EFFECT OF FEEDING DIFFERENT CONCENTRATE TO AYRSHIRE AND FRIESIAN COWS ON MILK YIELD AND FATTY ACID COMPOSITION OF BUTTER FAT

BY

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ABSTRACT

The effect of breed and different oil seed cake supplementation on milk yield, composition and milk fat fatty acid (MFFA) composition was studied using 12 Friesian and 12 Ayrshire lactating cows. Control diet contained maize bran (MB) plus minerals. For diet I and II, some of the MB in the control diet was replaced with cotton seed cake (CSC) (34.8%) and sun flower seed cake (SSC) (44.5%) respectively. Milk samples were collected for chemical composition analysis and butter making. The composition of fatty acid methyl esters (FAMEs) were analysed by gas chromatograph (GC). The data was analysed using Statistical Analysis System (SAS). The mean milk yield was significantly ($P<0.05$) higher in Friesian cows (8.36 kg/day) compared to Ayrshire breed (5.85 kg/day). The proportions of short chain fatty acids (SCFAs) were slightly higher ($P>0.05$) for Ayrshire breed (4.94%) compared to Friesian (4.34%). The proportion of medium chain fatty acids (MCFAs) for Friesian (12.73%) was slightly lower ($P>0.05$) whilst those of long chain fatty acids (LCFAs) (82.95%) were slightly higher ($P>0.05$) than those of Ayrshire (14.61 and 80.44) % respectively. The proportion of unsaturated fatty acids (UFAs) was slightly higher for Friesian (57.52%) than Ayrshire (46.03%) and for those fed CSC (57.42%) was slightly higher than those fed SSC (50.80%) and control diet (43.8%). The melting point (MP) range of milk butter fat (BF) in Friesian cows (39.5–41.5 ºC) and (39.5–41.0 ºC) was slightly higher compared to that of Ayrshire (39.5–41.0 ºC) and (39.5–40.5 ºC) when supplemented with CSC and MB diet respectively. It is concluded that both breed and oil seed cake supplements have no significant influence on the fatty acids (FAs) composition of BF.

DECLARATION

I, YUSTINA TESHA, do hereby declare to the Senate of Sokoine University of Agriculture that, this dissertation is my own original work and has never been nor concurrently been submitted for a higher degree awards in any other University.

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This Manuscript is dedicated to my husband Dr. Clemence Tesha, my late father Mathias Sekao Shau, my mother Philomena Mchaumbi Msoka and all my children Philip, Jovita, George and Scholarstica.

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LIST OF ABBREVIATION

AOAC	Association of Official Analytical Chemist
ADF	Acid Detergent Fiber
ASARECA	Association for Strengthening Agricultural Research in Eastern and Central Africa.
βHBA	β-hydroxybutyrate acid
BF	Butter Fat
CF	Crude Fiber
CLA	Conjugated Linoleic Acid
CoA	Co-enzyme A
CP	Crude Protein
CSC	Cotton Seed Cake
DASP	Department of Animal Science and Production
DCP	Digestible Crude Protein
DHA	Docosahexaenoic Acid
DM	Dry Matter
DMI	Dry Matter Intake
EE	Ether Extract
EFAs	Essential Fatty Acids
EPA	Ecosapentaenoic Acid
FA	Fatty Acid
FAs	Fatty Acids
FAME	Fatty Acid Methyl Ester
FCM	Fat Corrected Milk
GC	Gas Chromatography
GLM	General Linear Model
H	High
HDL	High Density Lipoprotein
IDF	International Dairy Federation
ILCA	International Livestock Centre for Africa
ISO	International Standard of Organization
LCFA	Long Chain Fatty Acids
LDL	Low Density Lipoprotein
LR	Lactometer Reading
M	Maintenance
MB	Maize Bran
MCFA	Medium Chain Fatty Acids
ME	Metabolisable Energy
MFFAs	Milk Fat Fatty Acids
MH	Maintenance High
MJ	Mega Joules
MP	Melting Point
MUFAs	Mono Unsaturated Fatty Acids
N	Nitrogen
NDF	Neutral Detergent Fiber

NFE	Nitrogen Free Extract
NPN	Non-Protein Nitrogen
NRC	National Research Council
PEG	Polyethylene Glycol
pH	Milk Acidity or Alkalinity
ppm	Part Per Million
PCSk*9*	Protein Convertase Subtilism/kexin type *9* series Protease Gene
PUFAs	Polyunsaturated Fatty Acids
RLDC	Rural Livelihood Development Company
SAS	Statistical Analysis System
SCFAs	Short Chain Fatty Acids
SFAs	Saturated Fatty Acids
SNF	Solid Not Fat
SSC	Sunflower Seed Cake
SUA	Sokoine University of Agriculture
TAWLAE	Tanzania Association of Women Leaders in Agriculture and Environment
TS	Total Solid
UD	University of Dar es Salaam
UFAs	Unsaturated Fatty Acids
VLDL	Very Low Density Lipoproteins

CHAPTER ONE

1.0 INTRODUCTION

Milk and milk products are regarded as highly nourishing foods that provide a large part of our dietary needs and are particularly important for children (Bascom, 2002). Milk is an important source of protein, minerals and vitamins in the human diet (Castle and Watkins, 1984). The fat content is considered as a measure of milk quality since it serves nutritionally as an energy source and supplies essential fatty acids (EFAs) (Frank, 1986).

Milk as a food, is widely consumed in many different food products but there has been a growing tendency for consumers to prefer ‘light’ milk rather than ‘full cream’ milk. This may actually be bad for very young children who require a greater proportion of fats in their diet. Nevertheless some dieticians have expressed concerns about some aspects of milk. The main issue seems to be the presence of significant amounts of saturated fats in milk and fats with *trans*-fatty acids. Understanding the responses of milk fat fatty acids (MFFA) composition to nutritional practices applied to dairy cows is important to the development of predictive models of functional characteristics of milk fat. Manipulation of the diet of cows, particularly the amount of supplemental fat, can have marked effects on the fatty acid (FA) profile of milk fat (Grummer, 1991; Palmquist *et al*., 1993). The FA composition of milk fat is an important variable that affects both the nutritional value of dairy products and processing characteristics (Berner, 1993). For example, increases of oleic acid (cis-C18:1) is generally associated with butter that is softer at refrigeration temperature (Banks *et al.,* 1989). The FA composition, through its influence on melting

behaviour, affects the churning time necessary to produce butter (Banks *et al*., 1989). Ruminant milk fat has abundant short- and medium-chain length fatty acids (FAs), which evolved as a compensation for the large amount of saturated fatty acids (SFAs) arising from ruminal biohydrogenation. Studies from rodents' shows that short fatty acyl chains are cleaved from the FA synthetase enzyme complex by thioesterase II. Although both rodents and ruminants contain thioesterase-I, specific for C16 FA, there is no thioesterase-II in ruminants; rather chain length seems to be determined by a multiple complex which includes the rate of chain-lengthening (largely determined by the supply of malonyl-Coenzyme A (CoA) and competition between enzymes that load and unload the acyl chain from the FA synthetase complex (Knudsen and Grunnet, 1982). Modification of lipid by rumen bacteria results in the formation of many odd-carbon and branched-chain FA probably through incorporation of propionyl-CoA into Carbon-skeleton (Wahle and Peterson, 1979). The major important products of hydrogenation that might pass to lower tract to be absorbed and incorporated into milk are trans-11- octadecenoic acid. The cream flavour compounds, 4-cis-heptenal, has been identified as arising from autoxidation of milk fat (Webb *et al*., 1974).

