# AGRICULTURAL AND FOOD CHEMISTRY

# Fatty Acid Composition of Traditional and Novel Forages

WILLIAM M. CLAPHAM,\* JOYCE G. FOSTER, JAMES P. S. NEEL, AND JAMES M. FEDDERS

Appalachian Farming Systems Research Center, Agricultural Research Service, U.S. Department of Agriculture, 1224 Airport Road, Beaver, West Virginia 25813-9423

Managing the fatty acid composition of grazing ruminant diets could lead to meat and milk products that have higher conjugated linoleic acid (CLA) concentrations, but forage fatty acid dynamics must be more fully understood for a range of forages before grazing systems can be specified. The fatty acid profiles of 13 different forages, including grasses, legumes, and forbs, grown under greenhouse conditions, were determined. Three separate harvests, at 3-week intervals, were made of each plant material.  $\alpha$ -Linolenic [C18:3, 7.0–38.4 mg g<sup>-1</sup> of dry matter (DM)], linoleic (C18:2, 2.0–10.3 mg g<sup>-1</sup> of DM), and palmitic (C16:0, 2.6–7.5 mg g<sup>-1</sup> of DM) acids were the most abundant fatty acids in all species at each harvest, together representing ~93% of the fatty acids present. Concentrations of fatty acids declined as plants developed, but the fractional contribution of each fatty acid to total fatty acids remained relatively stable over time. Grasses had a uniform composition across species with a mean of 66% of total fatty acids provided by C18:3, 13% by C18:2, and 14% by C16:0. The fractional contribution of C18:3 to total fatty acids was lower and more variable in forbs than in grasses. Intake of fatty acid by grazing ruminants would be affected by the forage species consumed.

KEYWORDS: Fatty acids; forage; ruminant nutrition; forage-finished beef; conjugated linoleic acid

### INTRODUCTION

The fatty acid profiles of meats and processed foods have recently become a subject of increased interest due to the beneficial or detrimental impact that individual fatty acids may have on human health. A number of studies demonstrate that fat profiles in ruminant meat and dairy products are affected by the animal's diet (1). Therefore, the fatty acid profiles of forage-finished meat products are expected to vary depending, in part, on the fatty acid content of the forage consumed during the finishing period. A detailed understanding of fatty acid dynamics among a range of forage species is needed to describe (and manage) the complex association between the fatty acid content of the forages consumed and the fatty acid profile of pasture-finished beef products.

 $\alpha$ -Linolenic and linoleic acids are the predominant unsaturated fatty acids in forages (2), with  $\alpha$ -linolenic acid concentrations as high as 50–75% of the total lipid fraction (3). Conjugated linoleic acids (CLA) are a family of fatty acids that are isomers of linoleic acid. In human diets CLA are associated with lower risk of vascular diseases, certain cancers, diabetes, and obesity (4). Meat and milk from ruminants are the main sources of CLA in human diets (5, 6). Milk from cows grazed on pasture without supplemental grain or concentrate had 500% more CLA than milk from cows fed grain and concentrate (1). The relative contents of CLA isomers in beef meat (7) and in bovine milk (8) are associated with diet. Supplementation of cattle diets with bruised linseed or fish oils that are high in  $\alpha$ -linolenic acid resulted in higher proportions of omega-3 polyunsaturated fatty acids and lower proportions of long-chain omega-6 polyunsaturated fatty acids (except for C22:5 omega-3) (9). Fish oil and high  $\alpha$ -linolenic acid diets resulted in increased *cis*-9, *trans*-11 CLA and *trans*-vaccenic acid concentrations in bovine milk (*10*).

Dewhurst et al. (11) demonstrated that plant species, cutting date, and cutting interval have a significant impact on polyunsaturated fatty acid concentrations in forage. They observed that fatty acid profiles were distinctly different among forage species; however, fatty acid profiles could not be used to differentiate among rye and fescue species. Boufaïed et al. (12) observed that polyunsaturated fatty acid concentrations were greater in *Phleum pratense* L. at earlier versus later stages of development and in plants receiving greater N fertilization. Concentrations of polyunsaturated fatty acids were significantly lower in *Lolium perenne* L. that had been wilted for an extended period than in corresponding fresh herbage (13).

This study is a preliminary investigation conducted in a greenhouse to determine the fatty acid profiles of conventional and novel forages and forbs that have potential applications in Appalachian pastures used for finishing beef cattle. The data will be used to design forage systems that maximize the intake of  $\alpha$ -linolenic acid by grazing cattle during the finishing phase.

#### MATERIALS AND METHODS

Plant Growth and Sample Preparation. Plant materials, representing a range of traditional and novel forages, were grown from seed

<sup>\*</sup> Author to whom correspondence should be addressed [e-mail william.clapham@ars.usda.gov; telephone (304) 256-2857; fax (304) 256-2921].

