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Review
Modification of Milk Fat

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Abstract. The potential health benefits associated with the intake of unsaturated fatty acids for the reduction of bad LDL cholesterol has been scientifically proven. Concentration of unsaturated fatty acids in milk and dairy products can be increased by many ways, however, many of the modification strategies do not have any significant impact on the reduction of cholesterol from milk and milk prodcuts. The concentration of unsaturated fatty acids in milk fat can also be decreased by dry fraction, interesterification, transeterification etc. Milk products with higher magnitude of unsaturated fatty acids may have significant influence on the reduction of serum cholesterol.

Keywords: fatty acid composition, cholesterol, interesterification, fractionation, milk fat

Introduction

Milk and dairy products are the regular part of the average diet of an individual, however, milk and dairy products are not the great source of beneficial unsaturated fatty acids, milk contains only 25-30% unsaturated fatty acids, the role of unsaturated fatty acids in the reduction of bad low density lipoprotien (LDL) cholesterol and enhancement of beneficial high density lipprotein (HDL)cholesterol is scientifically established (Williams, 2000). Due to the existence of lower magnitude of unsaturated fatty acids in dairy products, about 44% of the American population have started to avoid milk and milk products (Hansel *et al*., 2007). People are becoming more and more health conscious and currently, food industry is focusing on the development of functional foods.

Cardiovascular disease is the number one killer of mankind. Presently killing about 17 million people every year and mortalities are forecasted to reach beyond 25 million in the year 2020. In the USA 41.2% deaths are due to cardiovascular diseases (NCAHA, 2000). It is the biggest cause of deaths in the United Kingdom, Europe and Australia (BHF, 2005; EHN, 2005). About 21.5% of people over the age of 15 years living in the cities of Pakistan and one out of three people over 45 years suffer from hypertension (Nishter, 2002). American Heart Association and World Health Organization advised the consumers to intake unsaturated oils to decrease the risk of cardiovascular diseases (USDA, 2000). By increasing the concentration of unsaturated fatty acids, they may be used as a neutraceutical to decrease the risk of cardiovascular diseases (Sacks and Katan, 2002). Results of several investigations have disclosed that by increasing the concentration of unsaturated fatty acids, the concentration of medium chain fatty acids in milk decreases (Michael, 2007). Nadeem *et al*. (2013) reported that concentration of oleic acid in milk increased from 21 to 30%, through the manipulation in bovine feeding, the role of oleic acid in the reduction of serum cholesterol is well documented and nutritionists recommend that oleic acid must be included in the diet to reduce the incidence of cardiac diseases.

Milk fat with higher concentration of unsaturated fatty acid is susceptible to autoxidation as compared to unmodified milk fat (Gonzalez *et al*., 2003). Average temperature of Pakistan for the greater part of the year remains in the range of 35-40 °C. The exposure of fats and oils to high temperature results in oxidative and hydrolytic rancidity which not only decreases the nutritional value but also reduces the customer's acceptability (Fereidoon, 2005).

Production of higher concentrations of free radicals in the body lead to atherosclerosis, carcinogenesis, diabetes, cataract and accelerated ageing (Adedapo *et al*., 2008). The food industry has started to focus on the development of foods containing bio-active compounds for better health and prevention of fatal diseases (Dong *et al*., 2007). Recent studies have disclosed that diets containing poly phenolic antioxidants have cardiac and hepatic protective effects (Kris-Etherton *et al*., 2002). This paper describes various practicable and adaptable techniques and their efficiency for the reduction of saturated fatty acids and cholesterol from dairy and dairy products.

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Saturated fatty acids. Saturated fatty acids do not possess any double bond in the carbon-carbon chain. On the basis of chain length, fatty acids are usually divided into three groups; short-chain fatty acids C4-C10, medium chain fatty acids C12-C16, long chain fatty acids C18 and above. The concentration of short-chain and medium chain fatty acids in milk ranges from 10.6-12.8 and 42.4-44.0% on weight basis (Ghatak and Bandyoupadhayay, 2007). Palmitic, myristic and lauric acids are the major fatty acids among the category of saturated fatty acids promoting atherosclerosis (Michael, 2007).

