and orts was determined by soxhlet extraction with petroleum ether. The NDF and ADF of the TMR, orts, and feces were determined by the method of Robertson and Van Soest (1981) using an Ankom 220 Fiber Analyzer (Ankom Technology Co., Fairport, NY). Nutrient digestibility was calculated using indigestible ADF as a marker (Henderson et al., 1985) using a Daisy II 200 Rumen Fermentor (Ankom Technology Co.). Nutrient intake and apparent digestibility coefficients were corrected for orts.

Blood samples were collected from the coccygeal vein in preheparinized and NaF tubes at 0, 2, and 4 h after the a.m. feeding during wk 2 of the pretrial period and wk 3 and 6 of the experimental period. Samples were prepared for glucose and insulin analyses by refrigeration (8 h at 4°C) and centrifugation (3000 × g, 20 min). Plasma glucose was analyzed by a Tinder colorimetric

method (procedure #315; Sigma, St. Louis, MO). Insulin was determined by radioimmunoassay using a ^{125}I competitive binding method (ICN Pharmaceuticals Inc., Costa Mesa, CA). Plasma samples from wk 6 were pooled and prepared for AA analysis by deproteinizing with 5% sulfasalicylic acid, freezing for 12 h at -4°C , and centrifuged at $5000 \times g$ for 20 min (Amos et al., 1976). Plasma AA were determined by HPLC using a Beckman 1600 amino acid analyzer using norleucine as an internal standard (Beckman Instruments Inc., Fullerton, CA).

The DMI, milk yield, and milk composition data were analyzed using the PROC MIXED procedures of SAS (1996). The statistical model included the effect of treatment, week, and treatment $\times$ week interaction. Corresponding data from the preliminary week were included as a covariate for each variable, except milk FA concentration, along with DIM. Cow within treatment was included in the model as a random effect. A compound symmetry covariance structure was specified. Nutrient intake, apparent digestibility, and plasma AA data were analyzed using the GLM procedure of SAS (1996). Linear, quadratic, and cubic effects of FFA concentration were tested using one degree of freedom using contrast statements.

RESULTS AND DISCUSSION

The experimental diets contained similar concentrations of nutrients (Table 1). The two lots of WCS (Table 2) were similar in composition except that the 12% FFA had slightly higher concentrations of C16:0 and slightly lower concentrations of EE and C18:2.

Average DMI was similar for all treatments during the 8-wk trial (Table 3); however, there was a treatment $\times$ week interaction ($P < 0.05$). The interaction was due to an increase in DMI for cows fed WCS containing 6, 9, and 12% FFA during wk 7 and 8; whereas DMI remained constant for cows fed the diet containing WCS with 3% FFA (Figure 1). These results indicate that increasing concentrations of FFA in WCS do not negatively affect the acceptability of the diet.

Milk yield was similar for all treatments and averaged 33.5 kg/d. Milk fat percentage was higher ($P < 0.05$) for cows fed the 9% FFA WCS and lowest for cows fed the 6% FFA WCS compared with the other treatments. Yield of milk fat was similar for all treatments, so the higher milk fat percentage observed with 9% FFA WCS was most likely due to dilution as milk yield was numerically lower. No differences were observed among treatments in percentage or yield of milk protein, lactose, ash, total solids, and SNF or yield of energy-corrected milk.

Table 2. Chemical composition of whole cottonseed (WCS) containing different concentrations of FFA in the oil.

	FFA in oil of WCS	
	3%	12%
	(% of DM)	
DM	88.8	88.0
CP	26.7	24.8
NDF	43.2	43.3
ADF	30.3	30.8
EE ¹	17.0	16.2
	(% of oil)	
FFA	3.0	12.0
	(% of fatty acids)	
C14:0	0.70	0.72
C16:0	22.31	24.76
C16:1	0.52	0.57
C18:0	2.23	2.45
C18:1	14.97	14.91
C18:2	58.53	56.22
C18:3	0.15	0.14
C20:0	0.26	0.00
C22:0	0.16	0.00
C24:0	0.10	0.16
Other	0.07	0.07

¹EE = ether extract.

