SURVEY OF RETAIL MILK: COMPARISON OF THE FATTY ACID COMPOSITION OF CONVENTIONAL MILK AND MILK LABELED AS "rbST-FREE" AND "ORGANIC"

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Abstract

In recent years the dairy case has expanded in its selection of milk and the consumer must decide which management practice he will support and which is healthiest: conventional, rbST-free or organic labeled milk. Conventional, rbST-free and organic labeled milk are characterized by different management practices on the dairy farm. The fatty acid composition of milk can be modified by manipulating the cow's diet; however it is unclear whether the milk labeling differences in cow management affect the composition of milk fat. The objective of the present study was to compare the fatty acid composition of conventional milk with that labeled as rbST-free or organic. Samples were collected from retail stores in the 48 contiguous states. A total of 296 samples were collected in blocks containing conventional, rbST-free and organic milk. Fatty acids were extracted, methylated, and gas chromatography was used to quantify individual fatty acids. Conventional and milk labeled as rbST-free showed no differences in fatty acid composition. Organically labeled milk was similar, with minor statistical differences from conventional and rbST-free milk. These differences included greater levels of *trans*-11 18:1 (vaccenic acid; VA), *cis*-9, *trans*-11 C18:2 (rumenic acid; RA), as well as greater total saturated fatty acids, lower monounsaturated fatty acids and lower total *trans* fatty acids. The variation in RA and VA in organic milk was greater as compared to the variation observed for conventional and rbST-free milk samples. Least-squares mean of VA and RA in organic milk tended to be greater than the median whereas the least-squares mean and median were similar in value in conventional and rbST-free milk samples. Overall, some differences were significant in organic milk fatty acid composition, but these were minor and quantitatively of no biological importance in comparing conventional, rbST-free and organic milk.

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Introduction

In recent years the dairy case has expanded in its selection of milk available to the consumer. Not only is it a choice between whole and non-fat milk, the consumer must now decide which management practice he will support and which is healthiest for him and his family: conventional or milk labeled as rbST-free or organic, to an uninformed consumer, these labels may be confusing. It is important for the consumer to be aware of the latest scientific research on the nutrition and safety of dairy products so that he is not swayed by advertising or marketing.

Conventional, rBST-free and organic labeled milk are characterized by different management practices on the dairy farm. rbST-free labeled milk comes from cows not supplemented with recombinant bovine somatotropin (rbST). This protein hormone increases milk yield by coordinating physiological processes in well fed cattle (Bauman, 1999). A trend in labeling milk as "rbST-free" has appeared due to consumer concern about hormones such as rbST and insulin like growth factor (IGF-1) contaminating milk. Organic labeled milk is produced under specific management practices which include no use of antibiotics and the growth hormone, rBST. Organically certified feeds, grown with organically approved pesticides and fertilizers are also fed. Pasture grazing is not a requirement for organic milk, yet many brands advertise that their milk comes from pasture grazing cows.

It is well established that the fatty acid composition of milk can be modified by manipulating the cow's diet; however it is unclear whether differences in cow management related to the milk labeling described above will affect the composition of milk fat. The present study was performed in conjunction with a retail survey by Vicini et al. (2008) to determine differences in these three categories of milk. Vicini et al (2008) examined whether rbST supplementation caused differences in overall nutrient composition or concentration of hormones in retail milk. The present investigation examined variations in fatty acid composition among the differently labeled milk. There has been much research done examining health implications of saturated and unsaturated fats, *trans* fats and conjugated linoleic acids (CLAs) and their link to cardiovascular disease, cancer, diabetes and other human health issues. The study will help to clarify the extent to which milk labeled as “rbST-free” or “organic” differ in fatty acid composition from conventionally produced milk. The data provided by this study, combined with the results from Vicini et al. (2008) will provide more information for consumers on possible differences among labeled milks to allow for more informed choices.

Review of Literature

I. Milk Fat Composition

In recent years there has been an increasing public awareness of the link between diet and health as well as a greater interest in the role of fat in the human diet. Dairy products account for approximately 15-25% of fat consumed by humans (O'Donnell, 1993). A large body of scientific literature has found that some of the fatty acids in dairy products may impact human health (Lock and Bauman, 2004). Fat is the most variable component of milk and the fatty acid composition of milk fat is highly diverse (MacGibbon and Taylor, 2006). This diversity is due to the presence of over 400 fatty acids found in milk.

Triacylglycerols are the primary component of milk fat, approximately 98% of the total fat composition. The remaining 2% is composed of monoacyl- and diacylglycerols, unesterified fatty acids, phospholipids and sterols (Walstra and Jenness, 1984). Because triacylglycerols account for such a large proportion of fat, their composition is largely the focus when considering the impacts of milk fatty acids on human health. Triacylglycerols are composed of a glycerol backbone with three esterified fatty acids. In bovine milk there are only 15 fatty acids that are present at or greater than a 1% concentration. These constitute the major fatty acids and can be identified using gas chromatography. They range from 4 to 18 carbons in length and vary in geometric conformation and biological activity.The approximate fatty acid profile of these major fatty acids in bovine milk and their relative amount, different chain length and configurations is depicted in Figure 1.

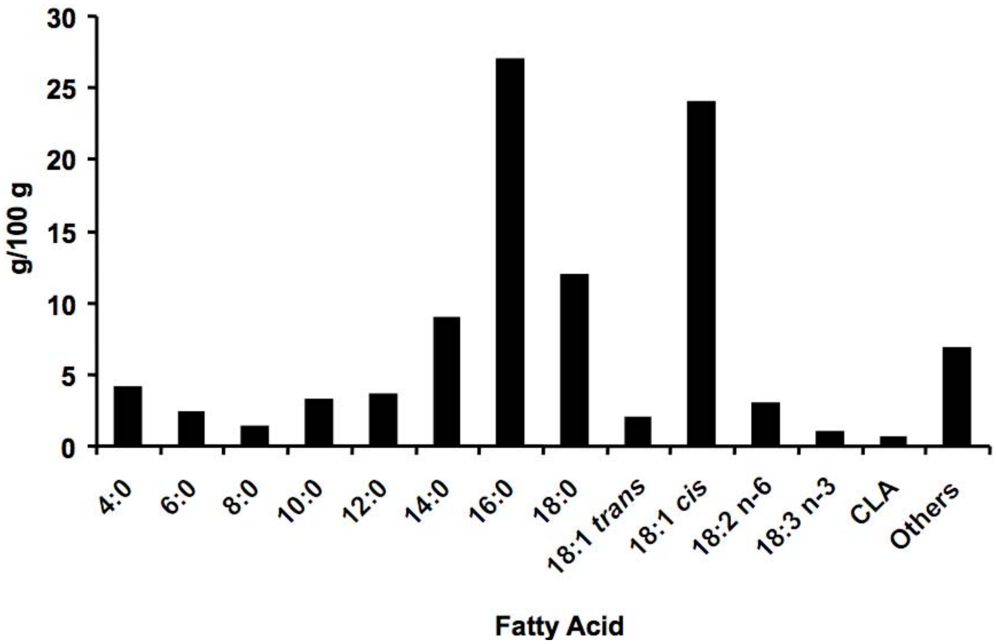

Figure 1. Fatty acid composition of bovine milk fat. Adapted from (Lock and Bauman, 2004).