Unsaturated fatty acids. Unsaturated fatty acids have double bonds in the carbon-carbon chain. Milk fat is characterised with higher concentration of oleic acid (C18:1) which accounts for about 22-25%. Oleic acid is relatively more stable towards autoxidation than linoleic acid and does not pose any threat for the beneficial HDL-cholesterol; polyunsaturated fatty acids decrease the harmful LDL-cholesterol. Nutritionists advise that the contribution of calories from fats and oils in the energy regime should not exceed 30% of the total requirement; also the contribution of saturated fatty acids should be less than 10%. American Heart Association guidelines suggest that the diet should contain about 15% oleic acid to decrease the risk of cardiovascular diseases (Aigster *et al*., 2000).

***Trans* fatty acids.** *Trans* fatty acids are produced during bio-hydrogenation of unsaturated fatty acids in the rumen by the rumen micro flora. Octadecenoic acid (*trans*-C18:1) is the most abundant form of *trans* fatty acids in the milk. The negative impact of *trans* fatty acids on human health is two times greater than saturated fatty acids; they decrease HDL and increase LDL. Cis-9, *trans*-11 (rumenic acid) belongs to the category of conjugated linoleic acid which possesses anti-carcinogenic, anti-diabetic and immuno modulating characteristics (Bessa *et al*., 2000). Conjugated linoleic acid (CLA) possess anticarcinogenic, immunomodulating, antidiabetic and antiatherogenic properties (Dhiman *et al*., 2000). Intake of about 3 g CLA on daily basis can prevent carcinogenesis (Baer *et al*., 2001).

Reducing saturated fatty acids by rumen protected feeds. Rumen contains a countless number of micro-organisms which convert unsaturated fatty acids into saturated and *Trans* fatty acids. Many strategies have been developed to protect the useful unsaturated fatty acids from rumen bio-hydrogenation. Given below is the summary of work conducted by the researchers to modify the fatty acid composition of milk by using rumen protected fats. Fatty acids in milk fat are derived from two major sources which are listed as:

Mammary gland synthesis. The fatty acids which are produced in the mammary glands are known as *de novo* fatty acids C4-C16 (Givens and Shingfield, 2006).

Fatty acids through diet. Long chain fatty acids are not synthesised by the mammary tissues but supplied from the blood stream, they are originated from the diet and therefore their content in milk can be modified by manipulating the rumen diets (Givens and Shingfield, 2006).

Oil seeds. Ashes *et al*. (1992) studied the effect of formaldehyde treated canola seeds on the fatty acid composition of cow milk. Feeding rumen protected canola seeds considerably reduced the concentration of saturated fatty acids in milk fat; C18:1, C18:2, and C18:3 were also higher in the milk. Feeding rumen protected soybean seeds significantly modified the fatty acid composition of the milk; linoleic acid in the milk of the treated group was 65% higher over the control. Supplementation of the feed with formaldehyde treated oil seeds at 14% level decreased the concentration of palmitic acid by 15% and increased linoleic and linolenic acids by 76% and 123%, respectively in the milk of Holstein-Friesian cows (Tymchuk *et al*., 1998). Feeding sunflower and flax seed supplemented diets to ewes for 42 days, significantly decreased the concentration of short and medium chain fatty acids, the concentration of CLA and C18:3 in the milk increased by 140 and 63% (Goodridge *et al*., 2001).

Calcium salts of fatty acids. The fatty acid profile of milk fat is governed by the genetic, environmental and feeding practices. Brzoska and Sala (2001) studied the effect of calcium salt of fatty acids on cholesterol content and fatty acid profile of cows, the cholesterol content of milk was not significantly different from the control, while, concentration of unsaturated fatty acids were significantly ($P<0.05$) increased in the milk. The effect of feeding calcium salts of long chain fatty acids of soybean oil on fatty acid composition of cow milk investigated, C18:1 and C18:2 were increased by 35.7% and 8.9%, respectively. The CLA content of milk was also higher than the control (Mandebvu *et al*., 2003). Lacasse *et al*. (2002) described that unsaturated fatty acids have a negative impact on rumen micro flora which tended to decrease the digestion of fibre, lower the ratio of acetic acid to propionic acid. Supplementation of calcium salts of fatty acids increased the fat content of milk ($P<0.05$), while protein content decreased ($P>0.05$) and milk production was greater than the control. The fatty acid profile of the milk revealed that the addition of calcium salts of fatty acids had a great effect on fatty acid composition (Gargouri *et al*., 2006). Feeding calcium salts of fatty acids to Holstein-Friesian cows increased the concentration of C18:1 from 22.5% to 30.2% (Juchem *et al*., 2008). Nadeem *et al*. (2013) studied the impact of feeding calcium salts of soybean oil fatty acids on fatty acid profile of Sahiwal cows. Calicum salts of soybean oil fatty acids were offered at 5% and 10% in an iso-nitrogenous feed. Fatty acid composition was determined after 2 weeks, gas chromatographic (GC) analysis showed that feeding calcium salts of soybean oil fatty acids at both concentrations considerably increased the concentration of beneficial unsaturated fatty acids and conjugated linoleic acid (oleic and linoleic).