There seems to be some compensation among groups of FA in milk fat, so that the melting point (MP) of the fat remains in a range that keeps it liquid at body temperature of the cow (39°C). Research with Jersey cows show higher proportion of short chain fatty acids (SCFAs) in milk than those of milk from Holstein cows which would decrease the MP; however, the oleic acid (C18:1) content is lower and stearic acid (C18:0) is higher in Jersey cows (Beaulieu and Palmquist, 1995 and Drackley *et*

al., 2001). An interaction of breed and dietary fat indicated that the content of cis-C18:1 in milk fat was increased more by supplemental fat in Holsteins than in Jerseys. This indicates that Jersey cows have lower activity of stearoyl-CoA desaturase, causing the milk to have higher MP and harder milk fat than other breeds (Drackley *et al*., 2001).

In the present study two hypotheses were tested. First, whether feeding cows concentrates containing different oil seed cake supplements will or will not affect milk fat composition in all cows. Second, if FA composition of milk fat from Friesian and Ayrshire cows receiving similar diets will be different or not. In developing countries like Tanzania there is limited information concerning milk quality. Past dairy cattle research concentrated on milk yield. So there is a need for dairy cattle research to address both parameters, which is quality and quantity of milk and milk products from Friesian and Ayrshire cows. The present study focus on two breeds, Friesian and Ayrshire cows which were available in Magadu farm and University farm at Sokoine University of Agriculture (SUA) and also due to constrain of budget only two oil seed cakes, cotton seed cake (CSC) and sunflower seed cake (SSC) that are commonly used by dairy farmers were used in the study. Of the oil seed cake produced and processed for local utilization in Tanzania, CSC is the largest in quantity followed by SSC (Nandonde, 2008).

Milk fat contains about 70% saturated fatty acids (SFAs), 25% mono-unsaturated fatty acids (MUFAs) and 5% poly unsaturated fatty acids (PUFAs). The higher content of SFAs in milk fat raised concern among heath conscious consumers

because of the purported relationship of SFAs with atherosclerosis. Butter and other high-fat dairy products are excluded from the diets designed to decrease blood cholesterol and prevent or treat coronary heart diseases (Ney, 1991; Bauman and Lock, 2004). Some people are avoiding consuming milk with the expectation of avoiding high level of cholesterol and FAs which may be detrimental to their health. The quality of butter, especially its MP and susceptibility to oxidative deterioration which is influenced by several factors among of which is its FA composition, is important from the consumer point of view. Therefore, there is need to investigate differences in FA composition that may be caused by genetic as well as environmental factors such as feeding of oil seed cakes supplement.

The present study was carried out to investigate differences in milk fat composition that may be caused by genetic as well as environmental factors such as feeding of oil seed cakes supplement. Therefore the specific objectives of the study were to study the effect of feeding different oil seed cakes supplement on milk yield and milk composition, FA composition and quality of butter fat (BF); and to compare FA composition and MP of BF of milk from Friesian and Ayrshire cattle breeds fed similar diets.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Milk Composition

The role of milk in nature is to nourish and provide immunological protection for the mammalian young. Milk has been found to have many positive effects to human health and it has been used as food for humans since prehistoric times (Bauman, 2004). The average gross composition of cows milk is as follows: 87.3% water (range of 85.5% - 88.7%), 3.9% milk fat or milk lipids or BF (range of 2.4 - 5.5%), 8.8% solids-not-fat (SNF) (range of 7.9 - 10.0%), protein 3.25% (0.75% casein), lactose 4.6%, minerals 0.65% - (Ca, P, citrate, Mg, K, Na, Zn, Cl, Fe, Cu) sulfate, bicarbonate, acids 0.18% - citrate, formate, acetate, lactate, oxalate; enzymes - peroxidase, catalase, phosphatase, lipase; gases - oxygen, nitrogen; vitamins - A, C, D, thiamine, and riboflavin (Watt and Merrill, 1963; Nangwala, 1996).

Report from O'Donnel (1989) and La Count *et al*. (1998) shows that ideal milk fat for use in the human diet should contain <10% PUFAs, <8% SFAs and >82% MUFAs. Typically, milk from cows fed diet based on grass silage contains approximately 40 g fat/l, of which up to 97% is in the form of triacylglycerol and the remainder as monoacylglycerol and free FAs. Although the FA content of milk is one of the most complex found in nature, with more than 500 different FAs identified there are usually only considerable amounts of 12 to 15 of these in any single fat (Hermansen, 1995). The predominant FAs in milk are the long chain fatty acids (LCFA), myristic (14:0), palmitic (16:0) and stearic (18**:**0). These SFA account for 75 g/l00 g total FAs, with a further 21 g/l00 g occurring as MUFA of which the most

prevalent is oleic acid (18:1). Only 4 g/l00 g milk FAs are PUFA occurring mainly as Linoleic (18**:**2) and linolenic (18:3**)** acids.

Table 1: Milk fat from different mammalian species (g/100 g fresh milk)

Species	Fat (g)
Cow	3.70 – 3.00
Human	4.20
Water Buffalo	9.00
Goat	3.80
Donkey	0.60
Elephant	5.00
Monkey rhesus	4.00
Mouse	13.10
Whale	42.30
Seal	49.40

Source: Webb *et al.* (1974)

2.2 Factors Affecting Milk Composition

Milk composition can be affected by a number of factors which include environment, breed, parity, lactation number and nutrition (Grummer, 1991; Beaulieu and Palmquist, 1995). Yield of milk and milk fat increases with fat supplementation (Avila *et al*., 2000)

2.2.1 Effect of breed

The composition of milk varies with a number of non- nutritional factors including breed (Caroll *et al*., 2006). Friesian, the breed with highest yield potentials has the lowest fat whereas the lower yielding Jersey and Guernsey have the highest fat contents (O'Mahony, 1986). Morales *et al*. (2000) showed that breed and fat source interaction had higher effect on Friesian than Jersey cows when dietary fat was changed. Zebu cows' milk fat can be as higher as 7%. The strain and genetic

constitutient of cows within a breed have a large effect on milk composition. An average value from many thousands of analyses show that, Ayrshire breed has higher fat content than Friesian (Holstein) breed (Table 2). The poorer milk quality from specific herd of one breed compared with another herd of a different breed can be due to individual breeding and management. Friesian breed consumed more dry matter (DM) and yielded more fat corrected milk (FCM) than Jersey cows but percentages of fat were higher in milk yielded by Jersey cows (Beaulieu and Palmquist, 1995). Milk fat from Jersey cows contained a higher proportion of SCFAs and medium chain fatty acids (MCFAs) and lower proportions of palmitic and oleic acid than in Holstein milk fat (Beauleu and Palmquist, 1995; Drackley *et al*., 2001).

Table 2: Milk fat percentages for various breed

Breed	%Fat
Ayrshire	3.64
Brown swiss	3.66
Guernsey	4.21
Holstein	3.48
Jersey	4.44
Shorthorn	3.42

Source: Neitz and Robertson (1991)

2.2.2 Intervals between milking and milking frequency

The fat content of milk vary considerably between the morning and evening because there is usually a much shorter interval between the morning and evening milking than between the evening and morning milking (O'Mahony, 1986). The same situation was observed in the study using Murrah buffaloes which showed 31% more milk and 26% more BF that resulted from milking three times per day as compared

to twice a day. Milking frequency affects both total milk and fat yield. Milking twice a day yields at least 40% more milk than once a day (Hogberg and Lind, 2003).