Fatty acids in bovine milk come from two sources: by *de novo* synthesis in the mammary gland or preformed in plasma lipids originating from absorption from the small intestine, and body stores of adipose tissue. *De novo* synthesized fatty acids include those 4 to 16 carbon in length and represent about 45% of the total fatty acids in milk fat on a mass basis while preformed fatty acids from circulation are 16 carbons in length or longer and account for the remaining percentage (Moore and Christie, 1979). In dairy cows, *de novo* fatty acids are synthesized in the mammary gland from acetate and β-hydroxybutyrate. These precursors are obtained from the microbial fermentation of cellulose and hemicellulose in the rumen and are absorbed in the blood stream and carried to the mammary gland for uptake. In the mammary gland, acetate is used to form acetyl-CoA which is then carboxylated to malonyl-CoA and used to produce fatty acids in a series of elongation steps by fatty acid synthase (MacGibbon and Taylor, 2006).

Fatty acids 18 carbons in length or longer as well as part of the 16 carbon fatty acids, originate from lipid components in the ruminant diet. When dietary lipid enters the rumen it is hydrolyzed by extracellular lipases produced by rumen bacteria. Polyunsaturated fatty acids (PUFA) then undergo a second transformation called biohydrogenation. PUFA are toxic to rumen bacteria so they excrete microbial reductase enzymes that isomerize and hydrogenate double bonds, thereby detoxifying the fatty acids (Jenkins, 1993). Biohydrogenation of multiple double bonds is a complex process that most rumen bacteria cannot carry out completely. Instead such bacteria have been classified into two groups; Group A that can hydrogenate 18 carbon PUFA to *trans* 18:1 fatty acids and Group B that can hydrogenate *trans* 18:1 fatty acids to stearic acid (Hartfoot and Hazlewood, 1997).

Ruminants consume PUFA in their diet; linolenic acid (18:3) and linoleic acid (18:2) are abundant in grass and other ruminant feed products. However, the lipid profile in bovine milk does not match the lipid profile of the cow's diet because of the biohydrogenation of most of the dietary PUFA (Jenkins et al., 2007). Ruminants consume a large proportion of unsaturated fatty acids while their milk contains mostly saturated fatty acids. Figure 2 shows the pathway of ruminal biohydrogenation of both linolenic and linoleic acid.

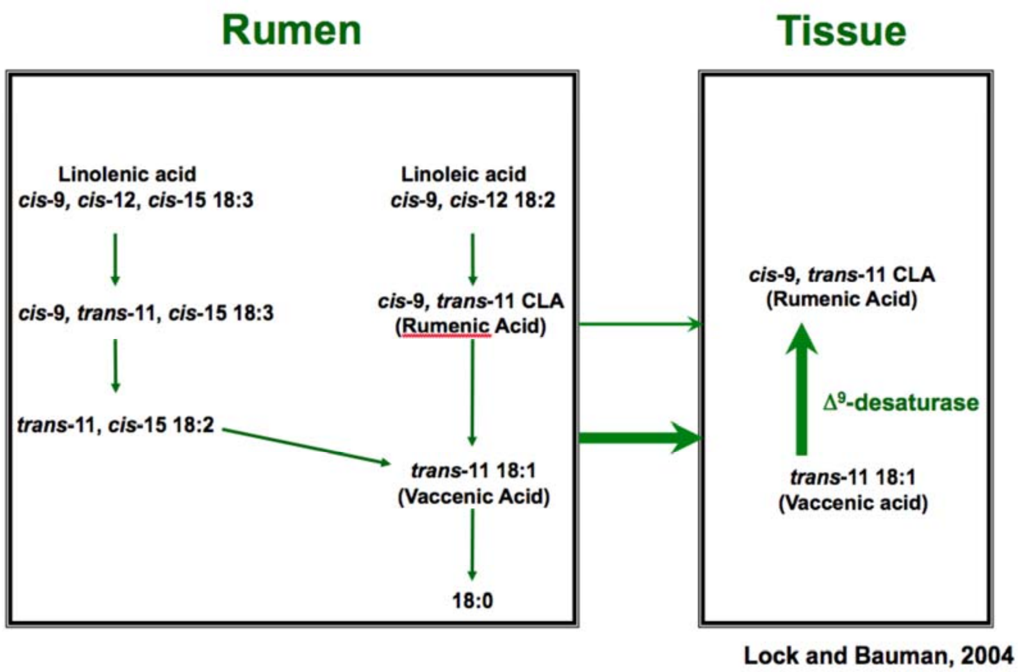

Figure 2. CLA synthesis in the ruminant. Adapted from Lock and Bauman (2004).

The transformation of linolenic acid involves several enzymes, saturating the double bonds until stearic acid is formed as the final product. Likewise, linoleic acid is first isomerized and then its double bonds hydrated to form stearic acid (18:0) (Bauman and Giinari, 2003). As a consequence of the rumen biohydrogenaton a high percentage of fatty acids that are absorbed from the small intestine are stearic acid. However, biohydrogenation of PUFA and rumen turnover are dynamic processes; as a result some *trans* C18:1 and conjugated C18:2 isomer intermediates in rumen biohydrogenation can escape from the rumen. Some are absorbed by the mammary gland and appear in ruminant milk fat. Samples from the duodenum and omasum have been collected and the fatty acid profile determined. As summarized by Palmquist et al. (2005) there are a range of intermediates produced during the biohydrogenation of linolenic and linoleic acid (Table 1). Thus, the extent to which dietary lipids are biohydrogenated in the rumen effects the fatty acid composition of ruminant milk.

Trans 18:1		Conjugated 18:2	
Isomer	Ruminal outflow (g/day)	Isomer	Ruminal outflow (g/day)
trans-4	0.5–0.7	*trans*-7, *cis*-9	<0.01
trans-5	0.4–0.6	*trans*-7, *trans*-9	<0.01–0.05
trans-6–8	0.4–6.7	*trans*-8, *cis*-10	0.01–0.02
trans-9	0.8–6.2	*trans*-8, *trans*-10	<0.01–0.10
trans-10	1.7–29.1	*cis*-9, *cis*-11	<0.01–0.01
trans-11	5.0–121.0	*cis*-9, *trans*-11	0.19–2.86
trans-12	0.5–9.5	*trans*-9, *trans*-11	0.22–0.55
trans-13 + 14	6.5–22.9	*trans*-10, *cis*-12	0.02–0.32
trans-15	3.2–8.5	*trans*-10, *trans*-12	0.05–0.06
trans-16	3.1–8.0	*cis*-11, *trans*-13	0.01–0.10
		trans-11, *cis*-13	0.01–0.46
		trans-11, *trans*-13	0.09–0.40
		cis-12, *trans*-14	<0.01–0.05
		trans-12, *trans*-14	0.08–0.19

Table 1. Distribution and ruminal outflow of *trans* 18:1 and isomers of conjugated 18:2 fatty acids in growing and lactating cattle. Adapted from Palmquist et al. (2005).

II. Milk Fatty Acids of Interest for Human Health

Milk and dairy products are nutritious components of the human diet; specific dietary lipids in milk have been implicated as impacting human health. Some fatty acids, such as saturated and *trans* fatty acids have been suggested to have a negative impact on human health (Keys et al., 1966; Mensink et al., 2003; Salter et al., 2007;), while others, like CLA and omega-3 fatty acids have proven to be beneficial to health maintenance and disease prevention (Griinari and Bauman, 1999; Simopoulos, 1999; Belury, 2002; Parodi, 2004; Bauman et al., 2005).