Table 1. Plant Materials Evaluated for Fatty Acid Content

	Station data system. The fatty acids quantified on a dry matter basis
	were lauric (C12:0), myristic (C14:0), palmitic (C16:0), palmitoleic
h a mira at	(C1(1)) stands $(C19(0))$ slide $(C19(1))$ limit $(C19(2))$ and a limit slide

common name	scientific name	cultivar	harvest schedule <sup>a</sup>
triticale	× Triticosecale Wittmack	Trical 102	4, 7, 10
orchardgrass	Dactylis glomerata L.	Benchmark	4, 7, 10
perennial ryegrass	Lolium perenne L.	Seville	4, 7, 10
tall fescue	Festuca arundinacea Schreb.	Ky 31	4, 7, 10
galega	Galega orientalis Lam.	Common	5, 8, 11
white clover	Trifolium repens L.	Huia	5, 8, 11
chicory	Cichorium intybus L.	Forage Feast	4, 7, 10
chicory	Cichorium intybus L.	INIA le Lacerta	4, 7, 10
chicory	Cichorium intybus L.	Grasslands Puna	4, 7, 10
rape	Brassica napus L.	Barnapoli	3, 6, 9
turnip	Brassica rapa L.	Barkant	3, 6, 9
borage	Borago officinalis L.	Common	3, 6, 9
plantain	Plantago lanceolata L.	Lancelot	5, 8, 11

<sup>a</sup> Weeks after sowing for first, second, and third harvests.

under greenhouse conditions from mid-January through mid-March 2002 (Table 1). Seeds (30-50 per 30-cm white plastic pot) were planted in commercial potting medium (Pro-Mix BX, Premier Horticulture Ltd., Dorval, PQ, Canada) amended with slow-release fertilizer [Osmocote 15-9-12 plus minor nutrients (Scotts, Marysville, OH), 25 g per pot, incorporated by hand to a depth of 8 cm]. Additional nutrient (same type and amount used at seeding) was surface-applied 60 days after seeding. The potting medium was thoroughly moistened with tap water prior to seeding and irrigated automatically thereafter to maintain adequate soil moisture. Greenhouse temperatures were maintained between 13 and 25 °C with automated heating and ventilation systems. A 12-h photoperiod was achieved with supplemental lighting (metal halide). Seedlings were thinned to 25 per pot within 3 weeks after sowing. The experimental design was a randomized complete block with pots as experimental units and tables representing blocks. A total of 195 pots (experimental units) were used to accommodate the 13 plant materials, 3 harvests, and 5 replicates. Initial harvest of a plant material was made when forage reached 95% cover of the pot surface, estimated visually (3-6 weeks after seeding, Table 1). Second and third harvests were taken at 3-week intervals thereafter from previously unharvested pots. This three-harvest regime allowed evaluation of fatty acid dynamics as the plants grew. Shoots (including any stem and all leaves) were frozen immediately in liquid nitrogen and then maintained at -85 °C or below until lyophilized. Herbage from the second and third harvests was cut to lengths of 5-8 cm prior to submersion in liquid nitrogen to facilitate drying. Dried samples were ground sequentially through a Wiley mill (2-mm screen) and a cyclone mill (0.5-mm screen) and then stored under a nitrogen atmosphere at -85°C until analyzed.

Fatty Acid Extraction and Quantification by GC. Fatty acids were analyzed using the protocol of Sukhija and Palmquist (14). Correction for incidental variations induced during extraction and methylation procedures was accomplished using an orchardgrass check sample (oven-dried and ground to a 0.5-mm particle size) that was collected from an ongoing field experiment. Chromotagraphic separation of fatty acid methyl esters was accomplished with a Hewlett-Packard (Wilmington, DE) model 6890 GC equipped with electronic pneumatics control, a model 7683 automatic liquid sampler, and a flame ionization detector. Samples  $(2 \mu L)$  were introduced by split injection (50:1 ratio) onto a WCOT fused silica, chemically bonded capillary column (Chrompack CP-select CB for FAME, 100 m long, 0.25-mm inside diameter, 0.39-mm outside diameter, 0.25-µm film thickness; Varian, Walnut Creek, CA). Helium (3 mL min<sup>-1</sup>) was used as the carrier gas. The temperature gradient (70-250 °C) consisted of the following steps: 70 °C for 1 min; increase to 135 °C at 90 °C min<sup>-1</sup>, hold for 1 min; increase to 160 °C at 1.5 °C min<sup>-1</sup>, hold for 0.5 min; increase to 185 °C at 1 °C min<sup>-1</sup>, hold for 0.5 min; increase to 195 °C at 60 °C min<sup>-1</sup>, hold for 5.5 min; increase to 250 °C at 90 °C min<sup>-1</sup>, hold for 3 min. Total run time was 54.7 min. Injector temperature was 280 °C; detector temperature was 300 °C. Fatty acids were identified according to their retention times using reference standards (GLC-63B, Nu-Check-Prep, Elysian, MN) and quantified using a Hewlett-Packard Chem(C16:1), stearic (C18:0), oleic (C18:1), linoleic (C18:2), and  $\alpha$ -linolenic (C18:3). Heptadecanoic acid (C17:0, 0.4 mg mL<sup>-1</sup> in hexane; Matreya, Pleasant Gap, PA) was used as the internal standard. Data Analysis. Significant effects of plant material, harvest time, and their interaction were assessed for individual and total fatty acid concentrations by analysis of variance. The fractional contribution of each fatty acid to the total measured fatty acid content was calculated. Fractional content values were square root transformed prior to performing analyses of variance (15). When the concentration of lauric