2.2.3 Stage of lactation and season

Milk fat decreases as milk yield increases to a peak at about two months after calving thereafter, increases slowly during the remainder of the lactation (Castle and Watkins, 1984; O'Mahony, 1986). The fat percentage varies with stage of lactation and with milk yield (Hogberg and Lind, 2003: Bailey and Henrichs, 2005). A study on Nili-Ravi buffaloes in Pakistan showed that the fat percentage increased steadily from 5.5% in the first month of lactation to 7.5% in the ten month of lactation (Hogberg and Lind, 2003; Bailey and Henrichs, 2005). Seasonal effects tend to be smaller than the lactational effect. Fat content is highest in October in the northern hemi sphere and then falls steadily to a minimum in June (Bailey and Henrichs, 2005; Hogberg and Lind, 2003).

A study conducted by Sargeant *et al.* (1998) indicated lowest protein and fat percentages during the summer months (June - August), and the highest percentage between October and December in the Northern Hemisphere. During the summer season, Ozrenk and Inei (2008) suggested strategies for reducing the impact of high heat loads on lactating cows such as shading, increased ventilation, changing the time of feeding to the late afternoon and an appropriate administration of mineral elements with the feed rations as a necessary strategies to prevent deterioration of cheese yield and quality.

Nutrition can be regarded as one of the most important sources of variation in the yield and composition of milk but climatic conditions, seasonal variation and regional differences can also play an important role (Ozrenk and Inei, 2008). The quality and composition of the milk are of the most importance to the dairy industry and human health because milk composition is related to milk process ability. When the milk has higher protein content it should be processed to cheese. If it has higher level of fat, then it should be used to produce butter (Ozrenk and Inei, 2008).

2.2.4 Effect of type of food (nutrition) on milk composition

Of all milk components, milk fat is influenced most by dietary manipulations. Partial replacement of cereal grains with fat supplements, which have a higher concentration of energy than starch-based supplements, is a feeding strategy to increase energy intake and LCFAs in milk fat of grazing cows (Agenäs *et al*., 2002). Changes in milk composition due to dietary manipulation are related to changes in ruminal acetate: propionate ratio (van Soest, 1994). When propionate: acetate ratio in the rumen is increased, the energy efficiency in the rumen also increased resulting in high milk and milk fat production (van Soest, 1994). Milk fat percentages tends to decline as the proportion of the concentrate in the ration increases above 50 to 60% of the ration due to lower ruminal production of acetate and butyrate precursors that are for milk FA synthesis in the mammary glands (Bauman and Griinari, 2001; 2003).

O'Mahony (1986) observed milk fat depression syndrome i.e drop of milk fat below 3% in lactating dairy cows when fed finely chopped forages. The drop in milk fat content was attributed to animals spending less time in chewing and therefore

production of less saliva hence ruminal pH drop due to reduced buffer effect in acid production in the rumen (Grant and Kubik, 1990; Daniel *et al*., 2003). As the ruminal pH drops below 6, the activity of the cellulolytic bacteria is reduced and so there is a reduced production of acetic and butyric acids precursors that are for SCFA synthesis in the mammary glands. CSC is rich in linoleic and other unsaturated fatty acids (UFAs), which adversely affect ruminal fermentation and milk production (Chouinard *et al*., 2001). Diets rich in linoleic acid often increase the formation of trans- FAs in the rumen, which can decrease *de novo* FAs synthesis in the mammary gland (Chouinard *et al*., 2001; Bauman and Griinari, 2003).

2.2.5 Effect of energy and protein

Samuelson (1996) mentioned that cows on a high plane of nutrition generally have a reduced fat content in the milk whereas cows on low energy intake have an increased fat content. A depression in milk fat content has been reported in cows fed diets containing high amounts of grain (fermentable starch) (Jenkins, 1993; Palmquist *et al*., 1993). When comparing four treatments with low (L), maintenance (M), maintenance high (MH) and high dietary energy concentration (H), fed to primiparous beef heifers, Lalman *et al.* (2000) found that increasing dietary energy led to increased milk fat percentage, however this effect was apparently seen in the L, M and MH treatments, but not in high energy concentration as the lowest milk fat percentage was found in the high treatment 3.5, 3.8, 3.7% and 3.3% respectively. Lowman *et al.* (1979) working with beef cows reported that increasing the energy allowance significantly increased milk yield but did not affect milk composition. The effect of dietary energy on milk composition vary depending on the source

(ingredients) of energy used in the cow's diet and other dietary factors like effective fiber (chewing time), particle size and additives (buffers) (Emery, 1988; Jelec, 1990). Study by Spain *et al.* (1984) showed that there is no effect on milk protein percentages when feeding dairy cows with low or high protein diet (soybean meal, fish meal or corn gluteal meal). Milk protein yield increases when cows are fed diet with high soy bean meal and fish meal than when fed corn gluteal meal. The increase in milk protein yield may reflect a positive intake of ruminal nitrogen balance, given the low intake of corn gluteal meal and fish meal (Spain *et al.*, 1984). Increase in milk protein yield when dairy cows are fed with high soy bean meal has been reported earlier by Henderson *et al*. (1985). Differences in undegraded amino acid composition in protein supplements tends to increase or decrease milk protein yield due to alteration of amino acid flow to small intestinal (duodenum) and plasma amino acid composition. Therefore, differences in protein flow to the small intestine between protein sources may account for differences in response to milk protein yield (Spain *et al.*, 1984). Milk fat and protein percentage increase with increase in dietary crude protein. Diet containing 16% crude protein (CP) resulted in highest milk fat while protein percentage remains unchanged (Burgress and Nicholson, 1984).

2.2.6 Diseases and medication

Effects of mastitis on milk composition are determined by severity of infection, extent of infection, secondary physiological changes which alter metabolism of mammary cells and/or the cow, and whether the epithelial cells are disrupted or milk components are destroyed by enzymatic action. The fat content from milk of two

different farms decreased due to mastitis infection and due to disruption of mammary gland secretory cells (Kiro and Stefanov, 2006).

2.3 Milk Fat - Functional Properties

Fat is the only form in which the body stores its energy requirement. These contain mostly SFAs and PUFAs content rarely exceeds 4%. Milk fat is more easily digested and large amounts of it can be absorbed without producing a digestive disturbance than any other common edible fat. Fat supplies 9 cal/g when metabolised. Edible fats of low MP have about the same degree of digestibility.

Like all fats, milk fats are primary source of energy for the body. They are also carriers of flavour and vitamin compounds and contributors to the mouth feel of food. In manufacturing food, fats perform as a heat transfer medium, lubricant, releasing agent and texturising agents. These sensory, functional properties of fats and oils are determined by the levels of palmitic (C16:0) and Stearic (C18:0) SFAs, oleic (C18:1) MUFAs, PUFAs, and trans-fatty acids. Milk that has higher proportion of MUFAs and conjugated linoleic acid (CLA) may have benefit to human for cardiovascular health (Rehl *et al*., 2004).

2.4 Composition of Milk Fat

The milk fat or BF is made of 98.5% triglyceride, 0.6% phospholipids, 0.1% monoglyceride, 0.1% free FAs, 0.4% diglyceride, 0.3% sterol (cholesterol and cholesterol esters) and 0.03% cholesterol (Vanstone and Daugall, 1960). Milk fat

globules vary in size from as small as 0.1µ to 22µ in diameter. Majority are within the range 1µ – 5µ in diameter (Webb *et al.*, 1974).