The higher standard error observed for milk yield and energy-corrected milk yield coupled with only 6 cows per treatment increases the difference required to detect a significant difference between treatments. Milk yield was numerically greatest for the diet containing WCS with 6% FFA and lowest for the diet containing WCS with 9% FFA, but energy-corrected milk yield was numerically higher for diets containing WCS greater than 3% FFA. These data suggest that feeding WCS with up to 12% FFA should not negatively impact milk yield; however, additional research is required to determine the impact of feeding WCS with even higher concentrations of FFA.

Table 3. Dry matter intake and milk yield and composition of lactating Holstein cows fed diets containing whole cottonseed (WCS) with increasing concentrations of FFA in the oil.

	FFA in oil of WCS				SE
	3%	6%	9%	12%	
DMI, kg/d	21.5	22.8	22.1	23.0	0.7
Milk, kg/d	32.9	35.7	32.0	33.8	1.9
Fat, ^a %	3.48	3.32	3.67	3.41	0.08
Protein, %	2.63	2.74	2.85	2.81	0.09
Lactose, %	5.39	5.30	5.37	5.22	0.15
TS, %	12.22	12.01	12.61	12.18	0.25
Ash, %	0.69	0.71	0.71	0.69	0.01
SNF, %	8.72	8.74	0.92	0.74	0.18
ECM, ¹ kg/d	32.7	33.2	33.5	33.1	1.6

¹ECM = energy-corrected milk.

^aCubic response to increasing concentrations of FFA in WCS ($P < 0.05$).

Figure 1. Interaction of FFA concentration in the oil of whole cottonseed and week on DMI (SE = 1.0 kg/d, $P < 0.03$).

Increased milk fat percentage and decreased milk protein percentage have been reported when WCS are supplemented to the diet (Coppock et al., 1987), but composition is not typically altered by type of cottonseed (Santos et al., 2002) or amount fed (Coppock et al., 1987). The change in BW during the 8-wk trial did not differ by treatment (−11.4, 18.7, 6.0, and 10.8 kg for WCS with FFA of 3, 6, 9, and 12%, respectively). In contrast to our results, Bruckmaier et al. (1998) reported a decrease in milk fat percentage when a prilled FFA supplement was fed. Compared with the hydrogenated triglyceride supplement, the prilled FFA supplement those researchers fed had much higher concentrations of C16:0 and lower concentrations of C18:1, which has reduced milk fat percentage (Palmquist and Jenkins, 1980).

Individual milk FA concentrations are presented in Table 4. Concentrations of C6:0 decreased linearly ($P < 0.05$) as FFA in WCS increased. A cubic response was observed for C8:0 ($P < 0.05$), C10:0 ($P < 0.02$), C12:0 ($P < 0.01$), and total short-chain FA because concentrations were highest for 6% FFA and lowest for 9% FFA. Concentrations of C16:0 ($P < 0.05$), C16:1 ($P < 0.05$), and total medium-chain FA ($P < 0.01$) increased linearly in response to WCS with increasing FFA. Individual and

total long-chain FA, total unsaturated FA, and total saturated FA were similar for all treatments.

Supplemental WCS has reduced concentrations of C6 through C14 milk FA because of the inhibition of mammary de novo FA synthesis (Palmquist et al., 1993; Wu et al., 1994; Jenkins et al., 1996). The increase in FFA did not appear to alter synthesis of short-chain FA greatly, although differences were noted for diets containing WCS with 6 and 9% FFA. Increasing FFA in WCS does not appear to alter medium- or long-chain FA. Generally off-flavors caused by spontaneous auto-oxidation are associated with increases in milk C18:2 and C18:3 FA, and these FA are often increased with addition of oilseed to the diet (Hermansen, 1995). Concentrations of C18:2 and C18:3 were similar for all diets in the current study, suggesting that increased FFA in WCS should not cause any off-flavors in the milk.

Nutrient intake and total tract apparent digestibility coefficients as measured during wk 5 are presented in Table 5. No differences were observed in intake of DM, CP, and NDF; however, intake of ADF increased linearly ($P < 0.02$) as FFA in WCS increased. No differences were observed in the apparent digestibility of DM among treatments. The CP digestibility was lowest for WCS with 6% FFA and highest for WCS with 9% FFA,

Table 4. Fatty acid (FA) concentrations in milk of lactating Holstein cows fed diets containing whole cottonseed (WCS) with increasing concentrations of FFA in the oil.