acid fell below detection limits (n = 20), a value of 0.001 mg g<sup>-1</sup> of dry matter (DM) was used to calculate total and relative fatty acid

concentrations. Forage quality parameters including crude protein content, net energy for maintenance (NEM), and net energy for gain (NEG) were determined for each sample using near-infrared reflectance spectroscopy (NIRS). Ground samples were scanned on a FOSS NIRSystems (Laurel, MD) model 6500 near-infrared reflectance spectrometer system (firmware version 156) using WinISI Winscan software (version 1.50). Dry matter intakes (DMI) were calculated from NIRS estimates of forage energy content and NRC (16) DMI eq 7-a. The estimates are based on a 431 kg (shrunk body weight) medium-frame steer (Bos taurus). Fatty acid intakes were then estimated as the product of DMI and fatty acid concentration. Animal performance estimates are based on animal energy requirements (16). Performance and intake estimates are presented purely for comparative purposes and are not intended to imply actual results under field conditions.

#### RESULTS

 $\alpha$ -Linolenic acid was the dominant fatty acid in all species with an average concentration across all species and harvests of 23 mg  $g^{-1}$  of DM (**Table 2**), contributing an average of 62% of the total measured fatty acids. Linoleic and palmitic acids were the next most abundant fatty acids. Each averaged  $\sim 5.5$ mg  $g^{-1}$  of DM (**Table 2**), and each contributed an average of 16% of the total measured fatty acids. Of the remaining measured fatty acids, lauric acid consistently had the lowest concentration, averaging only  $\sim 0.03 \text{ mg g}^{-1}$  of DM. Both stearic and myristic acids had concentrations averaging  $\sim 0.41 \text{ mg g}^{-1}$ of DM, and palmitoleic and oleic acids averaged 0.75 mg  $g^{-1}$ of DM. Combined, these minor fatty acids contributed only  $\sim$ 6% to the total measured fatty acid pool and will not be discussed further due to the minimal expected impact of these acids on the fatty acid profile of grazing ruminant products.

Plant material, harvest time, and their interaction had significant (P < 0.01) effects on  $\alpha$ -linolenic acid concentration as well as the fractional contribution of  $\alpha$ -linolenic acid to total fatty acids. The concentrations of  $\alpha$ -linolenic acid declined in all plant materials between the first and third harvests (Table **2**). First-harvest concentrations ranged from 14 mg  $g^{-1}$  of DM for borage to  $>38 \text{ mg g}^{-1}$  of DM for galega and Forage Feast and Puna chicories. The decline in concentration over the 6 weeks between the first and third harvests ranged from <30%for perennial ryegrass, galega, and Forage Feast chicory to >50% for triticale, Lacerta chicory, rape, and borage. The fractional contribution of  $\alpha$ -linolenic acid to total fatty acids at first harvest ranged from 0.44 to 0.72 g of  $\alpha$ -linolenic acid g<sup>-1</sup> of total fatty acids for borage and galega, respectively. Unlike concentrations, fractional contributions changed little as plants matured. Only white clover and Lacerta chicory showed modest declines in fractional contribution from 0.61 to 0.57 g of  $\alpha\text{-linolenic}$  acid  $g^{-1}$  of total fatty acids and from 0.62 to 0.56 g of  $\alpha$ -linolenic acid g<sup>-1</sup> of total fatty acids, respectively, between the first and third harvests.

Linoleic acid concentrations, like those of  $\alpha$ -linolenic and the other fatty acids measured, were significantly (P < 0.01)