Ruminant fats are relatively more saturated than fats present in vegetable oils. This is largely due to biohydrogenation in the rumen. Saturated fats have been implicated in causing human health issues like cardiovascular disease and high cholesterol, and as a result whole milk dairy products have been deemed by some consumers as unhealthy. The link between coronary heart disease and saturated fat intake was first demonstrated in the 1950s when international surveys found that countries consuming the least amount of saturated fats had the lowest incidence of cardiovascular disease (Keys et al., 1966). Approximately two-thirds of the fatty acids found in milk are saturated. This would suggest that consuming milk fat would increase the risk of high cholesterol and coronary heart disease; however, 20% of these saturated fats are short chain fatty acids (C4:0-10:0) and 20% are stearic acid (18:0). These fatty acids are generally accepted as not having a negative impact on human health and present no risk of coronary heart disease and high cholesterol (Salter et al., 2007). Saturated fatty acids such as lauric, myristic and palmitic, have been found to increase pro-atherogenic LDL (Hegsted et al., 1965). However, a meta-analysis by Mensink et al. (2003) suggests that although such saturated fatty acids increase LDL, these adverse effects are offset by the beneficial effect of raising HDL. Therefore, the presence of saturated fat in the diet is neutral when coronary heart disease is compared to replacement by energy equivalent carbohydrates (Mensink et al., 2003).

In contrast to saturated fats, polyunsaturated fats, particularly those in the n-3 series have been shown to be very beneficial to human health. EPA (20:5n-3) and DHA (22:6-3) are omega-3 fatty acids that are found in milk in only small quantities; as a result, there is an opportunity and a continued effort to increase their concentration. Omega-3 fatty acids are essential and need to be obtained from the diet because humans do not possess the enzyme necessary to form a double bond in the n-3 position. The consumption of omega-3 fatty acids has been shown to reduce the risk of cardiovascular disease, type 2 diabetes, hypertension, cancer and maintain or enhance neurologic function (Larson et al., 2004; Lock and Bauman, 2004). In Western diets there is commonly an imbalance in omega-6 and omega-3 fatty acids in a 20-30 to 1 ratio. The ideal ratio is 4:1 or less. This is a cause for concern because these fatty acids compete for enzymes used to synthesize eicosanoids and such an imbalance can lead to immune and inflammatory problems (Simopoulos, 1999). When EPA and DHA are supplemented to the cow's diet, transfer efficiency to the milk appears to be low, about 2-4% (McConnell et al., 2004). This is mainly due to the extensive biohydrogenation that occurs in the rumen. However, even when the rumen bacteria are bypassed by abomasal infusions, transfer efficiency of EPA and DHA is still only about 25%, a value about one-third that of 18-carbon fatty acids (Lock and Bauman, 2004). Nevertheless, increases in concentration of these fatty acids in milk would be beneficial to human health as it would serve to increase the number of foods that can supply omega-3s and increase the nutritional value of dairy products.

Another bioactive component of milk fat is conjugated linoleic acid (CLA). The incomplete biohydrogenation of linoleic and linolenic acid results in many isomers of C18:1 and C:18:2 (Table 1). One biohydrogenation intermediate in milk fat that is of interest is *cis*-9, *trans*-11 CLA, known as rumenic acid (RA). It is important to note that RA is an intermediate in the biohydrogenation of linoleic acid, but not linolenic acid (Figure 2). Of the CLA isomers in milk fat, RA makes up about 75-90% of the total. Initially, CLA found in milk was thought to originate only from the incomplete biohydrogenation of linoleic acid (Lawless et al., 1998; Griinari and Bauman, 1999), but further investigation found inconsistencies with this concept. Nutritional studies found that CLA content of milk increased as dietary sources of linolenic acid were increased (Kelly et al., 1998a), yet RA is not an intermediate in the biohydrogenation of linolenic acid. The ratio of *trans*-11 18:1 (vaccenic acid; VA) to RA in the rumen fluid was found to be 50:1 while the ratio in milk was 3:1. Based on this conflicting information, Griinari and Bauman (1999) proposed that RA found in milk fat arose from two sources: incomplete biohydrogenation of linoleic acid in the rumen and endogenous synthesis in the tissue using the enzyme delta-9 desaturase with VA as the substrate. This theory was supported in a study by Griinari et al. (2000) that infused VA into the abomasums of dairy cows and found a 31% increase in RA in milk fat. Further studies have demonstrated that endogenous synthesis of RA via delta-9 desaturase and VA is in fact the major source of RA in ruminant milk (Corl et al., 2001; Kay et al., 2004). Humans also have the ability to synthesis RA from VA (Palmquist et al, 2005). Therefore, increases in both RA and VA in milk would have important implications for human health.

CLA, particularly the *cis*-9, *trans*-11 isomer, has been found to have a number of health benefits. CLA was originally found to be a potent antimutagen (Pariza et al., 1979; Ha et al, 1987). The use of CLA in cancer prevention has been investigated (Parodi, 2004), especially its potential in preventing breast cancer and mammary tumors (Bauman et al., 2005). Biomedical studies with animal models have also indicated that dietary supplementation of CLA can reduce the risk of developing atherosclerotic lesions and can possibly cause the regression of existing lesions (Krichevsky et al., 2000; Bauman et al, 2005). Biomedical studies testing the effects of other isomers of CLA, such as *cis*-10, *trans*-12, have found additional health benefits such as antidiabetogenic and anti-obesity effects as well as immuno- and bone growth modulation (Belury, 2002). Milk fat appears to contain many bioactive components capable of improving human health and preventing chronic diseases. Rather than reduce the consumption of dairy lipids to lower the intake of saturated fats, it may be more beneficial to alter the composition of milk to contain higher amounts of CLA and omega-3 fatty acids.

III. Diet Modification

There are a number of factors that can influence the fatty acid composition of bovine milk, including animal genetics, stage of lactation and season (Jensen, 2002). Alterations in diet have proven to be a highly effective and repeatable way to modify milk fat composition. The total amount of fat found in milk can be altered by changing the forage to concentrate ratio of the diet. High amount of concentrate or starch in the diet results in reduced fat synthesis and milk fat depression (Palmquist et al., 1993; Bauman and Giinari, 2003). Oilseed supplements, such as soybean and cottonseed, have also been shown to modify the fatty acid composition. For example, the addition of full fat soybean to the basal diet increased fat content, lowered the percent of de novo synthesized fatty acids (<C16:0) and increased the percentage of unsaturated, *trans*, omega-3 and omega-6 fatty acids in milk fat (Murphy et al., 1990). The addition of cottonseed and canola showed similar trends with a few exceptions: cottonseed reduced the proportion of 18:2 (DePeters and Cant, 1992) and canola reduced the overall milk fat percentage (Khorasani et al., 1991).

Changes in the fatty acid composition of milk have also been attributed to long term pasture grazing. There is a strong correlation between pasture grazing and an increase of CLA content in milk (Jahreis et al., 1997; Kelly et al., 1998b; White et al., 2001). Jahreis et al. (1997) sampled German dairy bulk tanks and found that cows grazing on pasture year-round had the highest levels of CLA in their milk. As pasture grazing decreased, CLA content decreased. Kelly et al. (1998b) also found that cows fed pasture had a significantly higher proportion of CLA in the milk fat compared to cows fed a concentrate diet. In addition, the levels of increased CLA stayed constant as long as the cow was feeding on pasture. Additionally, White et al. (2001) found that Holstein cows on pasture produced 83% more CLA than cows confined and fed a total mixed ration. The aforementioned citations provide some examples to illustrate the effect of diet on milk fat composition in dairy cows. For further information, the reader is referred to reviews that have summarized the literature relating to the ability of diet to alter the fatty acid profile of milk (Jensen, 2002, Ashes et al., 1997, Grummer, 1991, Kennelly, 1996, Palmquist et al., 1993, Sutton, 1989).

