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Temporal changes in milk fatty acid distribution due to feeding different levels of rolled safflower seeds to lactating Holstein cows

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ABSTRACT

The objective of this experiment was to follow the time-course changes of the milk fatty acids (FA) and particularly conjugated linoleic acid (CLA), n-3, and n-6 FA in response to feeding whole rolled safflower seed (SS). Eighteen cows were blocked by milk production, days in milk, and parity, and randomly assigned to 1 of 3 diets by replacing whole cottonseed with SS. The control diet contained no SS (SS0), whereas the other diets contained 3% of dry matter as SS (SS3) or 6% SS (SS6). The study was conducted for 8 wk. Cows fed SS produced more milk than SS0, with SS3 producing more milk than SS6, but without a change in milk fat yield or milk fat %. Except for C8:0 FA, changes in milk FA were not observed until the third week of SS feeding. The C8:0 began decreasing during wk 1 of SS feeding and continued to decline to wk 8. Short-chain FA (C6:0 to C11:0) and medium-chain FA (C12:0 to C16:1) concentrations decreased in milk when cows were fed SS, whereas long-chain FA (C18:0 and higher) increased after wk 3. The milk long-chain FA increased from wk 3 until wk 5 and then reached a plateau with little difference between SS3 and SS6, whereas the short-chain FA decreased more in milk from cows fed SS6 than SS3. Total CLA increased slightly less than 5× in milk from cows fed SS compared with SS0. Over the same time frame, n-3 FA declined and n-6 FA increased in the milk from cows fed SS, with no difference between SS3 and SS6. This study indicated that SS fed at 3 and 6% of DM had the potential to increase milk production and the CLA in milk, but with a corresponding increase in n-6 FA.

Key words: safflower seed, conjugated linoleic acid, temporal fatty acid change

INTRODUCTION

Bioactive fatty acids (**FA**) in milk can have substantial benefits for human health and include CLA (C18:2 *cis*-9 *trans*-11 and C18:2 *trans*-10 *cis*-12) and the n-3 FA as well as C18:1 *trans*-11. The CLA are among the most potent anti-carcinogenic compounds (Parodi, 1999). The n-3 FA have been found to aid in the prevention of heart disease (Harris and von Schacky, 2004) and C18:1 *trans*-11 may also have health benefits due to their conversion to CLA (Bu et al., 2007). Dairy products are the primary source of CLA in the human diet and provide about 10% of n-3 FA and 25% of n-6 FA (van Valenberg et al., 2013). This suggests that increasing the CLA and n-3 levels in milk may make it a more preferred food source. However, CLA have been shown to cause milk fat depression (Chouinard et al., 1999; Bauman and Griinari, 2001), and increases in the n-6 to n-3 ratio have caused decreased milk production (Greco et al., 2015) and proinflammatory responses in humans and rodents (Calder, 2012). Studying the time-course of CLA responses in milk can provide insight into the biological mechanism involved in these changes (Harvatine and Bauman, 2011). However, few studies are available on the time course of CLA changes for cows fed safflower seed.

Safflower is well suited to cultivation in warm desert areas, producing a seed high in oil (Dubois et al., 2007). Also among 80 common oil seeds, safflower seed (**SS**) has the highest content of linoleic acid, which is considered to have an improved profile of milk FA, specifically C18:1 *trans*-11, CLA, and n-6 content (Bell et al., 2006; Dschaak et al., 2011; Alizadeh et al., 2012). Hence, SS is a viable candidate as a fat source for dairy cows in these environments. Differences in FA responses may be due to use of different sources, different amounts of SS fed, different processing methods (Bu et al., 2007), or the amount of linoleic and linolenic FA ingested (Lock and Bauman, 2004). The time course of milk FA changes due to feeding SS have not been well studied,

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although Bell et al. (2006) reported milk fat depression started the day of feeding, but CLA levels in milk did not occur until after 4 wk later.

The primary objective of this study was to determine the temporal relationships of milk FA changes due to feeding different SS amounts. Secondary objectives were to determine differences in milk fat yield (**MFY**) and milk fat % (**MFPct**) as well as differences in digestible OM between SS fed at 3 or 6% of the diet DM as an aid to explain FA changes.