		mg g <sup>-1</sup> of dry matter								
plant material	harvest	lauric	myristic	palmitic	palmitoleic	stearic	oleic	linoleic	$\alpha$ -linolenic	total
triticale	1	0.022	0.55	5.43	1.16	0.25	0.94	5.17	30.0	43.5
	2	0.064	0.38	3.83	0.56	0.19	0.62	3.38	19.4	28.4
	3	0.066	0.26	3.04	0.35	0.14	0.52	2.73	13.2	20.3
orchardgrass	1	0.031	0.56	6.81	1.19	0.30	1.10	7.97	34.4	52.3
	2	0.043	0.50	5.49	0.80	0.27	0.65	5.84	27.1	40.7
	3	0.077	0.41	4.41	0.56	0.23	0.42	4.66	21.0	31.7
perennial ryegrass	1	0.027	0.62	6.99	0.94	0.30	1.46	6.76	34.7	51.8
	2	0.046	0.62	6.35	0.74	0.32	1.01	5.74	31.5	46.3
	3	0.072	0.61	5.91	0.56	0.28	0.71	5.47	26.8	40.5
tall fescue	1	0.027	0.50	5.91	1.23	0.24	1.54	5.70	28.4	43.5
	2	0.044	0.48	4.94	0.97	0.22	1.03	4.12	25.3	37.1
	3	0.075	0.36	3.78	0.53	0.16	0.64	3.01	17.1	25.7
galega	1	0.019	0.58	7.24	1.38	0.97	0.70	5.15	38.4	54.5
	2	0.024	0.51	6.49	0.91	0.89	0.33	3.80	26.0	38.9
	3	0.045	0.46	5.98	0.86	0.78	0.33	3.66	30.7	42.8
white clover	1	0.019	0.42	6.52	1.01	0.54	1.40	8.23	26.7	44.8
	2	0.023	0.42	5.62	0.75	0.47	0.89	5.89	20.3	34.4
	3	0.104	0.51	4.85	0.59	0.44	1.21	6.27	17.8	31.8
chicory (Forage Feast)	1	0.014	0.46	7.63	1.33	0.29	1.33	10.69	39.6	61.3
	2	0.016	0.45	6.32	0.88	0.25	0.48	8.08	25.6	42.1
	3	0.007	0.42	5.69	0.78	0.25	0.33	6.42	25.0	38.9
chicory (Lacerta)	1	0.019	0.43	7.16	1.28	0.29	1.18	10.49	35.3	56.2
	2	0.024	0.38	5.42	0.74	0.25	0.40	6.88	21.1	35.2
	3	0.024	0.27	4.64	0.51	0.24	0.48	5.74	14.8	26.7
chicory (Puna)	1	0.013	0.46	7.39	1.25	0.25	1.24	9.69	42.5	62.8
	2	0.030	0.42	5.65	0.81	0.22	0.43	7.17	24.2	38.9
	3	0.013	0.35	5.01	0.63	0.22	0.31	5.88	19.8	32.2
rape	1	0.018	0.44	6.28	0.94	1.00	0.71	5.80	21.1	36.2
	2	0.022	0.27	3.03	0.25	0.60	0.25	2.84	10.4	17.6
	3	0.019	0.22	2.55	0.18	0.52	0.25	2.43	8.2	14.4
turnip	1	0.019	0.41	6.43	1.12	0.80	0.46	3.99	22.5	35.8
	2	0.025	0.28	3.95	0.42	0.56	0.18	2.04	13.9	21.3
	3	0.021	0.25	3.55	0.31	0.52	0.22	1.85	11.4	18.2
borage	1	0.001	0.39	7.01	0.78	0.74	2.37	7.18	14.0	32.5
	2	0.017	0.30	5.28	0.44	0.56	1.02	4.67	10.8	23.1
	3	0.020	0.22	3.98	0.27	0.45	0.81	3.37	7.0	16.1
plantain	1	0.000	0.51	6.64	1.03	0.37	0.73	8.68	26.1	44.0
	2	0.000	0.45	5.28	0.67	0.38	0.39	6.19	22.6	35.9
	3	0.011	0.33	3.72	0.39	0.34	0.30	4.49	15.2	24.7
SEM	0.009	0.03	0.17	0.04	0.02	0.05	0.24	1.98	2.30	
	df					mean square				
plant material (PM)	12	0.004**a	0.121**	11.0**	0.48**	0.718**	1.63**	48.1**	684**	1127**
harvest (H)	2	0.010**	0.267**	90.1**	6.60**	0.309**	8.37**	158.7**	2741**	6530**
$PM \times H$	24	0.001**	0.014**	1.0**	0.05**	0.029**	0.22**	1.8**	50**	73**

<sup>a</sup> The double asterisk (\*\*) indicates significance at the 0.01 level of probability.

influenced by plant material, harvest time, and the plant material × harvest time interaction. The concentration of linoleic acid at first harvest ranged from <4.0 mg g<sup>-1</sup> of DM in turnip to >9.6 mg g<sup>-1</sup> of DM in the three chicory cultivars (**Table 2**). The decline in concentration between first and third harvests ranged from <28% in ryegrass and white clover to ≥53% in rape, turnip, and borage. The fractional contribution of linoleic acid g<sup>-1</sup> of total measured fatty acid in galega to >0.20 g of linoleic acid g<sup>-1</sup> of total measured fatty acid in plantain and borage. The fractional contribution of linoleic acid g<sup>-1</sup> of total measured fatty acid in plantain and borage. The fractional contribution of linoleic acid g<sup>-1</sup> of total measured fatty acid in plantain and borage. The fractional contribution of linoleic acid changed little as the plants grew.

Palmitic acid is the dominant saturated fatty acid in the plant materials investigated (**Table 2**). At first harvest, concentrations ranged from 5.4 mg g<sup>-1</sup> of DM for triticale to 7.0 mg g<sup>-1</sup> of DM or more for perennial ryegrass, galega, borage, and the three chicory cultivars. Concentrations declined in all plant materials as plants grew. Declines ranged from only 19% in perennial ryegrass to 59% in rape between the first and third harvests. Borage had the highest fractional content of palmitic acid, ranging from 0.22 to 0.25 g of palmitic acid g<sup>-1</sup> of total fatty acid for the first and third harvests, respectively. Grasses tended to have the lowest fractional content of palmitic acid, with ranges from 0.13 to 0.15 g g<sup>-1</sup> of total fatty acid, depending on species and harvest time.