IV. Milk Labeling

Labeling milk as “rbST-free” or “organic” is becoming an increasingly popular trend; however, it is unclear to many consumers what these labels actually mean. It is also unclear whether there are differences in milk fat composition among labeled milk. All milk produced in the United States must adhere to strict government standards of quality and sanitation. The labeling of milk as between conventional or organic milk is not based on compositional differences, but instead is based on the production system and the practices followed on the farm. In fact, a statement from the American Dietetic Association says, “both organic and conventional farming supply nutritionally comparable foods” (The American Dietetic Association, 2009). Conventional based farming methods use the industry-accepted best standards to ensure that dairy cows are healthy and provide the animals with comfortable living conditions, a nutritious diet and good medical care. All milk producers are required to adhere to environmental practices to protect natural resources, properly dispose of waste and protect air and water quality (National Dairy Council, 2007).

Organically produced milk must follow additional standards outlined by the USDA's National Organic Program. These standards include: 1) cows must be given feed grown without the use of pesticides or commercial fertilizers, 2) cows must be given periodic (unspecified) access to pasture and sunlight, 3) cows must not be treated with supplemental hormones and 4) cows must not be given certain medications to treat illness (U.S. Department of Agriculture). In the United States, the commercial use of the term "organic" is legally restricted. Organically labeled milk must come from farms certified as adhering to the National Organic Program standards. Certification is handled by state, non-profit and private agencies approved by the USDA. There is an extensive application process, frequent farm inspections and record keeping for certified organic farms. Certification and labeling are necessary to differentiate organic dairy products from conventional dairy products because there is currently no test able to distinguish between organic and conventional milk (Molkentin, 2009).

Milk labeled as "rbST-free" is produced under the same standards as conventional milk, except that dairy farmers pledge not to administer recombinant bovine somatotropin to their cows. Somatotropin is a peptide hormone that occurs naturally in dairy cows to regulate growth and lactation; it is now commercially available as a supplement to increase the milk yield per cow (Bauman, 1999). Some consumers perceive that rbST supplements will contaminate the milk supply with a potentially harmful substance; however the safety of rbST has been tested extensively and without exception regulatory agencies through the world (> 50 countries) have concluded food products from rbST supplemented cows are safe for consumption. Most recently, Vicini et al. (2008) found that all commercial milk, conventional, rbST-free and organic, contained trace amounts of hormones and there were no biologically important differences among them. Additionally, in the case of rbST the activity of somatotropin is species specific and is not biologically active unless administered by injection to the appropriate species (Juskevich and Guyer, 1990).

The composition of milk from bST-supplemented cows has been examined in over 200 trials. The gross composition of milk, including milk fat, does not differ due to the use of bST (Juskevich and Guyer, 1990). Furthermore, the fatty acid composition of milk can be effected by a variety of genetic, physiological and environmental factors such as breed, stage of lactation, dietary ingredients, energy status and season, and these factors affect the milk composition of rbST-supplemented cows and non-supplemented cows in the same manner (Linn, 1988; Juskevich and Guyer, 1990). Fatty acid composition of milk fat can be altered by the energy balance of the cow and in early lactation when cows are in a negative energy balance. Many of the preformed lipids in milk are derived from body fat reserves. The resulting milk fat contains more long chain, unsaturated fatty acids (Chalupa and Galligan, 1989). These same effects are seen when rbST-supplemented cows are in negative energy balance.

The differences in the labeling of milk can be confusing and often consumers are making choices based on a perceived added value rather than actual differences among food products. This is certainly the case for milk labeled as conventional, rbST-free or organic. The current study focuses on milk fat and compares the fatty acid composition of retail milk fat from unlabeled and specialty labeled milk. This study represents the first research to investigate this and the information will help increase consumer awareness of the composition of the type of milk they purchase.

Materials and Methods

I. Milk Sampling

Milk samples were used from the Vicini et al (2008) study. Samples were collected from retail stores in the 48 contiguous states. They were purchased in blocks to minimize variation in shipping conditions. Each block contained milk of each of the following label types: 1) conventional 2) rbST-free and 3) organic. Milk was considered to be conventionally labeled when it did not contain any claims about rBST supplementation or organic production practices. rBST-free labeled milk claimed to be produced without the use of exogenous somatotropin. Organic milk was labeled as such and came from farms that were certified to meet the USDA organic standards.

Collection was as described by Vicini et al. (2008). Briefly, a block consisted of a shipping container collected on one day by one sampler in a specific city. Sample collectors were employees of the Monsanto Company living in each state. Milk was over-sampled from states that had larger populations or greater amounts of milk production. Milk samples were collected based on the following preferences: 1) whole milk for fat analysis 2) freshest milk based on expiration date and 3) paper or plastic container. Samplers made an effort to purchase all labels of milk from the same store, but if they could not locate one milk type, the samples were purchased and the block was incomplete. Purchased milk was pasteurized and did not include samples labeled as using ultra-high temperature pasteurization (UP). All samples were obtained in three weeks from October to November 2006. After purchase, the containers were sealed and shipped on ice to St. Louis by overnight mail. Subsequently, sample aliquots were shipped frozen overnight to Cornell University and kept frozen at -20º C until analysis for fatty acid composition. The geographic distribution for the milk samples is presented according to purchase location

(Figure 3) and processing location (Figure 4). Milk products have a code that indicates location of milk processing. Overall, the total samples analyzed were 111 conventional, 82 rbST-free and 99 organic.

II. Fatty Acid Analysis

The extraction of fatty acids was based on the Hara and Radin (1978) method and transmethylation according to Christie (1982) as modified by Chouinard (1999). Briefly this involved separation of the fat cake from milk by centrifugation, extraction by hexane isopropanol and sodium sulfate and methylation by NAoME reagent. Fatty acid methyl esters were quantified by gas chromatography (Hewlett Packard GCD system HP G 1800 A, Avondale, PA) fitted with a CP-Sil 88 capillary column (100mm x 0.25 mm i.d. with 0.2 µm film thickness; Varian Instruments, Walnut Creek, CA.) The oven temperature was initially set at 80ºC and ramped to 190º at 2 ºC/min and held for 20 minutes. The temperature was ramped again at 10 ºC/minute till 225 ºC was reached and held for 12 minutes. Fatty acid peaks were identified and quantified using pure methyl ester standards (NuCheck Prep, Elysian, MN). A butter oil reference standard (CRM 164; Commission of the European Communities, Community Bureau of Reference, Brussels, Belgium) was analyzed to control for column performance and to determine correction factors for fatty acids.

III. Statistical Analysis

Fatty acid composition of all 296 samples was used for statistical analysis. Data was analyzed using JMP 7.0; treatment (conventional, rbST-free and organic) was included in the model as a fixed effect and shipping block as a random effect. We are currently working on calculating standard error and P-values using nonparametric techniques. The data collected does not comply with the assumption of normality in variance and non skewed results. Organic samples showed especially high variance and larger range than either conventional or rBST-free samples. This trend was observed in many fatty acids; Figure 3 shows the variance in two particular fatty acids: C18:1, *t*11 and CLA, 9-11.