2.4.1 Triglyceride

The main milk lipids are a class called triglycerides which are comprised of a glycerol backbone bonded to three different FAs. The FAs are composed of a hydrocarbon chain and a carboxyl group. Milk fat contains both SFAs and UFAs (Lehninger, 1970). LCFAs include C14:0 - myristic 11%, C16:0 - palmitic 26%, C18:0 - stearic 10% and C18:1 - oleic 20%. SCFAs (11%) include C4:0 - butyric, C6:0 - caproic, C8:0 - caprylic, and C10:0 - capric. Butyric FA is specific for milk fat of ruminant animals and is responsible for the rancid flavour when it is cleaved from glycerol by lipase action. Milk fat triglycerides are synthesised in the mammary epithelial cells. However, the FAs used to synthesise the milk triglycerides may arise from 3 sources namely from breakdown of blood lipids, dietary origin and from *de novo* synthesis within the mammary epithelial cells (Feingold *et al*., 1990).

2.4.1.1 Blood lipids

Forty to 60% of the triglycerides FAs come from the blood. These are derived from very low density lipoproteins (VLDL) composed of 90 to 95% lipids, 55 to 60% triglycerides on the inner core and five to 10% proteins at the outer surface. Chylomicrons, containing ingested FAs from the intestine, act as a source of blood-derived FAs for the mammary gland (Lehninger *et al*., 1993). The FAs contained in VLDL and Chylomicrons are dependent upon dietary lipids and on mobilised fat from body adipose tissues (Stryer, 1985). The bypass of fat in diets of ruminants

directly to the intestine became part of the FA profile of the VLDL and Chylomicrons (Van Soest, 1982). The free FAs, mon-acylglycerides, diacylgycerides and glycerol can all be taken up by the mammary epithelial cell and reused for triglyceride synthesis. Finding by Feingold *et al*. (1990) shows that FA in VLDL depend on dietary lipid and on mobilisation of fat from body adipose tissue.

2.4.1.2 *De novo* fatty acid synthesis

De novo synthesis of FAs means synthesis of new molecules of FAs from precursors absorbed from the blood (Lehninger, 1970). *De novo* synthesis of FAs occurs in the cytoplasm of the mammary epithelial cells. In the ruminant, the carbon sources used for FAs synthesis are acetate and β-hydroxybutyrate acid (ßHBA) absorbed from the rumen. Feeding unsaturated dietary fats resulted in lower concentrations of SCFAs and MCFAs and higher concentrations of LCFAs in milk fat and butter. This is due to reduced *de novo* biosynthetic activities for SCFAs (Enjalbert *et al.,* 1998). Modification of lipid by rumen bacteria also results in the formation of many odd-carbon and branched-chain FAs probably through incorporation of propionyl-CoA into Carbon-skeleton (Wahle and Peterson, 1979). The relative proportions of milk FAs derived from *de novo* synthesis or blood lipids are dependent on carbon chain length of the FA. Shorter chain FAs arise predominantly from *de novo* synthesis in the epithelial cell, while longer chain FAs arise directly from blood lipids.

2.4.1.3 Cholesterol

Cholesterol is a waxy substance consisting of fats/lipids and proteins. It is present in milk to the extent of 0.03% (Vanstone and Daugal, 1960). It is formed in the liver

and carried in the blood on the carrier molecule known as lipoproteins which are either the VLDL or low density lipoprotein (LDL) or high density lipoprotein (HDL). Once bound to carrier molecule HDL, the cholesterol is known as HDL cholesterol. HDL in effect transport fats from the cell to the liver. When the HDL cholesterol level is high, it is more cardio protective. Therefore, HDL cholesterol is considered good cholesterol (Stryer, 1985). LDL is the carrier of oxidised cholesterol as LDL cholesterol from the liver to the cell. Elevated LDL cholesterol is directly correlated with increased cardiovascular risk. LDL cholesterol is therefore called bad cholesterol. Zoosterol is a cholesterol synthesised by the animal cells. Zoosterol combined with oleic and palmitic acid form about 0.03% in animal body (Vanstone and Daugal, 1960).

2.5 Factors Affecting Milk Fat Composition

2.5.1 Dietary fat supplementation

Yield of milk and milk fat increases with dietary fat supplementation (Contarin e*t al.*, 1990). Oilseed supplementation increases concentration of MUFA, PUFA, and LCFA and decreases concentration of saturated SCFA and MCFA in both milk and cheese (Zhan *et al.,* 2005). Most fat supplements contain LCFAs. Feeding supplementary dietary fat increases milk fat yield and FA composition of the milk fat (Beaulieu and Palmquist, 1995). The bypass fat in diets of ruminants, the lipids pass directly to the intestine and become part of the FA profile of the VLDL and chlyomicrons. Fats that are protected from biohydrogenation have a less inhibitory effect on the cellulolytic organisms in the rumen and pass into the lower tract relatively intact. Cow’s milk FA composition can be altered by bypass of dietary lipid.

2.5.2 Hydrogenation of fatty acid in the rumen

The major important products of hydrogenation that might pass to lower tract to be absorbed and incorporated into milk and meat are the *trans-11*- octadecenoic acid. Rumen microorganisms cause a marked biohydrogenation of unsaturated LCFAs and this process results in production of trans-octadecenoic acids (Erdman and Sharma, 1991). Literature data (Palmquist and Jenkins, 1980) shows that unsaturated vegetable fat protected from rumen degradation can increase milk fat percent while unprotected vegetable fat or hydrogenated vegetable fat can depress milk fat percent.

2.5.3 Effect of feeding on composition of milk fat

The FA composition of milk is affected by the cow's diet. Thus, while the fresh pasture as compared to grains provides a higher intake of UFAs, their content in milk is only moderately increased. Findings by Jenkins (1998) show that supplementation of oleamide to the diets of lactating dairy cows would similarly resist biohydrogenation and cause the content of MUFAs to increase appreciably in milk fat. The effect of oleamide was compared with the effect of a diet that contained an equivalent quantity of high oleic canola oil and with the effects of a control diet with no added fat. Total FA content of the oleamide averaged 87.6% by weight and the amide content averaged (89.7 ± 4.4%) for eight measurements (Mkuu, 2001). Oleic acid was the primary FA in both the oleamide and the canola oils. Several FAs were present at 10% or less. Oleamide in the diets

of lactating cows was more effective than canola oil for enhancing milk C18:1. The content of C18:1 was more than doubled in milk when cows were fed with oleamide (Mkuu, 2001).

Studies from Avila *et al*. (2000) shows that FAs composition of milk fat were altered by inclusion of supplemental fats and administration of calcium salts of FAs containing at least 10 carbon atoms to ruminants with increases in milk production. The major cause of decreased output of SCFA in the milk when dairy cows consume rations containing added fat is mediated within the mammary gland and is probably due to the dietary LCFAs acting as partial inhibitors of the synthesis of the SCFAs (Avila *et al*., 2000). Feeding fat to dairy cows can increase milk yield (Ruppert *et al*., 2004) as well as an increase in milk fat and LCFAs content in milk (Aldrich *et al*., 1997). However, feeding fat above a certain level reduces feed intake and reduces fiber digestion by inhibiting microbial fermentation that occurs in the rumen.

2.6 Product Quality

With the advancement in health perspectives, the need for high quality milk and milk products in the developed and developing countries has become an important issue. The types of FAs present in milk fat can influence the flavour and physical properties of dairy products (Stegeman *et al*., 1992). Changing FA composition affects milk fat MP directly because each FA has a characteristic MP, and indirectly, by influencing

the array of FAs in the various positions of the glycerol backbone that affect the MP. This structure is the most important determinant of crystallisation behavior and hardness of milk fat, as well as MP. Factors such as disease (mastitis), hygiene, feeding and management can also affect MP of milk fat.