The dynamics of total fatty acid concentration followed the patterns of the dominant fatty acids discussed above. Total concentration was significantly (P < 0.01) influenced by plant

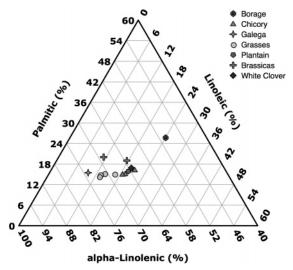


Figure 1. Relationships among the 13 plant materials with respect to relative concentrations of  $\alpha$ -linolenic, linoleic, and palmitic acids averaged over the three harvests. The value plotted for an individual fatty acid is the concentration of that fatty acid as a percentage of the total concentration of the three fatty acids shown, not the total concentration of all fatty acids measured.

material, harvest, and their interaction (**Table 2**). Total concentration was highest for all plant materials at the first harvest and declined with time. Total concentration at the first harvest ranged from 55 mg of fatty acid  $g^{-1}$  of DM for Forage Feast and Puna chicories to  $\leq 35$  mg  $g^{-1}$  of DM for rape, turnip, and borage. Declines between the first and third harvests ranged from 60% for rape to ~25% for perennial ryegrass and galega.

Differences in the fatty acid profiles among the plant materials are apparent in Figure 1, which spatially depicts the 13 plant materials as a function of the mean fractional concentrations (averaged over three harvests) of  $\alpha$ -linolenic and linoleic acid, the two dominant unsaturated acids, and palmitic acid, the single dominant saturated acid. The highest proportion of palmitic and linoleic acids along with the lowest proportion of  $\alpha$ -linolenic acid is a unique suite of characteristics that sets borage apart from the other plant materials analyzed. Intermediate levels of palmitic acid in brassicas separate them from the remaining plant materials. The relative proportions of palmitic acid among the grasses, legumes, plantain, and chicory cultivars are similar (ranging from 15 to 17%); however, there are differences among these plant materials in regard to  $\alpha$ -linolenic and linoleic acids. For instance, galega has the lowest proportion of linoleic acid and the highest proportion of  $\alpha$ -linolenic acid among the species studied. The chicory cultivars, white clover, and plantain tend to have a higher proportion of linoleic acid and a lower proportion of  $\alpha$ -linolenic acid, whereas the grasses are intermediate between this group and galega.

Forage nutritive values, estimated DMI, ADG, and intakes of linoleic and  $\alpha$ -linolenic acids are all significantly (P < 0.01) affected by plant material, harvest time, and their interaction (**Table 3**). Crude protein contents for all forages from the first harvest were high, ranging from 25 to 31%. Crude protein contents (<9.0%) were in the third harvests of triticale, plantain, and rape. Estimated intake of linoleic acid ranged from 17 g day<sup>-1</sup> (turnip, third harvest) to 89 g day<sup>-1</sup> (Forage Feast chicory, first harvest), and intake of  $\alpha$ -linolenic acid ranged from 67 g day<sup>-1</sup> (borage, third harvest) to 350 g day<sup>-1</sup> (Puna chicory, first harvest).

#### DISCUSSION

Interest in fat profiles in human diets, particularly increasing the amount of CLA, is driving development of forage systems for production of pasture-raised beef products. In cattle, dietary substrates high in  $\alpha$ -linolenic acid appear to facilitate establishment of rumen microflora that are associated with the greatest deposition of CLA in meat and milk (7). Data from the current study demonstrate that herbages from grasses, legumes, and forbs vary greatly in their fatty acid profiles, particularly in concentrations of  $\alpha$ -linolenic, linoleic, and palmitic acids. These data therefore suggest that the amount of CLA in pasture-raised beef can be influenced by the plant species in the pasture. Although absolute concentrations of all measured fatty acids (except lauric acid) declined as plants developed, fatty acid composition remained stable. Even plants, such as white clover and borage, that became reproductive and flowered by the third harvest showed little change in relative amounts of individual fatty acids. These results are consistent with those reported by Boufaïed et al. (12). Observed decreases in the concentrations of forage fatty acids can be attributed to dilution effects of growth and increased concentrations of other metabolites such as cellulose, hemicellulose, and lignin.

 $\alpha$ -Linolenic acid concentrations were higher in our lyophilized samples than in samples that had been oven-dried at 55 °C for 2 days (12), conditions that could have reduced concentrations of labile fatty acids. We observed similar differences in fatty acid composition when comparing subsamples of freshly harvested tissues that were either frozen and lyophilized or ovendried at 55 °C (unpublished data). In an experiment examining the effects of forage conservation method on fatty acid composition, Dewhurst and King (13) observed that fresh grass contained higher concentrations of palmitic, linoleic, and  $\alpha$ -linolenic acids than either wilted grass or grass hay. Thus, grazing is likely to be the best approach for increasing CLA in meat and milk of ruminants consuming all-forage diets.

Pasture management will play an important role in forage fatty acid composition. Dewhurst et al. (11) observed that fatty acid composition differed among grass species and that fatty acid concentration declined with cutting date until late-season harvests under cool conditions. However, differences among grass species were not great enough to differentiate between fescues and ryegrasses. Concentrations of palmitic, linoleic, and  $\alpha$ -linolenic acids in the grasses we studied (orchardgrass, perennial ryegrass, triticale, and tall fescue) were also similar. Other plant materials such as Puna chicory, which had a high mean fatty acid concentration but markedly greater variability (Table 2), may present special challenges when defining pasture species combinations to achieve defined CLA objectives in beef and dairy products. Variability in fatty acid concentration with plant development could affect the utility of forages in grazing systems. It is possible that variability in forage fatty acid content could be minimized in some species by management that maintains the forage in a vegetative state. Other opportunities to influence the fatty acid composition of forages were revealed by Mayland et al. (17), who observed that total fatty acid composition of perennial ryegrass was correlated with chlorophyll a + b concentrations and increased with herbage N concentration.