(A) C18:1, *t*11 **(B) CLA, 9-11**

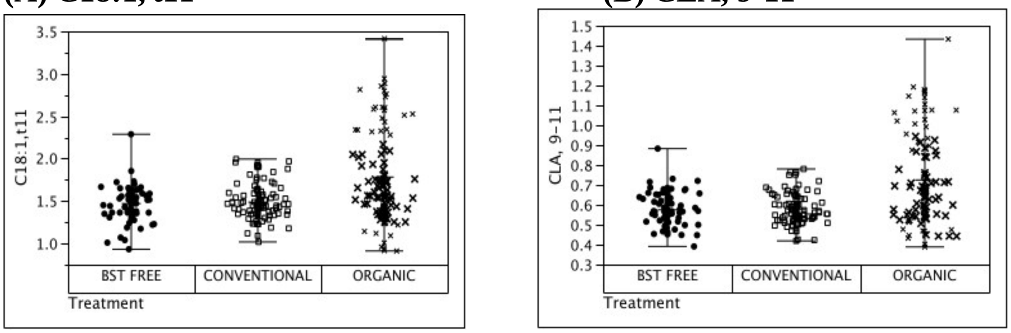

Figure 3. Variance in quantity of C18:1, *t*11 and CLA, 9-11 in 296 samples analyzed.

Results and Discussion

Conventionally labeled retail milk samples were purchased from all 48 contiguous states (Figure 4). Milk labeled rbST-free was not located in AR, ID, IN, MS, OH, OK, SD, TN and WY. Organic milk samples pasteurized by high temperature/short time (HTST) method were not obtained in LA, ME or MS.

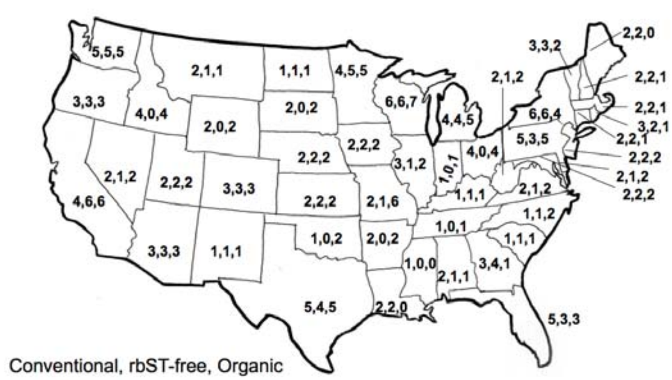

Figure 4. Number of samples purchased in the 48 contiguous states. Numerals represent the number of conventional, rbST-free and organic samples, respectively.

Although milk samples were purchased in all 48 contiguous states, the distribution of where the milk was processed differed with none being processed in ND, SD, WY, OK, WV, NJ and RI (Figure 5). This should not have a significant effect on the results of the data analysis seeing as milk was intentionally over-sampled in states that produce large quantities of beverage milk for consumer consumption. Such oversampling makes these data more representative of milk available in retail stores.

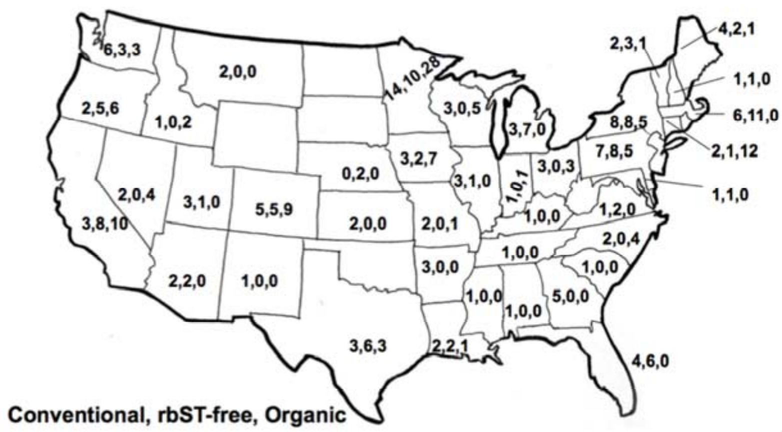

Figure 5. Number of samples processed in the 48 contiguous states. Numerals represent the number of conventional, rbST-free and organic samples, respectively. Milk samples processed in ND, SD, WY, OK, WV, NJ and RI were not obtained due to milk distribution between processing plants and retail stores.

Total Fatty Acid Composition

Least square means and standard errors were calculated for fatty acid composition of milk samples. Differences were observed between treatments for all fatty acids except C22:6 (Table 2). However, differences were minor and the total composition of fatty acids was similar among all treatments (Figure 6). Conventional, rbST-free and organic milk samples also showed the same general patterns in milk fatty acid content as has been described historically (Figure 1). Overall, conventional and rbST-free milk fatty acid composition was not statistically different; organic milk samples showed statistically different quantities of saturated fatty acids, *trans* fatty acids and CLA.

Table 2. Least squared means and standard errors for fatty acid composition of milk fat from 278 samples collected from the 48 contiguous states.

Variable, g/100g FA	Treatment[3]					
	Conventional		rbST-free		Organic	
	Mean	SD[4]	Mean	SD[4]	Mean	SD[4]
C4:0	4.22	0.30	4.17	0.28	4.36	0.31
C6:0	2.11	0.14	2.12	0.12	2.30	0.11
C8:0	1.13	0.07	1.15	0.09	1.26	0.07
C10:0	2.49	0.19	2.53	0.02	2.81	0.21
C12:0	2.83	0.22	2.89	0.26	3.24	0.28
C14:0	9.42	0.57	9.61	0.65	10.62	0.54
C14:1	0.86	0.09	0.89	0.11	0.97	0.12
C15:0	0.87	0.08	0.89	0.10	1.07	0.09
C16:0	27.78	0.93	27.93	1.34	29.27	1.78
C16:1	1.53	0.12	1.55	0.17	1.47	0.17
C17:0	0.50	0.03	0.50	0.04	0.56	0.04
C18:0	11.04	0.85	10.86	1.02	10.21	1.09
C18:1,*t*6-8	0.30	0.03	0.30	0.04	0.23	0.05
C18:1,*t*9	0.29	0.02	0.28	0.04	0.21	0.04
C18:1,*t*10	0.54	0.12	0.52	0.04	0.28	0.12
C18:1,*t*11	1.47	0.19	1.45	0.26	1.71	0.54
C18:1,*t*12	0.52	0.06	0.50	0.19	0.40	0.09
C18:1,*c*9	24.38	0.93	23.96	0.08	21.44	1.12
C18:2,*c*9,*c*12	3.44	0.37	3.52	1.42	2.58	0.81
C20:0	0.09	0.01	0.09	0.55	0.10	0.01
C18:3	0.40	0.07	0.41	0.01	0.66	0.12
CLA, *c*9, *t*11	0.57	0.07	0.57	0.01	0.70	0.22
C22:0	0.04	0.01	0.04	0.07	0.06	0.01
C20:3	0.11	0.01	0.11	0.01	0.08	0.02
C20:4	0.14	0.01	0.14	0.01	0.11	0.02
C20:5	0.03	0.01	0.03	0.01	0.06	0.01
C24:0	0.03	0.01	0.03	0.01	0.05	0.01
C22:4	0.03	0.004	0.03	0.004	0.02	0.007
C22:5	0.06	0.01	0.06	0.01	0.11	0.02
C22:6	<0.001	0.32	<0.001	0.01	0.001	0.01
Other[2]	2.73	2.43	2.73	2.13	2.96	0.31
Desaturation Index[1]						
CLA, *c*9, *t*11	0.28		0.28		0.29	
16:1/16:0	0.05		0.05		0.04	
18:1/18:0	0.69		0.68		0.67	