MATERIALS AND METHODS

Animals and Management

Animal care and sampling procedures were approved by the Animal Care Committee of Bu-Ali University. For this study 18 lactating cows with a mean ± SD of 28.6 ± 4.1 kg average daily milk yield, 157 ± 19 DIM, 545 ± 48 kg of BW, and a BCS of 2.3 ± 0.3 (5 point scale) were used in a randomized complete block design with repeated measurements of variables. Animals were blocked by parity (either primiparous or multiparous (mean parity of 2.8 ± 1.4), average daily milk yield, and DIM allowing 6 cows for treatment to be assigned to 1 of 3 dietary treatments: no added SS (**SS0**); SS included at 3% of ration DM (**SS3**); or SS included at 6% of ration DM (**SS6**). Sufficient SS (variety IL-111) were obtained from a commercial source to last the whole experimental period (8 wk). Seeds were rolled through 5-mm mesh of a Wiley's Pulverizer (Ogaw Seiki, Tokyo, Japan) roller mill and were added to a TMR. The nutritional and FA content of the SS is presented in Table 1, whereas the ingredient and nutritional content of the diet, and FA profiles, are presented in Tables 2 and 3, respectively. Diets were designed to be iso-nitrogenous and nearly iso-energetic, as well as being nutritionally adequate for 550-kg cows producing 32 kg of milk according to NRC (2001). Cows were housed in individual tie stalls with continuous access to water and allowed access to an outside lot 2 times a day for 1 h of exercise. The TMR was fed twice daily at 0800 and 1800 h, and cows were milked 3 times daily at 0500, 1300, and 2100 h. Cows were weighed and body scored weekly during the 8 wk of the experiment.

Samples and Analysis

Feed intake and milk yield were recorded daily, whereas samples of TMR were collected on d 2 and 6 of each week and were stored at −20°C until analyzed for nutrient composition. Dry matter of feeds was determined by drying at 110°C until a constant weight was obtained. Following drying, ash content was determined as the residue after 8 h in a 500°C furnace. Feed samples were analyzed for ether extract (**EE**) as method 920.39 and CP (as N × 6.25) method 955.04 (AOAC International, 2002). Fiber, as both NDF and ADF, was determined by the methods of Van Soest et al. (1991). Milk samples were obtained the last 2 d of each week. The 3 daily milk samples were mixed proportionally and a sub-sample of the mixed milk was taken for each cow per day and preserved using Bronopol tablets (Valio Ltd., Helsinki, Finland), and was analyzed for milk true protein, fat, and lactose using a Milko-Scan model 6 (Foss, Hillerød, Denmark). Milk samples were processed for SCC using a DeLaval cell counter (DeLaval, Tumba, Sweden) according to the manufacturer's protocol. Digestibility of ration nutrients were determined using acid insoluble ash as an indigestible marker using the technique of Van Keulen and Young (1977) and the equations from Zhong et al. (2008).

Rumen samples were taken on d 2 of each week 2.5 h after morning feeding by means of rumenocentesis as described by Tajik et al. (2011). The pH of the sample was measured immediately using a mobile AR 50 pH meter (Fisher Scientific GmbH, Hamburg, Germany). Another 10 mL was preserved with 1 mL of 5% sulfuric acid and frozen at −20°C until analysis for VFA and ammonia-N. After thawing, 5 mL of rumen fluid was vortexed with 1 mL of 250 g/L of meta-phosphoric acid and centrifuged at 3,000 × *g* for 20 min at 4°C to separate the supernatants. The VFA were determined by gas chromatography on a Chrompack CP 9002, model no. CP-9002 (Vulcanusweg 259, 2600 AM, Delft, the Netherlands) with a 50 m (0.32 mm ID) silica-fused

Table 1. Nutrient and fatty acid content of safflower seeds fed at 3 levels to mid-lactation dairy cows

Item	Value
Nutrient	
DM (%)	93.0
CP (% of DM)	19.5
NDF (% of DM)	38.1
ADF (% of DM)	29.7
EE[1] (% of DM)	40.2
Ash (% of DM)	1.2
Fatty acid (% of EE)	97.5
Fatty acid[2] (%)	
C14:0	1.2
C16:0	5.5
C18:0	2.2
C18:1	14.2
C18:2	74.0
C18:3	0.3
Others[3]	2.6

[1]EE: ether extract, a measure of crude fat.
[2]Fatty acids expressed as number of carbons:number of double bonds.
[3]Others determined by difference (100 − Σ fatty acids determined).