The immature, rapidly growing tissues collected during our first harvest had levels of crude protein that exceeded nutritional needs of beef cattle (16). Consumption of this herbage by cattle could negatively affect rumen microflora energy balance (18), thereby contributing to the development of off-flavors in the meat (19). Crude protein contents for plantain (third harvest),

Table 3. Forage Quality and Modeled Dry Matter Intake, Average Daily Gain, and Fatty Acid Intake for the 13 Plant Materials Evaluated<sup>a</sup>

		crude protein (%)	NEM <sup>b</sup> (MJ kg <sup>-1</sup> )	NEG (MJ kg <sup>-1</sup> )	DMI (kg day <sup>-1</sup> )	ADG <sup>c</sup> (kg day <sup>-1</sup> )	fatty acid intake		
plant material	harvest						linoleic (g day <sup>-1</sup> )	$lpha$ -linolenic (g day $^{-1}$ )	linoleic/ α-linolenic
triticale	1	26.9	8.6	5.8	8.8	1.4	46	264	0.17
	2	12.0	7.7	5.1	9.4	1.2	32	182	0.17
	3	8.7	7.5	4.9	9.5	1.2	26	125	0.21
orchardgrass	1	29.9	8.8	6.0	8.6	1.4	69	296	0.23
	2	19.3	7.8	5.1	9.4	1.2	55	254	0.22
	3	11.4	7.1	4.5	9.6	1.1	44	198	0.22
perennial ryegrass	1	30.6	8.7	5.9	8.8	1.4	59	304	0.19
	2	23.0	8.5	5.8	8.9	1.4	51	279	0.18
	3	19.0	8.0	5.4	9.2	1.3	51	248	0.20
tall fescue	1	29.7	8.8	6.0	8.7	1.4	49	246	0.20
	2	19.8	8.3	5.6	9.0	1.3	37	229	0.16
	3	11.5	7.5	4.9	9.5	1.2	29	162	0.18
galega	1	29.6	8.3	5.6	9.0	1.3	46	342	0.13
	2	23.4	7.7	5.1	9.4	1.2	36	244	0.15
	3	20.8	7.7	5.1	9.4	1.2	34	288	0.12
white clover	1	26.1	8.5	5.8	8.9	1.4	73	236	0.31
	2	21.2	8.1	5.4	9.2	1.3	54	187	0.29
	3	18.1	8.0	5.3	9.2	1.3	58	165	0.35
chicory (Forage Feast)	1	26.3	9.0	6.2	8.4	1.4	89	331	0.27
	2	18.1	8.7	5.9	8.7	1.4	71	224	0.32
	3	16.4	8.5	5.8	8.9	1.4	57	221	0.26
chicory (Lacerta)	1	26.6	9.0	6.2	8.4	1.4	88	295	0.30
	2	15.5	8.7	5.9	8.7	1.4	60	184	0.33
	3	11.7	8.2	5.5	9.1	1.3	53	135	0.39
chicory (Puna)	1	27.5	9.1	6.3	8.2	1.4	80	350	0.23
	2	16.3	8.7	6.0	8.7	1.4	62	210	0.30
	3	12.8	8.4	5.6	9.0	1.3	53	178	0.30
rape	1	30.9	9.0	6.1	8.4	1.4	49	178	0.28
	2	9.3	7.9	5.2	9.3	1.3	27	97	0.27
	3	8.2	7.4	4.8	9.5	1.2	23	78	0.30
turnip	1	29.9	9.2	6.3	8.2	1.4	33	185	0.18
	2	12.6	8.5	5.8	8.9	1.4	18	123	0.15
	3	12.1	8.2	5.5	9.1	1.3	17	104	0.16
borage	1	33.8	8.6	5.9	8.8	1.4	63	123	0.51
	2	15.3	8.1	5.4	9.2	1.3	43	99	0.43
	3	9.6	7.0	4.5	9.6	1.1	32	67	0.48
plantain	1	25.2	9.0	6.2	8.4	1.4	73	218	0.33
	2	14.6	8.2	5.5	9.1	1.3	57	206	0.27
	3	7.7	8.2	5.5	9.1	1.3	41	138	0.30
SEM		0.7	0.1	0.1	0.1	0.01	2	17	0.01
plant material (PM) harvest (H) PM × H	df 12 2 24	102** <i>c</i> 3617** 43**	1.53** 13.48** 0.30**	1.13** 9.96** 0.23**	mea 0.709** 7.112** 0.098**	n square 0.033** 0.222** 0.015**	$\begin{array}{c} 34 \times 10^{2**} \\ 76.3 \times 10^{2**} \\ 1.0 \times 10^{2**} \end{array}$	$\begin{array}{c} 48.9 \times 10^{3**} \\ 134 \times 10^{3**} \\ 3.4 \times 10^{3**} \end{array}$	0.116** 0.004** 0.034**