Enhancing unsaturated fatty acids by fractionation. Effect of increasing saturated fatty acids of milk fat by fractionation has been studied by many earlier researchers. Reddy (2010) fractionated the milk fat by dry crystallization technique into olein, stearin and mid fraction, fatty acid composition of all the fractions was significantly different from the parent milk fat. Shea *et al*. (2000) fractionated milk fat from 63 to10 °C in a carefully controlled temperature programme and found that olein fraction contained 63.2% more conjugated linoleic acid, also enriched with polyunsaturated fatty acids over the native milk fat. Fractionation of milk fat is a good tool to increase the concentration of conjugated linoleic acid in the milk. Up to 89% conjugated linoleic acid increased in the fractionated milk fat (Romero *et al*., 2000).

Improving functionality of milk fat by fractionation. Milk fat has low functional properties due to its natural fatty acid composition and melting characteristics. The use of milk fat is restricted to the manufacturing of butter, butter oil and some bakery products. Fractionation process not only improves the fatty acid profile of milk fat but also enhance the functional properties of milk fat. Fractionated milk fat can be used in the formulation of cookies, cheese, pastries etc. Several companies offer milk fat fractions as functional food ingredients for bakery, confectionery, chocolate and dairy industries. High-melting milk fat fractions are used in laminated pastries to promote layering and in the chocolate industry to inhibit fat blooming. Low-melting milk fat fractions can be used for creaming in the biscuits and cheese industries (Gibon, 2006; Deffense, 2002). Reddy (2010) fractionated the milk fat into stearin and olein fractions to improve the functional properties of milk fat, stearin fraction revealed higher solid fat index with improved plasticity. Physical properties, melting profiles and solidification properties of stearin were similar to commercial bakery shortenings and vanaspati. Olein fraction contained relatively higher concentrations of monounsaturated fatty acids, which can be successfully used as a cooking fat or as a salad oil for having lower solid fat index as compared to original milk fat. Bazmi and Relkin (2009) studied the effect of using milk fractions rich in unsaturated fatty acids on some quality characteristics of ice cream emulsions, formulated according to the typical ice cream formulation by using anhydrous milk fat alone or in combination with low melting fractions of milk fat at different concentrations, enrichment of milk fat with olein fraction increased the whipping ability of the emulsions.

Fat modification by interesterification and transesterification. For the modification of the physical properties of butter and canola oil blends, 1, 3 specific lipases (*Rhizopus arrhizus***)** were employed. The first blend was comprised of 60% butter and 40% canola oil and the second blend contained 60% canola oil and 40% butter. Solid fat index of both the blends were less than the control and the lowest value was observed between 5 and 10. However, both the blends were characterised for higher concentration of unsaturated fatty acids (Rousseau and Marangoni, 1998). In another attempt to incorporate unsaturated fatty acids in the table margarine, the blends of butter fat and corn oil were chemically interesterified, the interesterified blends showed better spread ability, interesterification significantly decreased the trisaturated glycerides, improved the omega-6 fatty acids in the interesterified blends (Ract and Gioielli, 2003). Functional properties, fatty acid composition and physical properties of the transesterified blends of anhydrous milk fat, linseed oil and rape seed oil blends were significantly different from the starting materials (Aguedo *et al*., 2008).