	FFA in oil of WCS				SE
	3%	6%	9%	12%	
	(% of total FA)				
C4:0	3.62	3.61	3.28	3.40	0.15
C6:0 ^a	2.38	2.55	2.25	2.27	0.07
C8:0 ^b	1.30	1.49	1.25	1.26	0.05
C10:0 ^c	2.56	3.06	2.50	2.60	0.10
C11:0	0.04	<0.01	0.04	0.04	0.02
C12:0 ^c	2.70	3.23	2.69	2.79	0.10
Total C4-12 FA ^b	12.60	13.94	12.01	12.36	0.22
C14:0	10.13	10.81	10.31	10.36	0.21
C14:1	1.22	1.30	1.29	1.22	0.08
C15:0	0.90	0.90	0.89	0.90	0.02
C16:0 ^a	28.74	27.94	30.01	30.33	0.74
C16:1 ^a	0.94	0.92	1.20	1.18	0.09
C17:0	0.52	0.52	0.51	0.51	0.02
Total medium-chain FA ^{a,b}	42.45	42.39	44.21	44.50	0.60
C18:0	14.97	14.37	14.27	14.43	0.59
t18:1	2.01	2.13	2.03	2.17	0.08
C18:1	20.70	19.60	20.30	19.40	0.65
C18:2	2.09	2.26	2.11	2.07	0.08
C18:2 c9t11	0.36	0.37	0.38	0.39	0.06
C18:3	0.37	0.38	0.36	0.36	0.05
Other CLA	0.12	0.13	0.12	0.14	0.03
C20:0	0.24	0.21	0.24	0.23	0.01
C22:0	0.08	0.07	0.08	0.06	0.02
C22:1	0.08	0.05	0.05	0.04	0.02
Total long-chain FA ²	41.02	39.57	39.94	39.29	0.60
Unidentified FA	3.93	4.10	3.84	3.85	
Unsaturated FA	27.88	27.15	27.85	26.93	0.51
Saturated FA	68.26	68.71	68.32	69.23	0.58

^aLinear response to increasing concentrations of FFA in WCS ($P < 0.05$).^bCubic response to increasing concentrations of FFA in WCS ($P < 0.05$).^cCubic response to increasing concentrations of FFA in WCS ($P < 0.02$).^dLinear response to increasing concentrations of FFA in WCS ($P < 0.01$).¹Fatty acids with 14 to 18 carbons.²Fatty acids greater than 18 carbons.**Table 5.** Nutrient intake and apparent total tract digestibility of lactating Holstein cows fed diets containing whole cottonseed (WCS) with increasing concentrations of FFA in the oil during wk 5.

	FFA in oil of WCS				SE
	3%	6%	9%	12%	
Intake, kg/d					
DM	21.2	23.0	21.8	21.9	0.9
CP	3.4	3.4	3.6	3.3	0.1
NDF	6.01	6.67	6.60	7.16	0.6
ADF ^a	10.4	11.6	10.2	10.7	0.4
Apparent digestibility, %					
DM	50.3	51.2	49.7	49.6	1.0
CP ^c	54.1	48.0	53.2	46.7	1.1
NDF ^c	39.7	42.8	35.4	38.6	0.7
ADF ^b	35.1	36.7	39.9	45.6	1.6

^aLinear response to increasing concentrations of FFA in WCS ($P < 0.05$).^bLinear response to increasing concentrations of FFA in WCS ($P < 0.01$).^cCubic response to increasing concentrations of FFA in WCS ($P < 0.05$).

resulting in a cubic response ($P < 0.05$). The NDF digestibility was highest for WCS with 6% FFA and lowest for WCS with 9% FFA, resulting in a cubic response ($P < 0.05$). Apparent digestibility of ADF increased linearly ($P < 0.01$) as FFA in WCS increased.

Plascencia et al. (1999) did not observe any negative effect on the total tract digestibility of nutrients when yellow grease containing increasing concentrations of FFA were fed to steers. However, the increased ADF intake with increasing FFA levels in our trial might have minimized any negative effects at the highest levels of FFA in WCS. Others have shown that increasing the fiber content of a diet can mitigate the fermentation problems normally associated with increased unprotected dietary fats (Mir, 1988; Doreau et al., 1991; Tackett et al., 1996). The reason for the differences in CP digestibility is not apparent, but might reflect nonsignificant differences in intake that parallel the digestibility coefficients.