1. Defined as [product of Δ^9 desaturase ÷ product of Δ^9 desaturase + substrate of Δ^9 desaturase]
2. CLA *t*9, *c*11 and CLA *t*10, *c*12 were found in only a small number of samples. The remaining samples contained trace amounts below the level of detection.
3. Coventionaly labeled milk: did not contain claims about supplementation with recombinant bovine somatotropin (rbST) or organic production practices. rbST-free: contain claims that cows were not supplemented with rbST. Organic: milk from farms that were certified to meet the USDA organic standards.
4. Standard deviation is being presented instead of standard error due to the inequality of variance in samples. P-values and standard error could not be presented as we are currently working with statisticians to determine the most appropriate statistical method to acquire accurate values.

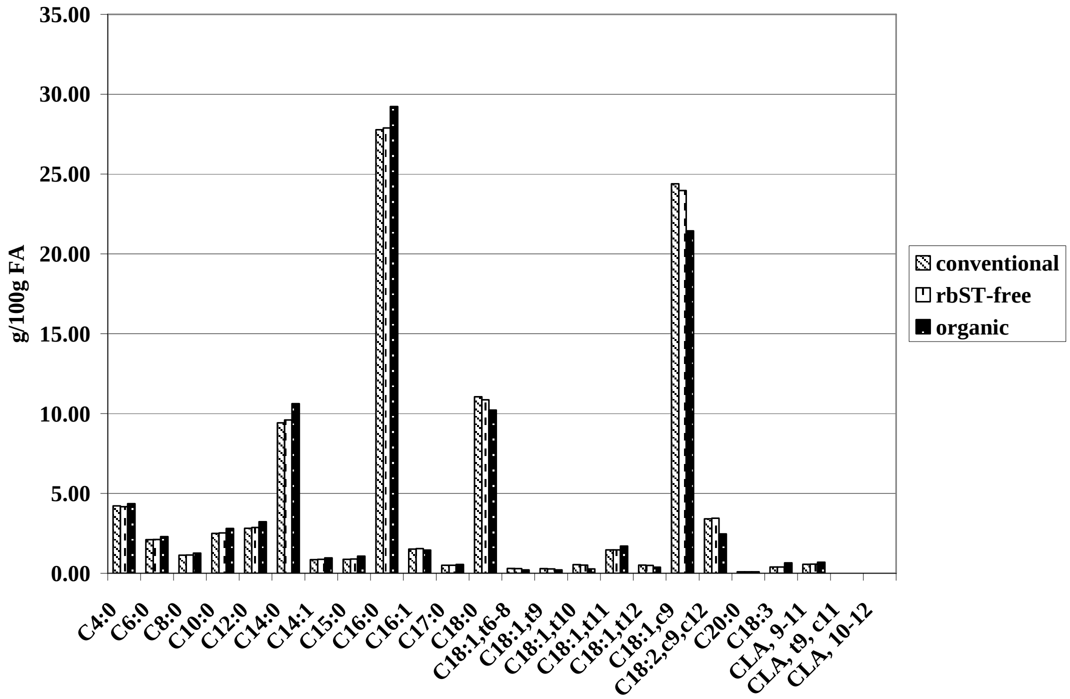

Figure 6. Fatty acid composition of milk and the effect of treatment (conventional, rbST-free, organic). Fatty acid quantity (g/100g FA) represents least-squares means.

Saturated Fatty Acids

Conventional and rbST-free milk samples did not contain statistically different quantities of saturated fat. Organic milk contained higher total saturated fat and a higher amount of lauric, myristic and palmitic fatty acids (Figure 7). These specific saturated fatty acids in particular have been implicated in increasing the risk of developing coronary heart disease when consumed by humans (Maijala, 2000) (Figure 7). Conversely, total monounsaturated and polyunsaturated fatty acids were found in lower concentrations in organic milk than in conventional and rbST-free samples (Figure 8).

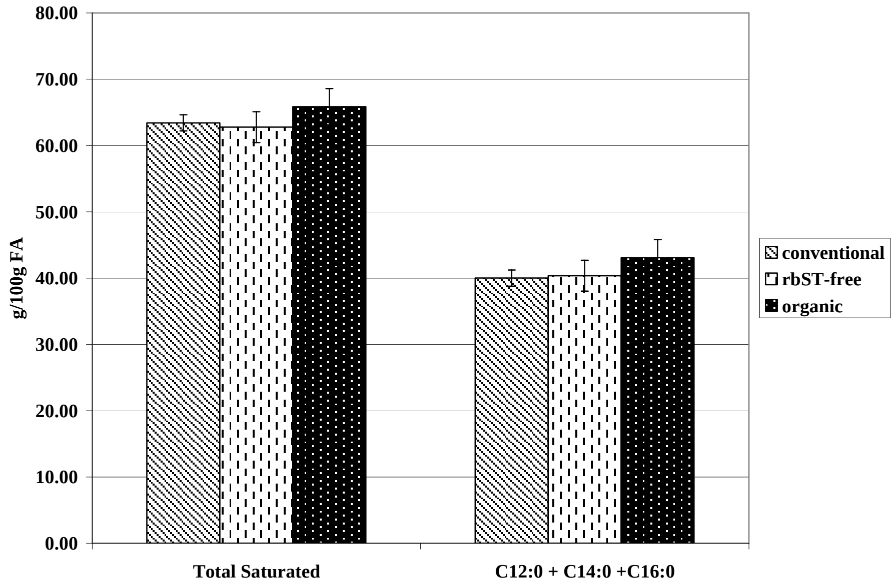

Figure 7. Comparison of total saturated fatty acids among treatments (conventional, rbST-free, organic). Saturated fatty acids included: C4, C6, C8, C10, C12, C14, C15, C16, C17 and C18. Fatty acids of health concern included: C12, C14 and C16. Fatty acid quantity (g/100g FA) represents least-squares means. Error bars represent standard deviation.

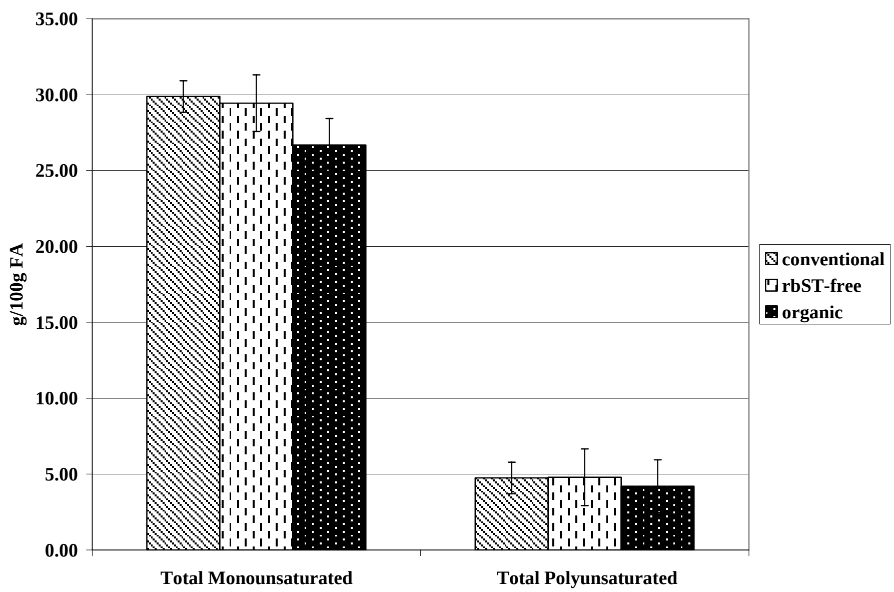

Figure 8. Comparison of total monounsaturated fatty acids among treatments (conventional, rbST-free, organic) Monounsaturated fatty acids were mainly C16:1 and C18:1. Polyunsaturated fatty acids were mainly C18:2, *c*9, *t*11, C18:3, CLA, *c*9, *t*11. Fatty acid quantity (g/100g FA) represents least-squares means. Error bars represent standard deviation.