Dairy products with higher concentration of unsaturated fatty acids. The milk with higher content of unsaturated fatty acids was turned into butter; solid fat index of the butter over a temperature range of 10-38 °C was lower in the samples. Sensory characteristics were not different from the control (Lin *et al*., 1996). Texture of butter with modified fatty acid composition was softer as compared to the control; panel of trained judges was unable to detect any flavour difference (Ramaswamy *et al*., 2001). Flavour quality of ultra-high temperature treated (UHT) milk, ice cream and butter with modified fatty acid composition was similar to the control, milk with modified fatty acid composition developed slight oxidized flavour (Avramis *et al*., 2003). Lynch *et al*. (2004) studied the effect of CLA fortification on oxidative stability of homogenised milk packed in plastic bottles and stored at 4 °C, non-significant changes were observed in the fatty acid profile of milk at zero and 14 days of analysis. Sensory evaluation by a panel of untrained judges did not reveal any flavour difference between the control and treated stuff. Jones *et al*. (2005) studied the effect of fortification of this-9, *trans*-11 on physical-chemical and sensory characteristics of some dairy products. Milk with higher content of CLA was turned into UHT milk, butter and cheese, most of the sensory characteristics of dairy products with altered fatty acid composition were similar to the unmodified milk however, the texture of butter with modified fatty acid profile was relatively soft.

Effect of modified fats on blood cholesterol level. Feeding modified fats to hypercholestrolemic adults reduced the blood cholesterol up to 95% as compared to those who consumed unmodified milk fat (Davis *et al*., 1993). The subjects who consumed fat-modified dairy products had significantly low total and LDL-cholesterol in plasma, representing a 9% reduction in the risk of developing coronary heart disease (Jacques *et al*., 1999). Feeding modified bovine butter fat decreased LDL and total cholesterol from 78-80% (Poppitt *et al*., 2002). LDL cholesterol decreased in the hyper-cholesterolemic subjects when milk-fat-based cheese was replaced with rape seed oil based cheese in the diet (Karvonen *et al*., 2002).

Effect of modified fatty acid composition on fat oxidation. Lipid oxidation is a chemical reaction which takes place at the double bond site of unsaturated fatty acids as a result of free radical mechanism (Fox and McSweeney, 1998). Oxidation may be different types, for example thermal oxidation, autoxidation and enzymatic oxidation (hydrolytic rancidity), but the autoxidation of unsaturated fatty acids is more important for nutritional and keeping quality viewpoints. Oxidation is catalyzed by the metal ions, especially copper, iron, oxygen, temperature, moisture fatty acid profile and the presence of fat hydrolysing enzymes (Gonzalez *et al*., 2003). Milk with modified fatty acid composition is susceptible to oxidation (Focant *et al*., 1998; I'm and Marshall, 1998; Ashes *et al*., 1997).

Oxidised milk may transfer off-flavour to the products which are made from it (Abd El-Rahman *et al*., 1997). Fats having higher content of monounsaturated fatty acids are less vulnerable to autoxidation than those having higher extents of polyunsaturated fatty acids.

Natural antioxidants for the enhancement of keeping quality of milk fat with modified fatty acid composition. The oxidation of milk fat products is accelerated by the catalytic activity of temperature. The exposure of fats and oils to high temperature results in oxidative rancidity which not only decreases the nutritional value but also reduces the customer's acceptability (Potter and Hotchkiss, 1995). Consumption of rancid fats promotes many types of diseases such as cardiovascular disease (McSweeney and Fox, 2003). The quality assessment of butter sold in Lahore (2^{nd} largest city of Pakistan) showed higher concentrations of oxidation products (Hussain *et al*., 2011). More than 5000 natural antioxidants have been isolated from various plant sources (Dong *et al*., 2007; Kris-Etherton *et al*., 2002). These antioxidants have anti-carcinogenic, anti-inflammatory, antimicrobial, cardiac and hepatic protective activity. The diets rich in phenolic antioxidants have many health benefits and confer longer life expectancy Kris-Etherton *et al*. (2002). Mohdaly *et al*. (2011) stabilised the sunflower oil by sesame cake extract and found that the concentration of total phenolic content in the cake extract was 1.94% on dry weight basis. Canola hull also contains some phenolic substances but the concentration was much less than *Moringa oleifera* (Naczk and Shahidi, 1998). The free radical scavenging activity of *M. oleifera* oil at 200 µL was 70.15% (Ogbunugafor *et al*., 2011).

Conclusion

Fatty acid composition of milk fat can be significantly modified by manipulation in the diet of dairy animals, fractionation, interesterification and transesterification of milk fat with liquid vegetable oils. Structured lipids have better nutritional and physical characteristics owing to the incorporation of new fatty acids or to alter the current fatty acids on the glycerol molecule. Functional properties of milk fat can be considerably improved through dry and solvent fractionation. Stearin fraction of milk fat can be used as bakery shortening. Oxidative stability of modified version of milk fat can be improved through natural antioxidants.

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