Under socio-economic point of view consumers decrease consumption of SFAs, which in turn are associated with increased risk of cardiovascular disease and prefer milk with more UFAs (Cohen *et al.*, 2006). Milk from cows that are grazed on grass has higher concentration of long UFAs, MUFAs and PUFAs than cows that are fed concentrates and silage (Elgersma *et al.*, 2006). This type of milk is preferred by consumers (Elgersma *et al.*, 2006). Farmers in Netherlands that produce milk from cows grazed on grass received more price for their milk compared to those that produce milk from concentrate and silage (Elgersma *et al.*, 2006). This implies that in future milk price might be based on the fatty acid composition of milk fat rather than the content of milk fat and protein that are currently used as a measure of milk quality (Schroeder, 1996).

2.7 Melting Point of Milk Fat

Milk fat is composed mainly of Triacylglycerol esters of FAs and glycerol, which represent 97 to 98% of total fat. More than 400 FAs have been identified, varying in chain length and unsaturation. The extreme diversity of milk fat FAs and triglyceride, each characterized by its own MP induces a wide melting range, which spans from about –40 to 40°C. At intermediate temperature, milk fat is partially crystallized and corresponds to a mixture of solid fat (crystals) and liquid fat (oil) (Jensen and Newburg, 1995).

Table 3: Principal fatty acids found in milk and their melting point (°C)

	Molecular formula	Chain length	Melting point
Butyric	$CH_3(CH_2)_2COOH$	C_4	–8°C
Caproic	$CH_3(CH_2)_4COOH$	C_6	–2°C
Caprylic	$CH_3(CH_2)_6COOH$	C_8	16°C
Capric	$CH_3(CH_2)_8COOH$	C_{10}	31.5°C
Lauric	$CH_3(CH_2)_{10}COOH$	C_{12}	44°C
Myristic	$CH_3(CH_2)_{12}COOH$	C_{14}	58°C
Palmitic	$CH_3(CH_2)_{14}COOH$	C_{16}	64°C
Stearic	$CH_3(CH_2)_{16}COOH$	C_{18}	70°C
Arichidonic	$CH_3(CH_2)_{18}COOH$	C_{20}	
Oleic	$CH_3(CH_2)_7$ $CH=CH(CH_2)_7COOH$	$C_{18:1}$	13°C
Linoleic	$CH_3(CH_2)_4(CH=CHCH_2)_2(CH_2)_6COOH$	$C_{18:2}$	–5°C
Linolenic	$CH_3CH_2(CH=CHCH_2)_3(CH_2)_6COOH$	$C_{18:3}$	

Source: O'Mahony (1986)

The MP of FAs is proportional to chain length (Table 3). As expected, the molecular weight and the unsaturation of the FAs are the major factors, which determine the MP properties of integral fat. In addition, the softness of the fat depends on the distribution of the FAs in the triglycerides and on the physical state of the fat. Jersey cows (Beaulieu and Palmquist, 1995) tends to have a higher proportion of short-chain FAs than other breeds, which would decrease the MP, however, the oleic acid (C18:1) content is lower and stearic acid (C18:0) is higher thus increasing the MP of FAs. The MP of FAs is lowered both by increased SCFAs and MCFAs and by increased ratio of C18:1 to C18:0 (Morales *et al.,* 2000).

Table 4: Chemical composition of oil seed meals (percent dry matter)

Item	Cotton	Sesame	Groundnut	Coconut	Afr. palm	Castor	Sunflower
C.P.	34.8	36.3	46.2	15.6	14.1	26.7	37.9
C.F.	16.6	7.0	4.5	40.3	14.1	8.4	19.0
E.E.	7.5	11.6	7.5	18.8	17.4	2.8	8.9
Ash	6.6	11.1	4.4	5.4	7.8	8.5	6.5

Source: Tanzania Feedstuff Table for Ruminants (2004)

2.8 Plant Protein Supplements

2.8.1 Oil seed cakes

Oil seed cakes and meals are the residues remaining after the removal of oils from oil seeds (Doto *et al.*, 2003). Although the biological value of these oil seed cakes as protein source are lower compared to that of animal protein sources, they are still valuable in the feeding of animals because they are cheaper compared to animal protein sources. Numerous seeds and fruits are cultivated for their oil content in the tropics. Those yielding notable volumes of by-product material are cotton, sesame, sunflowers, groundnuts, coconuts, African palm, and castor (Table 4). The principal by-products of these crops are the above-ground-plant residues following harvest and the oil seed cakes remaining after oil removal. Oil seed cakes are rich source of proteins and are used as cattle feed all over the world including Tanzania (Kinabo, 1980). Table 4 presents the chemical composition of oil seed cakes of plant oil seeds cultivated in Tanzania.

In general, all these meals have high protein levels, ranges between 14.1 to 46.2% (Doto *et al*., 2004). Sesame, groundnut, and castor meal have low fiber levels, while coconut, African palm and sunflower vary in fiber content according to the oil

extraction process and the amount of hulls included. These meals are the main protein sources for animals. Because of their high value for non-ruminants, they are partially replaced in ruminant feeding by non-protein nitrogen (NPN) sources (McDonald *et al*., 1973). Kinabo (1980) indicate that National milling Corporation figures for 1978 showed that a total of 7,675 tones of oilseed cakes were used in the production of different livestock feeds of which 51% was CSC.

The demand of protein for human and animal is increasing and is likely to lead to an increased protein scarcity and cost. It is therefore, essential to exploit fully the potential of any protein sources including SSC as livestock feeds (Ngowo, 1986). Due to high quality of Tanzanian organic cotton, it is growing for international markets. Assessment conducted by RLDC (2005) indicated that cotton is cultivated at least in 5 regions of central corridor (Morogoro, Dodoma, Singida, Shinyanga and Tabora) and it has a higher potential for increased production and income to farmers. Therefore, improving processing of CSC will be beneficial to livestock keepers as well as poorest communities of rural areas in Tanzania. Whole oilseeds (e.g. soybean, sunflower, and canola) contain primarily UFAs; however, most of the UFAs fed to cows are converted to SFAs by ruminal microorganisms. This results in primarily SFAs in milk fat despite consumption of a diet high in UFAs. Saturation of UFA in the rumen can be prevented experimentally by infusing FAs directly into the abomasum (bypassing the rumen) (La Count *et al.,* 1998). An increased energy concentration in the feed ration, with no change in the ratio of concentrate to bulky feed, may be achieved by adding fat (Espindola *et al*., 1997).

2.8.1.1 Cotton seed cake

Whole cotton seed meal has been used as a source of CP, fiber and energy for dairy cows. Findings by Noftsger *et al.* (2000) shows that cow fed whole CSC has higher yield of milk, milk protein, milk fat, SNF and 4% FCM. Cottonseed oil is rich in palmitic acid (22 - 26%), oleic acid (15 - 20%), linoleic acid (49 - 58%) and 10% mixture of arachidic acid, behenic acid and lignoceric acid. It also contains about 1% sterculic acids and malvalic acids in the crude oil *(*Kelly *et al.*, 1998). The gossypol content has been considered non-toxic depending on the amounts of cottonseed meal fed to cattle.