<sup>a</sup> Intake and average daily gain values were generated from the National Research Council (*16*) model for a 431 kg (shrunk weight) medium frame steer. <sup>b</sup> Abbreviations: NEM, net energy for maintenance; NEG, net energy for gain; ADG, average daily gain; DMI, dry matter intake. <sup>c</sup> The double asterisk (\*\*) indicates significance at the 0.01 level of probability.

rape (second and third harvests), and triticale (third harvest) were low and could also negatively affect animal or rumen microflora productivity. On the basis of estimated DMI and energy content, all forages would support ADG ranging from 1.1 to 1.4 kg and would be highly acceptable with regard to finishing cattle on forage (**Table 3**).

With our forages, estimated intake of linoleic acid ranged from 17 (turnip, third harvest) to 89 g day<sup>-1</sup> (Forage Feast chicory, first harvest), and intake of  $\alpha$ -linolenic acid ranged from 67 (borage, third harvest) to 350 g day<sup>-1</sup> (Puna chicory, first harvest). Supplementation of dairy cattle diets to provide total dietary levels of 400–675 g day<sup>-1</sup> of linoleic acid was shown to increase CLA and polyunsaturated fatty acid content of milk,

both as a percent of total fatty acids and in overall quantity produced per day (20, 21). Increases in total dietary  $\alpha$ -linolenic acid from 200 to 570 g day<sup>-1</sup> also increased CLA content in milk (20, 21). Estimated forage linoleic acid intakes (**Table 3**) are well below supplemented levels provided by Dhiman et al. (21) and AbuGhazaleh et al. (20), but  $\alpha$ -linolenic acid intakes for many of our forages are comparable to  $\alpha$ -linolenic acid levels in the supplemented treatments. In beef, French et al. (22) showed increased CLA content in intramuscular fat when cattle were finished on grass versus concentrate diets. In their experiment, the grass had similar fatty acid profiles and concentrations as our forages. Total linoleic acid intakes were similar between concentrate and grass diets (~40 g day<sup>-1</sup>); however,  $\alpha$ -linolenic acid intakes were approximately 4.8 and 140 g day <sup>-1</sup> for the concentrate and forage diets, respectively. Some of our forages collected during the second harvest would provide  $\alpha$ -linolenic acid intakes from 128 to 196% greater than those reported by French et al. (22). When cattle were finished on grass versus concentrate (22), intramuscular CLA, as a percent of total fatty acid, rose from 0.5 to 1.1%, whereas average milk CLA content increased from 0.5 to 1.75% across all treatments (20, 21).

Increased fat CLA content may be related to the ratio of linoleic to  $\alpha$ -linolenic acids in the diet. Supplementation of linoleic acid increased CLA percent when the linoleic acid/ $\alpha$ -linolenic acid ratio in the diet was 5:1 or greater, whereas supplemental  $\alpha$ -linolenic acid increased CLA concentration when the ratio was 1:1 or less (21). The ratio of linoleic to  $\alpha$ -linolenic acids in the grass was approximately 0.3:1.0 in the study conducted by French et al. (22) and is 0.5:1.0 or less for plant materials in our study (**Table 3**). On the basis of these data, increasing fat CLA content (as a percent of total fatty acids) through supplementation of oil in forage-based systems may be limited. Using forages with higher  $\alpha$ -linolenic acid content, relative to other forages, may be the better approach.

Finishing cattle on pasture could benefit the producer through increased profitability and benefit the consumer through creation of meat products with a higher CLA content than similar products from cattle finished on grain in the feedlot. Development of pasture-finishing systems that produce consistent, highquality products will require consideration of forage fatty acid, crude protein, and energy contents along with forage dry matter production. The range in forage fatty acid content reported here suggests that forage species selection and management will likely affect CLA content of pasture-finished beef products.

#### **ABBREVIATIONS USED**

ADG, average daily gain; DM, dry matter; DMI, dry matter intake; CLA, conjugated linoleic acid; NEG, net energy for gain; NEM, net energy for maintenance.

#### ACKNOWLEDGMENT

We thank Nathan Snyder for assistance with plant propagation and management, Robert Arnold and the late Julie de Thourars for help with plant harvest, and David Bligh, Jared Robertson, Chris Nacci, and Sarah Coffey for help with sample preparation and analysis.

## LITERATURE CITED

- Dhiman, T. R.; Anand, G. R.; Satter, L. D.; Pariza, M. W. Conjugated linoleic acid content of milk from cows fed different diets. J. Dairy Sci. 1999, 82, 2146–2156.
- (2) Harfoot, C. G.; Hazlewood, G. P. Lipid metabolism in the rumen. In *The Rumen Microbial Ecosystem*; Hobson, P. N., Ed.; Elsevier: New York, 1988; pp 285–322.
- (3) Hawke, J. C. Lipids. In *Chemistry and Biochemistry of Herbage*; Butler, G. W., Bailey, R. W., Eds.; Academic Press: New York, 1973; Vol. 1, pp 213–263.
- (4) Pariza, M. W. Perspective on the safety and effectiveness of conjugated linoleic acid. Am. J. Clin. Nutr. 2004, 79 (Suppl.), 1132S-1136S.
- (5) Chin, S. F.; Liu, W.; Storkson, J. M.; Ha, Y. L.; Pariza, M. W. Dietary sources of conjugated dienoic isomers of linoleic acid, a newly recognized class of anticarcinogens. *J. Food Compos. Anal.* **1992**, *5*, 185–197.