Table 6. Amino acid concentrations of diets containing whole cottonseed (WCS) with increasing concentrations of FFA in the oil.

	FFA in oil of WCS			
	3%	6%	9%	12%
	(mg AA/g of DM)			
Arginine	9.7	9.9	9.5	8.6
Histidine	3.5	3.5	3.5	3.3
Isoleucine	6.6	6.7	6.4	6.3
Leucine	11.4	11.4	11.4	10.7
Lysine	6.0	6.0	5.8	5.6
Methionine	2.3	2.3	2.2	2.0
Phenylalanine	6.6	6.7	6.5	6.2
Threonine	5.6	5.7	5.7	5.3
Valine	0.4	0.4	0.5	0.4
Total essential AA	52.1	52.6	51.5	48.4
Alanine	9.5	9.6	9.5	8.9
Aspartamine	12.8	13.0	12.8	12.4
Cystine	3.8	3.8	3.7	3.6
Glutamine	26.2	26.3	26.0	24.9
Glycine	7.4	7.6	7.5	7.4
Proline	10.8	10.8	10.8	10.2
Serine	7.5	7.5	7.7	7.1
Tyrosine	4.7	4.6	4.7	4.3
Total non-essential AA	82.7	83.2	82.7	78.8
Total AA	134.8	135.8	134.2	127.2

There were no differences among treatments in average plasma glucose concentrations, which averaged 54.5 mg/dL (data not shown). Plasma insulin concentrations were 18.0, 20.5, 19.7, and 17.4 μ U/mL for WCS with FFA of 3, 6, 9, and 12%, respectively. A quadratic effect ($P < 0.005$) was observed for plasma insulin because of higher concentrations when WCS containing either 6 or 9% FFA were fed compared with that when WCS containing either 3 or 12% FFA were fed. An interaction ($P < 0.001$) of week $\times$ sampling time was observed for both glucose and insulin concentrations because of a shift in eating patterns. As the trial progressed, temperatures increased, and the cows ate a greater proportion of their DM after the p.m. feeding.

Dietary concentrations of AA are presented in Table 6, and plasma AA concentrations measured during wk 6 are presented in Table 7. A quadratic response was observed for concentrations of isoleucine ($P < 0.01$), leucine ($P < 0.03$), valine ($P < 0.01$), and total branch ed-chain AA ($P < 0.01$) because of lower concentrations when WCS with 6 and 9% FFA were fed. Plasma lysine exhibited a cubic response ($P < 0.02$) because of higher concentrations with 3 and 9% FFA in WCS than with 6 and 12% FFA in WCS. Phenylalanine and ornithine

Table 7. Plasma amino acid concentrations of lactating Holstein cows fed diets containing whole cottonseed (WCS) with increasing concentrations of FFA in the oil.

	FFA in oil of WCS				SE
	3%	6%	9%	12%	
	(mg AA/dL)				
Arginine	8.64	7.26	7.34	7.15	0.50
Histidine	3.93	3.14	3.50	3.54	0.35
Isoleucine ^b	23.79	17.58	17.36	19.26	1.23
Leucine ^c	15.18	11.22	11.28	11.86	0.94
Lysine ^d	8.78	6.79	7.40	5.82	0.63
Methionine	4.12	3.84	3.84	3.97	0.19
Phenylalanine ^a	3.91	3.35	3.37	3.01	0.18
Threonine	7.87	6.70	8.17	7.72	0.53
Tryptophan	2.97	7.12	7.38	5.48	2.60
Valine ^b	25.84	18.44	17.44	20.45	1.49
Total essential AA	105.03	85.43	87.09	87.50	5.15
Alanine	20.09	19.30	18.34	17.88	1.85
Cysteine	4.86	5.05	4.85	5.06	0.17
Cystithionine ^a	0.19	0.21	0.20	0.29	0.01
Glutamine	149.38	146.58	144.38	122.25	8.56
Glutamate	5.62	5.87	5.90	5.72	0.48
Glycine	30.01	33.27	27.66	29.64	1.70
Ornithine ^d	4.77	3.81	4.41	3.89	0.26
Proline	6.80	6.00	5.91	6.33	0.48
Serine	6.54	6.92	6.77	6.26	0.39
Tyrosine	3.50	5.27	2.82	2.94	0.91
Branched-chain AA ^b	64.81	47.24	46.09	51.56	3.48
Total essential AA ^a	230.01	230.48	220.94	198.73	7.92
Total AA ^a	335.04	315.92	308.03	286.23	10.47

^aLinear response to increasing concentrations of FFA in WCS ($P < 0.01$).