Observation made by McNight (1968) indicated that when up to 880 ppm gossypol were in the total diet, adding ferrous sulphate to provide an iron: gossypol ratio of 1: 1 improved weight gains in cattle. Because of incomplete rumen and micro flora development in young calves and lambs, only 10 to 15% is included in calf and lamb concentrate mixes. Quality is improved by mild heating by making the amino acid more available while excessive heating is undesirable as it impairs the nutritive value of CSC and unavailability of amino acids (Kinabo, 1980). Heated cotton seed meal had a better feeding value than raw seed because heat inactivated the toxic polyphenols constituent gossypol. Decorticated cottonseed meal is one of the best protein supplements for dairy cows, buffaloes and sheep. It has been highly recommended for incorporation in the formulated feed so as to function as a bypass-protein to raise the milk yield. Feeding protected cotton seeds result in a large increase in milk fat C18:2 percentages and in the C18:0 to C18:1 ratio despite the fact that CSC is poor in C18:0 and contains only 16% of C18:1. This response is

related to the fact that cotton seeds (Chilliard *et al*., 2003) are rich in cyclopropenoic FAs, which are strong inhibitors of mammary delta-desaturase activity. The rumen modifications depend to a large extent on the FA composition of the fat in the ration. The modifications in the mammary gland are catalysed by desaturation enzymes, especially delta-9-desaturase that converts *trans-11* C18:1 to *cis-9*, *trans-11* C18:2, the most abundant CLA and C18:0 to C18:1 (Jensen, 2002).

With unprotected cotton seeds, the increase in milk C18:2 does not occur, whereas the increase in milk C18:1 was probably due to unidentified isomers of C18:1 arising from hydrogenation in the rumen (Chilliard *et al*., 2003). Results from studies indicate that the transfer efficiency of Eicosapentaenoic acid (EPA) and Docosahexaenoic acid (DHA) was low, averaging 2 to 4% (Chilliard *et al*., 2001).

2.8.1.2 Sunflower seed cake

Sunflower oil is one source of fat that can be used for feed supplement, which contains 12% SFA and 88% UFA (Grant and Kubik, 1990). Sunflower oil consist of 8% palmitic (C16:0), 3% stearic (C18:0), 13.5% oleic (C18:1), 75% linoleic (C18:2) and 0.5% linolenic (C18:3). Incorporating sunflower oil into rations may be a successful way to get more energy into the cow with the same feed volume (Palmquist, 1988).

Sunflower seed extract contains about 30% CP with 30% CF. It is widely used as protein supplemental for animal feed (Dorsi *et al.*, 1965). The undecorticated sunflower oilseed meal has been used extensively in ruminant feeding. It is superior

to most vegetable proteins in digestibility. The diet with SSC (Zhan *et al.*, 2005) had no effect on milk yield but reduced milk TS, fat, and protein proportion. Sulphur amino acid is higher in SSC and is near or above the average content of other essential amino acids from oilseed sources, except for lysine and leucine (Roberts, 1974).

Sunflower oil has high level of linolenic acid (Kelly *et al.*, 1998) required for the cell membrane structure, cholesterol transportation in the blood and for prolonged blood clotting. The FA composition of milk can be altered by providing specific FAs to the abomasum of lactating dairy cows (La Count *et al.*, 1998). Research indicate that contents of SCFAs and MCFAs (including myristic and palmitic acids) decreased, and the content of oleic acid (MUFA) increased when FAs from high-oleic sunflower or canola mixtures were infused into the abomasum (La Count *et al*., 1998; Jenkins and Lundy, 2001). This indicates that the content of SFAs in milk fat can be decreased and the content of MUFAs- and PUFAs in milk fat can be increased by providing UFAs directly to the abomasum.

Sunflower oil helps to reduce the serum cholesterol level. The protein content of sunflower meal ranged between 30 to 40% and is low in lysine content (Rehm and Espig, 1991). The amino acid profile of the meal is strongly affected by treatment during processing. Prolonged heating severely depresses the availability of aspartic acids, arginine, threonine, leucine, lysine and tryptophan at the same time increasing the content of glutamic acid, serine and ammonia (Donell, 1978; and Smith, 1968) as cited by Ngowo (1986). The addition of sunflower oil increase the concentration of

octadecenoic acid (C18:1) in milk fat relative to when peanut or linseed oils were fed (Kelly *et al.*, 1998). Feeding sunflower oil results in greater CLA concentration than other treatments. Cows fed with high oleic sunflower seeds and regular sunflower seeds produced butter which is softer, high proportion of UFAs in milk and butter; more spreadable UFAs and exhibited acceptable flavour, manufacturing and storage characteristics (Middaugh *et al.*, 1988).

2.8.1.3 Sesame meal

Sesame meal has been used in various proportions as a protein supplement for both young and mature ruminants, particularly in countries where this crop represents a major source of edible oil. Feeding more than 3 kg daily of sesame meal to lactating cows produced excessive UFAs in butter and disagreeable taste in milk (Göhl, 1975). Sesame meal has nearly 1 g/16 gN more methionine and cystine than the average of other oil meals but the lowest lysine content of any economically important oil meal. The high phytic acid content of the cake appears to promote a natural chelation and lowering of diet calcium and zinc availability (Göhl, 1975). Sesame cake is digestible and utilised as a good source of proteins. The oil in sesame cake has a good taste and keeps well due to the antioxidants sesamin and sesamolin. It contains 35 to 47% linolenic acid. The cake contains 35% CP with high methionine content (Rehm and Espig, 1991).

2.8.1.4 Groundnut meal

The use of groundnut (peanut) meal has no general limitations in ruminant feeding. Replacing protein supplements with the undecorticated meal, relatively lower in

protein and higher in fiber, has reduced gains in cattle grazing dry-season savanna, but this was overcome by compensatory response in the following rainy season (Quinn *et al.,* 1966). Mould groundnuts may contain toxic substances; the most dangerous being aflatoxin, produced by the fungus *Aspergillus flavus*.

2.8.1.5 Coconut meal

Coconut meal commonly known as copra cake is fed mostly to dairy cattle. Though it is comparatively a moderate protein (20 - 22%) source, it is highly palatable and its protein has a very high by-pass value. Although feeding more than 2 kg of coconut meal daily to dairy cows may result in tallowy butter, beef cattle can consume more without carcass quality impairment (Göhl, 1975). Copra cake is obtained from dried endosperm of coconut palm cakes by pressing process. The cake contains 20% protein which is used as animal feed (Kinabo, 1980).

2.8.1.6 Palm kernel meal

Palm kernel meal **c**ontains about 50% lauric acid and about 17% mystic acid. Although palm kernel meal may have appreciable oil content, this by-product is usually dry and unpalatable unless mixed with other feedstuffs such as molasses. There are no nutritional limitations to its use in ruminant feeding when it is blended with palatable feeds. In this manner 2 to 3 kg/day have given satisfactory results in adult cattle (Göhl, 1975). Palm kernel cake is relatively low in protein content as compared to the other oilseed cakes in Sub-Saharan Africa. Although some observations (Chicco and Shultz, 1977) indicate that palm kernel cake can be used in rations for monogastrics, it has been largely used in Sub-Saharan Africa for cattle

feeding, especially dairy animals where it can serve as the main protein source (Adegbola, 1977). Palm oil from Palm kernel cake contained approximately 47% palmitic acid which is higher than most source of fat fed to dairy cows (Elliot *et al.*, 1996; Allen, 2000).