- (6) Wood, J. D.; Enser, M. Factors influencing fatty acids in meat and the role of antioxidants in improving meat quality. *Br. J. Nutr.* **1997**, 78 (Suppl.), S49–S60.
- (7) Mir, P. S.; McAllister, S.; Scott, S.; Aalhus, J.; Baron, V.; McCartney, D.; Charmley, E.; Goonewardene, L.; Basarab, J.; Okine, E.; Weselake, R. J.; Mir, Z. Conjugated linoleic acidenriched beef production. *Am. J. Clin. Nutr.* **2004**, *79* (Suppl.), 1207S-1211S.
- (8) Jiang, J.; Bjoerck, L.; Fonden, R.; Emanuelson, M. Occurrence of conjugated *cis-9*, *trans-11-octadecadienoic acid in bovine* milk: effects of feed and dietary regimen. *J. Dairy Sci.* **1996**, 79, 438–445.
- (9) Scollan, N. D.; Choi, N. J.; Kurt, E.; Fisher, A. V.; Enser, M.; Wood, J. D. Manipulating the fatty acid composition of muscle and adipose tissue in beef cattle. *Br. J. Nutr.* **2001**, *85*, 115– 124.
- (10) AbuGhazaleh, A. A.; Schingoethe, D. J.; Hippen, A. R.; Kalscheur, K. F. Conjugated linoleic acid and vaccenic acid in rumen, plasma, and milk of cows fed fish oil and fats differing in saturation of 18 carbon fatty acids. *J. Dairy Sci.* 2003, *86*, 3648–3660.
- (11) Dewhurst, R. J.; Scollan, N. D.; Youell, S. J.; Tweed, J. K. S.; Humphreys, M. O. Influence of species, cutting date and cutting interval on the fatty acid composition of grasses. *Grass Forage Sci.* 2001, *56*, 68–74.
- (12) Boufaïed, H.; Chouinard, P. Y.; Tremblay, F. F.; Petit, H. V.; Michaud, R.; Belanger, G. Fatty acids in forages. I. Factors affecting concentrations. *Can. J. Anim. Sci.* **2003**, *83*, 501–511.
- (13) Dewhurst, R. J.; King, P. J. Effects of extended wilting, shading and chemical additives on the fatty acids in laboratory grass silages. *Grass Forage Sci.* **1998**, *53*, 219–224.
- (14) Sukhija, P. S.; Palmquist, D. L. Rapid method for determination of total fatty acid content and composition of feedstuffs and feces. *J. Agric. Food Chem.* **1988**, *36*, 1202–1206.
- (15) Gomez, K. A.; Gomez, A. A. Statistical Procedures for Agricultural Research, 2nd ed.; Wiley: New York, 1984.
- (16) National Research Council. Nutrient Requirements of Beef Cattle, 7th rev. ed.; National Academy of Science, National Research Council: Washington, DC, 1996; pp 1–242.
- (17) Mayland, H. F.; Molloy, L. F.; Collie, T. W. Higher fatty acid composition of immature forages as affected by N [nitrogen] fertilization. *Agron. J.* **1976**, *68*, 979–982.
- (18) Wallace, R. J.; Cotta, M. A. Metabolism of N-containing compounds. In *The Rumen Microbial Ecosystem*; Hobson, P. N., Ed.; Elsevier: New York, 1988; pp 217–249.
- (19) Lane, G. A.; Fraser, K. A comparison of phenol and indole flavour compounds in fat, and of phenols in urine of cattle fed pasture or grain. N. Z. J. Agric. Res. 1999, 42, 289–296.
- (20) AbuGhazaleh, A. A.; Schingoethe, D. J.; Hippen, A. R.; Kalscheur, K. F. Milk conjugated linoleic acid response to fish oil supplementation of diets differing in fatty acid profiles. *J. Dairy Sci.* 2003, 86, 944–953.
- (21) Dhiman, T. R.; Satter, L. D.; Pariza, M. W.; Galli, M. P.; Albright, K.; Tolosa, M. X. Conjugated linoleic acid (CLA) content of milk from cows offered diets rich in linoleic and linolenic acid. J. Dairy Sci. 2000, 83, 1016–1027.
- (22) French, P.; Stanton, C.; Lawless, F.; O'Riordan, E. G.; Monahan, F. J.; Caffrey, P. J.; Moloney, A. P. Fatty acid composition, including conjugated linoleic acid, of intramuscular fat from steers offered grazed grass, grass silage, or concentrate-based diets. J. Anim. Sci. 2000, 78, 2849–2855.

Received for review July 15, 2005. Revised manuscript received October 13, 2005. Accepted November 1, 2005. Mention of trade names or commercial products in this paper is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

JF0517039