Trans Fatty Acids

Organic milk contained lower total *trans* fat than conventional or rbST-free milk (Figure 9). Organic milk also contained a different profile of *trans* fatty acids, specifically in the *trans* 18:1 region (Figure 10). Organic milk contained significantly lower quantities of *t*6-8, *t*9, *t*10 and *t*12 and significantly higher quantities of *t*11. *Trans*-11, also referred to as vaccenic acid, is a precursor of the enzyme delta-9 desaturase for rumenic acid synthesis. Increases in vaccenic acid concentration in milk may be beneficial to human health because of the action of delta-9 desaturase in human tissue (Bauman and Lock, 2006). The concentration of rumenic acid was also found to be higher in organic milk (Figure 9).

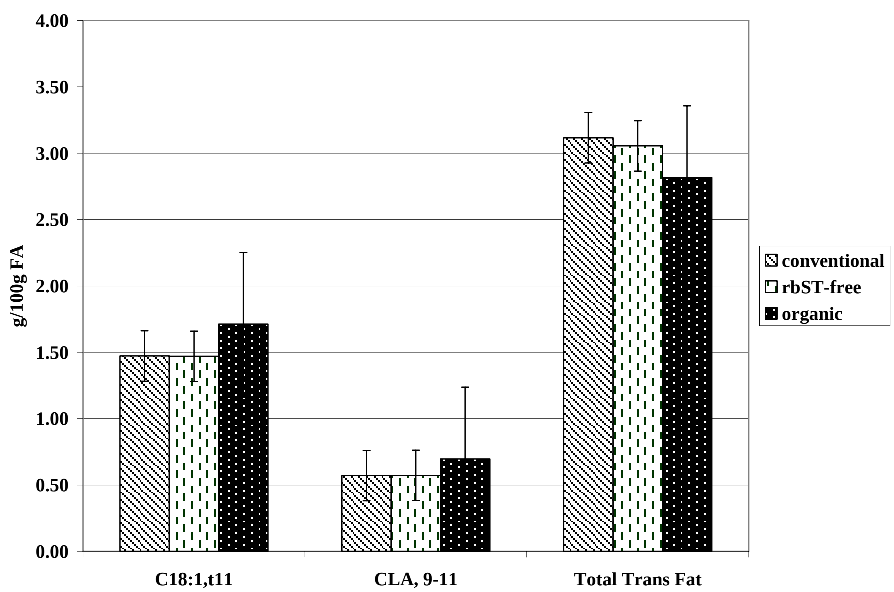

Figure 9. Comparison of vaccenic acid (C18:1, *t*11), rumenic acid (CLA, *c*9, *t*11) and total *trans* fatty acids among treatments (conventional, rbST-free, organic). Total *trans* fatty acids include: C18:1, *t*6-8, C18:1, *t*9, C18:1 *t*10, C18:1, *t*11, CLA, *c*9, *t*11. Fatty acid quantity (g/100g FA) represents least-squares means. Error bars represent standard deviation.

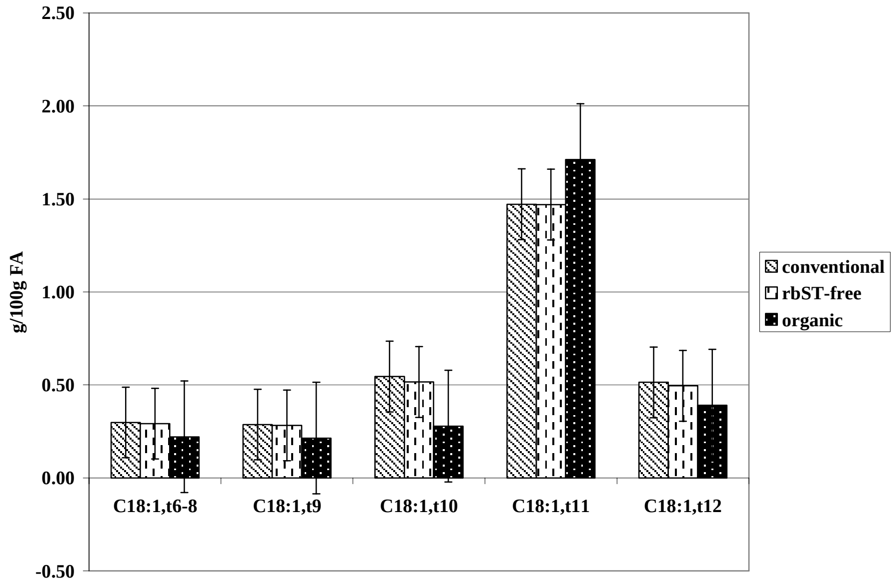

Figure 10. *Trans* fatty acid composition of milk samples and the effect of treatment (conventional, rbST-free, organic). Fatty acid quantity (g/100g FA) represent least-squares means. Error bars represent standard deviation.

It is important to note, that although organic milk may contain statistically greater quantities of vaccenic acid and rumenic acid, these differences are small and thus of little or no biological importance. This is caused by the low level of vaccenic and rumenic acid in milk (1.5% and 0.5%, respectively). Organic milk contains only 0.25% more vaccenic acid and rumenic acid than conventional or rbST-free milk. This would not significantly affect human health due to the relatively small quantity of milk consumed as part of the diet. In contrast, the manipulation of the diet of cows with the use of plant oils can result in a 6 to 10 fold increase in VA and RA (Bauman and Lock 2006).

The range of rumenic acid concentration was found to be larger in organic milk samples than conventional and rbST-free (Figure 11). As a result, the mean (0.72) was significantly higher than the median (0.67) of organic milk samples. This distribution also demonstrates the variability of rumenic acid content that can be found in organic milk. This may be a result of variation in pasture grazing as lush pasture is a dietary manipulation that will increase the milk fat concentration of RA. Pasture grazing is required by the USDA National Organic Program standards (U.S. Department of Agriculture, 2009) but the extent of grazing is not specified. Vaccenic acid distribution showed similar trends (Figure 12). Organic milk samples were more variable in VA than conventional or rbST-free samples; likewise, the mean (1.78) for VA was significantly higher than the median (1.65) of organic samples.