2.8.1.7 Soy bean meal

Soybean meal is the most common plant protein supplement meal and it is the product obtained by grinding the flakes which remain after removal of most of the oil from either whole or dehulled soybeans by a solvent extraction process (solvent extracted meals). The 44% CP soybean meal is the most common. Two other sources of soybean meal also are available: a dehulled soybean meal at 48% CP and expeller or cold processed soybean meal (42% CP and 5% oil). The product resulting from dehulled soybeans must contain not more than 3.5% crude fibre (CF) and not more than 12% moisture. The product resulting from expeller or cold processed must contain not more than 7% CF and not more than 12% moisture. Soybean meal is highly palatable and has a moderate level of rumen undegradable protein (RUP) (Walker, 2001). The most limiting amino acid to milk protein synthesis in soybean meal is methionine (Maiga *et al.*, 1996). Mechanically-extracted (expeller) meals contain more fat than solvent extracted meals. Expeller meals are also higher in RUP than solvent-extracted meals (55 vs 35%). The CP content of soybean meal and its ruminal degradability are highly variable. The higher fraction of RUP makes heat-processed soybean meal particularly attractive in diets for lactating dairy cows. The RUP in heat-processed soybean meal has high value because of its high lysine content (Howard, 1988).

2.8.1.8 Linseed meal

Linseed meal (Göhl, 1975) is the product obtained by grinding the flakes which remain after the removal of most of the oil from flaxseed by a solvent extraction process (solvent-extracted meal) or by grinding the cake or chips which remain after removal of most of the oil from flaxseed by a mechanical extraction process (mechanically-extracted meal). Protein degradability in linseed is high and similar to soybean meal (65%). Linseed meal is palatable and mildly laxative. The fiber content in Linseed meal is higher, 19 vs 10% acid detergent fiber (ADF) and energy content was lower compared to soybean meal (Howard, 1988). Efforts have been made to enhance level of EPA in milk by feeding linseed cake to ruminants (Jensen *et al*., 1991).

2.8.1.9 Canola seed cake

Canola seed cake commonly known as rape seed cake consists of the cake obtained after the removal of most of the oil, either by direct or prepress solvent extraction processes from rapeseed (Brassica spp.). The oil component of which contains less than 2% erucic acid and the solid component of which contains less than 30 micromoles of glucosinolates per gram of air-dry oil-free solid (Bell, 1993). It must contain a minimum of 35% CP, a maximum of 12% CF, and a maximum of 30 micromoles of glucosinolates per gram (Bell, 1993). The inclusion rate of canola meal in milking cow diets is often restricted to 35% CP and not more than 12% CF because of its high content of ruminally degraded protein (72% CP). Research has demonstrated its value as a very satisfactory protein and mineral source for all

classes of dairy animals (Hickling, 2001). Canola meal protein is degraded relatively fast in the rumen.

2.9 Lipid Composition of Feedstuffs

The lipid content of feedstuffs varies widely, from less than 1% in some by products to 100% in some fat supplements (Palmquist, 1988; van Soest, 1994). Similarly, the composition of the crude lipid (ether extract (EE)) also varies widely. Triacylglycerol (90% FA) is the major lipid class in rendered fats, in most cereals, and in oil seeds (> 95, 2 to 8; and 18 to 45% EE respectively), whereas total FA in forages is often less than 50% of the EE (Palmquist and Jenkins, 1980; van Soest, 1994). A large part of EE in forages is comprised of non-saponifiable substances (waxes, chlorophyll, cutin, etc). The majority of lipid in forages is found in the chloroplasts and its proportion of the plant dry weight decreases as the plant matures (Hawke, 1973). Triacylglycerol is potentially completely metabolisable by animals, whereas the non saponifiable fraction has no energy value, although it may offer other desirable nutritional characteristics (e.g., fat-soluble vitamins, carotene). Glycerol (10 to 11% of the glyceride by weight) has an energy value comparable to other carbohydrates, whereas the FAs contribute the highly dense energy value of fats. The EE fraction of plants, because it contains numerous nonnutritive substances, is not a nutritionally uniform fraction, whereas FA constitutes a uniform fraction that can be measured and used in computer modeling to estimate the energy value of feedstuffs (Weiss, 1993; NRC, 2001).

2.10 Summary of the Literature Review

Milk contains different nutrients including fat. The role of fats and FAs is to provide energy, lubrication and laxative effect to human being. But nowadays consumers prefer milk with low amount of fat to prevent heart diseases that can be caused by the milk fats. Milk fat contains SFAs and trans-FAs which may be detrimental to their health. Scientific evidence from the literature shows that milk fat can be altered through different feeding practices (Grummer, 1991; Palmquist *et al*., 1993). Feeding supplemental fats may increase the content of C18:0 and C18:1 while decreasing the content of C14:0 and C16:0; at the same time, however, content of more desirable SCFAs also may be decreased. Oil seed cake supplement differ in FA composition but through rumen modification (escape or biohydrogenation) milk fat can have the required FAs which is not detrimental to the human health. The present study focused on the effect of feeding dairy cattle concentrates containing different oil seed cake supplement on FA composition and quality of BF.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experimental Location

The study was conducted at Magadu dairy farm, Department of Animal Science and Production (DASP) of the SUA. SUA is situated at Latitude S 6.8528º and Longitude E 37.6575º, and some 2.5km south of Morogoro town. The area is 550 meters above sea level and receives both long and short rains in the months of February to May and October to November, respectively. The average annual rainfall is about 800mm per annum. The temperature ranges from 27 to 31°C during day time and falling to about 14°C at night in the coolest months of June and July. The study was conducted during the period of long rains season (March to June).

3.2 Experimental Animals and their Management

Two *Bos taurus* breeds (Friesian and Ayrshire) from Magadu Dairy Research Unit and University farm at SUA were used in the experiment. A total of 24 lactating cows were selected that is, 12 cows of each breed. The criteria used for selection of the experimental animals included:

- fair to good body condition,
- good health with no history of recent clinical diseases,

Before the start of the experiment, animals from Magadu farm were subjected to pregnant diagnosis while pregnant and non pregnant animals from University farm were identified from their records. The weights of the selected animals were estimated using weighing band. “Dominex” was applied fortnight to control ectoparasites.

3.3 Allocation of Animals to Experimental Diets

Animals were blocked according to breed and allocated to 3 groups of 8 animals (4 Friesian and 4 Ayrshire) each. Animals used in the experiment varied with respect to parity and stage of lactation hence in the allocation of animals to treatments, these sources of variation (parity and stage of lactation) were partly taken care of by ensuring that each treatment group contain animals belonging to the early parity, medium parity, early lactation and medium lactation stage. Each group of 8 animals was fed each of the 3 dietary treatments in Latin Square Design, with a change over period of 21 days as shown in Table 5. A preliminary period of 2 weeks where all animals were fed standard supplementary diet was allowed before the animals were fed the treatment diets. This period allowed the animals to get accustomed to the experimental condition. During the experiment animals were allowed to consume their treatment diet for 7 days before data collection.

Table 5: Allocation of animals to treatments

P	G1		G2		G3	
P1	CO FR	AY	CSC FR	AY	SSC FR	AY
P2	CSC FR	AY	SSC FR	AY	CO FR	AY
P3	SSC FR	AY	CO FR	AY	CSC FR	AY

Note: P = period, P1 = Period1, P2 = Period2, P3 = Period3; G = group; CO = control diet (Maize bran), CSC = cotton seed cake concentrate diet, SSC = sunflower seed cake concentrate diet, FR = Friesian and AY = Ayrshire.

3.4 Experimental Feeds and Feeding

3.4.1 Experimental feeds

Napier grass **(***Pennisetum purperium)* used in the study was harvested daily from Magadu plots. It was chopped into smaller parts using a hand-held machete to make it easy for the animals to consume and minimize the selection of palatable parts during feeding. Concentrates for the different dietary treatments were formulated using the appropriate ingredients to contain 13.00 MJME/kg DM and 92.26 gDCP/kg DM. Mixing of the ingredients was done manually at Magadu Dairy Farm.