^bQuadratic response to increasing concentrations of FFA in WCS ($P < 0.01$).

^cQuadratic response to increasing concentrations of FFA in WCS ($P < 0.05$).

^dCubic response to increasing concentrations of FFA in WCS ($P < 0.01$).

concentrations decreased linearly ($P < 0.02$), whereas cystithionine increased linearly ($P < 0.01$), as FFA in WCS increased. Total essential AA ($P < 0.07$), nonessential AA ($P < 0.01$), and total AA ($P < 0.01$) decreased linearly as the concentration of FFA in WCS increased.

Supplemental free soybean oil and whole soybeans have been observed to reduce plasma concentrations of isoleucine, ornithine, and leucine; however, no changes in plasma AA were noted when free oil and WCS were supplemented (Mohamed et al., 1988). Decreased concentrations of plasma branched-chain AA may indicate reduced microbial protein synthesis (Mohamed et al., 1988), which could account for the linear decline in concentrations of total essential and nonessential AA as the FFA in WCS increased. However, the flow of microbial protein to the abomasum was not altered when steers were fed WCS with FFA concentrations of 8 to 14.7% (Sullivan, 2002). Digestibility of CP was lowest when WCS with either 6 or 12% FFA were fed, but this did not alter yield of milk or milk protein.

CONCLUSIONS

Feeding WCS with FFA concentrations up to 12% does not alter nutrient intake, milk yield, or composition. Minor changes were noted for several short-chain FA in the milk, but, overall, the FA profile was not greatly altered. There was no change in concentrations of C18:2 and C18:3 in milk, suggesting that increased FFA in WCS would not cause any off-flavors in the milk or milk products. Changes in nutrient digestibility and plasma AA concentrations suggest that changes in ruminal fermentation might have occurred, which warrants additional research.