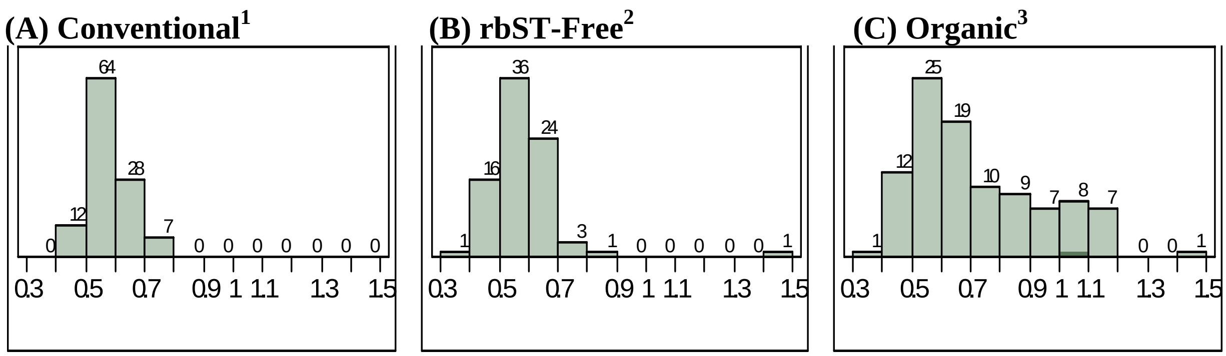

Figure 11. Distribution of CLA 9-11 (rumenic acid) in treatment types: (A) Conventional (mean = 0.57, median = 0.57) (B) rbST-Free (mean = 0.58, median = 0.57) (C) Organic (mean = 0.72, median = 0.67).
[1] Conventionally labeled milk; did not contain any claims about supplementation with recombinant bovine somatotropin (rbST) or organic production practices.
[2] rbST-free milk; processor claim that cows were not supplemented with rbST.
[3] Organic milk; from farms that were certified to meet the USDA organic standards.

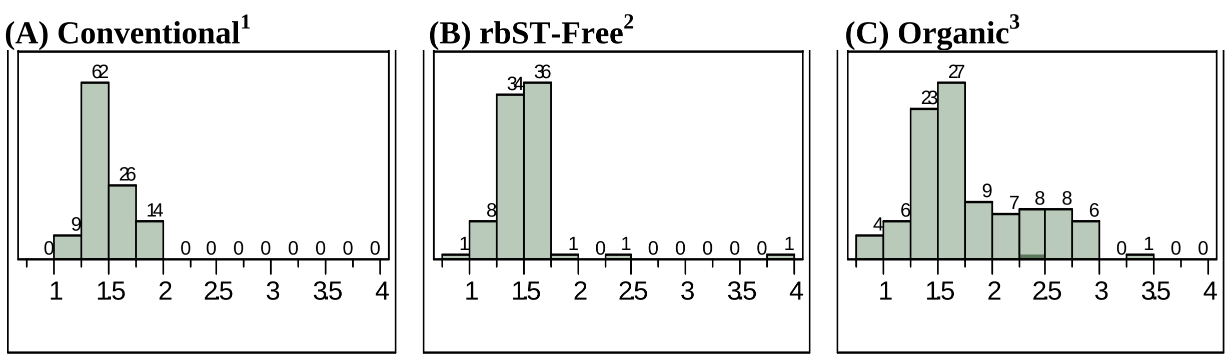

Figure 12. Distribution of C18:1 t11 (vaccenic acid) in treatment types: (A) Conventional (mean = 1.48, median = 1.44) (B) rbST-Free (mean = 1.49, median = 1.48) (C) Organic (mean = 1.78, median = 1.65)
[1] Conventionally labeled milk; did not contain any claims about supplementation with recombinant bovine somatotropin (rbST) or organic production practices.
[2] rbST-free milk; processor claim that cows were not supplemented with rbST.
[3] Organic milk; from farms that were certified to meet the USDA organic standards.

Omega-3 Fatty Acids

Omega-3 fatty acids were very low in milk averaging less than 1% of all milk fatty acids. However, organic milk contained a higher concentration of omega-3 fatty acids and a lower concentration of omega-6 fatty acids than conventional and rbST-free milk samples (Figure 12). Conventional and rbST-free omega-3 fatty acid concentrations were not found to be statistically different. This resulted in organic milk having a more favorable omega-6 to omega-3 ratio (3.7:1), close to the ideal ratio of 3:1 proposed by Simopolous (1999). However, only small quantities of EPA (~0.06% of total fatty acids) were detected and quantities of DHA were too low to be determined (<0.01% of total fatty acid). These longer chain omega-3 fatty acids are thought to confer the health benefits, such as reducing the risk of cardiovascular disease and neurologic enhancement (Lock and Bauman, 2004). Linolenic acid (0.64% of total fatty acid) accounted for a majority of the total omega-3 concentration. Due to the low conversion of alpha-linolenic acid to EPA and DHA in humans (Pawlosky et al, 2001), the greater omega-3 concentration in organic milk and the magnitude of the difference is of little or no biological consequence for human health as compared to an increase in EPA and DHA.

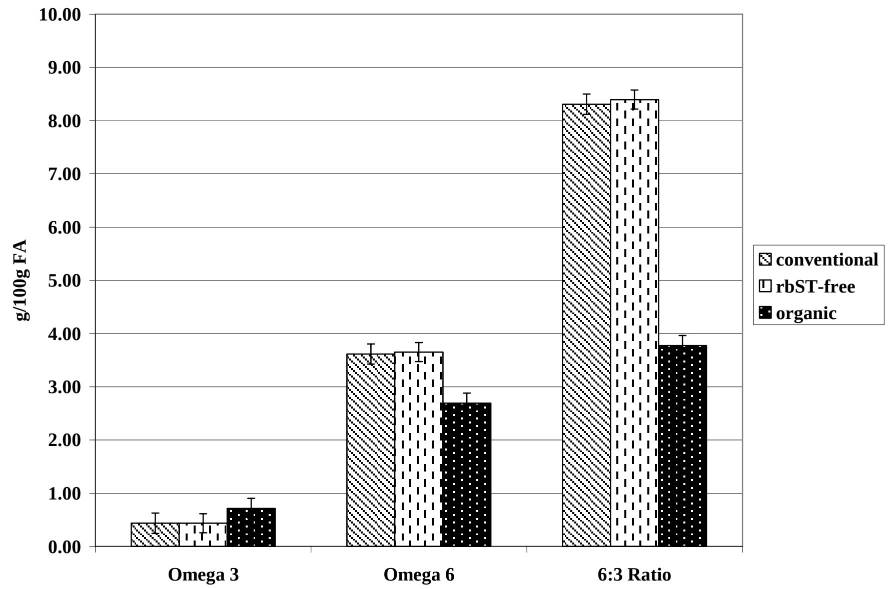

Figure 12. Comparison of omega fatty acids among treatments (conventional, rbST-free, organic); total omega-6 to omega-3 ratio among treatments. Omega-3 fatty acids included: C18:3, C20:5, C22:6. Omega-6 fatty acids included: C18:2, C20:4, C22:5. Ratio compares the summation of omega-6 to omega-3, respectively. Fatty acid quantity (g/100g FA) represents least-squares means. Error bars represent standard deviation.

Conclusion

The overall composition of milk non-labeled and labeled as “rbST-free” and “organic” was very similar. Although there were some statistically significant differences among the labeled milks for specific fatty acids, in all cases the differences were minor and therefore of little or no biological importance. All milk should be considered nutritious and beneficial to human health regardless of production label. Organic production practices or the restriction of rbST-supplementation have no biologically important impact on milk fatty acid composition.

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