REFERENCES

- Amos, H. E., J. J. Evans, and D. Burdick. 1976. Abomasal protein recovery and microbial protein synthesis in wethers fed high and low quality forage diets. *J. Anim. Sci.* 42:970–976.
- Bruckmaier, R. M., L. Gregoret, F. Jans, D. Faissler, and J. W. Blum. 1998. Longissimus dorsi muscle diameter, backfat thickness, body condition scores and skinfold values related to metabolic and endocrine traits in lactating dairy cows fed crystalline fat or free fatty acids. *J. Vet. Med.* 45:397–410.
- Coppock, C. E., J. K. Lanham, and J. I. Horner. 1987. A review of the nutritive value and utilization of whole cottonseed, cottonseed meal and associated by-products by dairy cattle. *Anim. Feed Sci. Technol.* 18:89–129.
- DePeters, E. J., and J. P. Cant. 1992. Nutritional factors influencing the nitrogen composition of bovine milk: A review. *J. Dairy Sci.* 75:2043–2070.
- Doreau, M., F. Legay, and D. Bauchart. 1991. Effect of source and level of supplemental fat on total and ruminal organic matter and nitrogen digestion in dairy cows. *J. Dairy Sci.* 74:2233–2242.
- Eastridge, M. L., and J. L. Firkins. 1991. Feeding hydrogenated fatty acids and triglycerides to lactating dairy cows. *J. Dairy Sci.* 74:2610–2616.
- Henderson, S. J., H. E. Amos, and J. J. Evans. 1985. Influence of dietary protein concentration and degradability on milk production, composition, and ruminal protein metabolism. *J. Dairy Sci.* 68:2227–2237.
- Hermansen, J. E. 1995. Prediction of milk fatty acid profile in dairy cows fed dietary fat differing in fatty acid composition. *J. Dairy Sci.* 78:872–879.
- Jenkins, T. C. 1993. Lipid metabolism in the rumen. *J. Dairy Sci.* 76:3851–3863.
- Jenkins, T. C., H. G. Bateman, and S. M. Block. 1996. Butylsoyamide increases unsaturation of fatty acids in plasma and milk of lactating dairy cows. *J. Dairy Sci.* 79:585–590.
- Keele, J. W., R. E. Roffler, and K. Z. Beyers. 1989. Ruminal metabolism in non-lactating cows fed whole cottonseed or extruded soybeans. *J. Anim. Sci.* 67:1612–1622.
- Kramer, J. K., V. Fellner, M. E. Dugan, F. D. Sauer, M. M. Mossoba, and M. P. Yurawecz. 1997. Evaluating acid and base catalysts in the methylation of milk and rumen fatty acids with special emphasis on conjugated dienes and total trans fatty acids. *Lipids* 32:1219–1228.
- MacLeod, G. L., and J. G. Buchanan-Smith. 1972. Digestibility of hydrogenated tallow, saturated fatty acids and soybean-supplemented diets by sheep. *J. Anim. Sci.* 35:890–895.
- Martinez, N., E. J. DePeters, and D. L. Bath. 1991. Supplemental niacin and fat effects on milk composition of lactating Holstein cows. *J. Dairy Sci.* 74:202–220.
- Mir, Z. 1998. A comparison of canola acidulated fatty acids and tallow as supplements to a ground alfalfa diet for sheep. *Can. J. Anim. Sci.* 68:761–768.
- Mohamed, O. E., L. D. Satter, R. R. Grummer, and F. R. Ehle. 1988. Influence of dietary cottonseed and soybean on milk production and composition. *J. Dairy Sci.* 71:2677–2688.
- Moore, J. A., R. S. Swingle, and W. H. Hale. 1986. Effects of whole cottonseed, cottonseed oil, or animal fat on digestibility of wheat straw diets by steers. *J. Anim. Sci.* 63:1267–1273.
- NCPA. 1997. Rules of the National Cottonseed Products Association, Inc. Natl. Cottonseed Products Assoc., Memphis, TN.
- National Research Council. 2001. Nutrient Requirements of Dairy Cattle. 7th rev. ed. Natl. Acad. Sci., Washington, DC.
- 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.
- Palmquist, D. L., and T. C. Jenkins. 1980. Fat in lactation rations: A review. *J. Dairy Sci.* 63:1–14.
- Plascencia, A., M. Estrada, and R. A. Zinn. 1999. Influence of free fatty acid content on the feeding value of yellow grease in finishing diets for feedlot cattle. *J. Anim. Sci.* 77:2603–2609.
- Robertson, J. B., and P. J. Van Soest. 1981. The detergent system of analysis and its application to human food. Page 123 in *The Analysis of Dietary Fiber*. W. P. T. James and O. Theodor, ed. Marcel Dekker, New York, NY.
- Santos, J. E. P., M. Villaseñor, E. J. DePeters, P. H. Robinson, and B. C. Baldwin, Jr. 2002. Type of cottonseed and level of gossypol in diets of lactating dairy cows: Effects on lactation performance and plasma gossypol. *J. Dairy Sci.* 85:1491–1501.
- SAS/STAT Software; Changes and Enhancements Through Release 6.11. 1996. SAS Inst., Inc., Cary, NC.
- Sullivan, H. M. 2002. Effect of high free fatty acid cottonseed on production in Holstein cattle. Ph.D. Diss., Univ. Georgia, Athens.
- Tackett, V. L., J. A. Bertrand, T. C. Jenkins, F. E. Pardue, and L. W. Grimes. 1996. Interaction of dietary fat and acid detergent fiber diets of lactating dairy cows. *J. Dairy Sci.* 79:270–275.
- Timmons, J. S., W. P. Weiss, D. L. Palmquist, and W. J. Harper. 2001. Relationships among dietary roasted soybeans, milk components, and spontaneous oxidized flavor. *J. Dairy Sci.* 84:2440–2449.
- Wu, Z., J. T. Huber, S. C. Chan, J. M. Simas, K. H. Chen, J. G. Vargela, F. Santos, C. Fontes, Jr., and P. Yu. 1994. Effect of source and amount of supplemental fat on lactation and digestion in cows. *J. Dairy Sci.* 77:1644–1651.