3.4.2 Feeding the experimental animals

The animals were allowed to graze during the day time for 5 hours from 0800 hours to 1300 hours. It was assumed that intake from grazing will supply 1% of BW dry matter intake (DMI). From 1300 to 1600 hours animals were allowed to rest in a resting paddock that was supplied with clean water troughs for animals to drink. Animals were supplied with 3 kg of treatment concentrate diets at each milking that was done twice per day at 0600 and 1800 hours. The amount offered was assumed to supply 1.2% of BW DMI. It was hypothesized that any differences in milk yield as a result of feeding with and without oil seed cake supplement would be due to the different source of protein in the concentrate components of the two diets. After evening milking animals were confined in individual stalls during the night and they were fed Napier grass to supply 0.8% of BW DMI. Every morning all refusals from the previous day's feeding were removed from each individual stall feeding and measured using a slatter spring balance. The daily intake of Napier grass by each

animal was estimated to be the difference between the amount offered and the quantity refused.

Table 6: Feed formulation and nutrient composition

Ingredients	Standard supplementary diet	Control diet	CSC (Diet I)	SSC (Diet II)
MB(%)	56.40	87.00	63.30	53.50
CSC(%)	18.30	-	34.80	-
SSC(%)	23.25	-	-	44.50
Premix(%)	0.50	0.50	0.50	0.50
L/stone(%)	1.00	1.00	1.00	1.00
Salt(%)	0.50	0.50	0.50	0.50
CP(%)	15.88	11.08	19.32	17.24
ME (MJ/kg)	13.79	15.23	12.22	13.92

Note: CSC=cotton seed cake, SSC=Sunflower seed cake, MB = Maize bran.

3.5 Sampling of Feeds

3.5.1 Sampling of forage from the grazing paddock

Sampling of feed ingredients was done before the start of the trial to assess the nutritive value of the forage. Forage samples were collected from Sokoine University of Agriculture (Magadu farm) in a grazing paddock. Systematic sampling was used whereby forage samples were collected after every 10 foot-steps. Species identification was done before clipping. Fresh weights of harvested samples were measured using spring balance. Harvested fresh samples were kept in a dry paper bags then brought to the DASP laboratory at SUA. The samples were oven dried at 72 °C for 48 hours then cooled in desiccators and reweighed. DM content was obtained by difference. In the laboratory, samples were ground in a mill to pass through a 1mm diameter sieve and stored in air tight containers for chemical analysis.

3.5.2 Sampling of Napier grass (*Elephant grass)*

Fresh, chopped *Elephant grass* of about 500 g was collected and kept in a paper bag 3 times during the experiment that is once at each experimental period. The samples were brought to DASP laboratory immediately and oven dried at 72 °C for 48 hours then cooled in desiccators and reweighed; then milled and stored in a sealed sample bottles ready for chemical analysis.

3.5.3 Concentrate diet sampling

Concentrate samples of 500 g of each test diet was collected in a dry paper bag once during each of the test period making a total of 3 samples per test diet. The 3 samples of each test diet were mixed together and a representative sample of 500 g from composite sample of each test diet was collected and stored in a dry paper bag. In the laboratory, the samples were oven dried at 72 °C for 48 hours then cooled in desiccators and reweighed. The samples were ground in a mill to pass through 1mm diameter sieve and stored in air tight containers ready for chemical analysis.

3.6 Milking and Milk Sample Collection

Milking was done by milking machine at Magadu dairy farm twice a day at 0600 hours and 1800 hours. When there was no power, milking was done by hand, by 3 experienced hand milkers. Cooper® milking salve, of Cooper Pharmaceuticals (Kenya Ltd) was applied to the teats and adjacent areas of the udder after milking to prevent cracking of the udder and teats. The amount of milk from each individual cow was recorded. Milk samples were collected once a week (morning and evening) during test period. Aliquot milk samples of 50 ml from each cow were collected into

pre-cooled 250 ml plastic containers during the morning and stored in a deep freezer located at the farm. During the evening milking, the sample collection procedure was repeated. An aliquot of evening milk samples were mixed with a respective aliquot of morning milk sample which gave a total aliquot milk sample for individual animal on that day that is 100 ml for chemical analysis.

3.7 Collection of Milk for Butter Fat Preparation

The amount of milk from each individual animal (morning and evening) was recorded daily. Milk sample from 4 animals receiving the same treatment diet was collected during test period and mixed together to get 10 litres of milk. The milk was separated into cream by cream separator at Magadu Dairy Unit. The cream was pasteurized at 90 ºC for 10 minutes and immediately cooled by water and kept in 250 ml glass bottles and stored in a deep freezer for later churning into butter. The cream was churned into butter by shaking in a bottle. Butter obtained from the cream was stored in a deep freezer located at DASP for FA composition determination. Butter was transferred to the University of Dar es Salaam chemistry laboratory in a cool box ready for analysis of FA composition by gas chromatography (GC).

3.8 Chemical Analysis

3.8.1 Experimental feeds and forages

All feed ingredients and formulated feeds used in the experiment were analyzed at the DASP Laboratory. The feed samples for chemical analysis were ground to pass through a 1 mm screen in a Christy and Norris 20cm laboratory hammer mill. DM percent was determined by drying the sample in an oven at 103 to 105 ºC for 24

hours. CP percent was determined by Kjeldahl method as outlined by Bremner and Mulveney (1992) and Nitrogen (N) percent obtained was used to calculate the CP percentage using the relationship: CP% = N% x 6.25 (AOAC, 1990). EE percent was determined using the Soxtec System HT- extraction technique (AOAC, 1990) and ash percentage was determined immediately after the DM determination by incinerating the samples in a muffle furnace at 600 ºC for 4 hours. The ash was cooled in a decicator and then weighed. CF percent was determined by using the fiber system and Weende method (AOAC, 1990).

Nitrogen free extracts (NFE) percent was calculated by differences:

NFE% = 100 - (moisture percentage + CP% + EE% + CF% + Ash percentage), where:

CP% = crude protein percent.

EE% = nitrogen free extract percent.

CF% = Crude fiber percent.

3.8.2 Milk composition

The milk samples were analyzed for pH, milk density, milk composition with respect to BF, protein, TS and SNF. Milk pH was determined by milk pH meter; milk density (lactometer test) was done by using hydrometer (ILCA, 1988); BF was determined by using Gerber method (ILCA, 1988); N percent was determined by Kjeldahl method outlined by Bremner and Mulveney (1992) and the CP percent was calculated by the formula:

CP% = N% x 6.38, where;

N% = Nitrogen percent.

CP% = crude protein percent.

6.38 = conversion factor for milk.

TS were calculated using the standard formula (ILCA, 1988) as:

L/4 + (1.22 x BF percent) + 0.72, where;

L = lactometer reading.

SNF was computed from the equation (ILCA, 1988);

SNF% = TS% - BF% where;

SNF% = Solid not fat percent.

TS% = total solid percent.

BF% = butter fat percent.

3.8.3 Melting point determination

MP of BF was analysed using MP apparatus Stuart Model SMP1, made in Great Britain. The apparatus has horizontally, mounted mercury-in-glass thermometer, maximum temperature of 350 °C and wells capable of viewing 3 capillary tubes simultaneously.

Glass capillaries with two opening ends were used. Small quantity of solid butter sample at room temperature was filled in 1 end of MP capillary to a depth of about 1 mm and placed into one of the three sample wells of the MP apparatus. The MP power switch was turned on. The samples were observed through a magnifying glass as the temperature changed. The temperature of MP apparatus was recorded when discoloration of samples occurred. While monitoring the temperature, the samples

were observed until the appearance of some liquid and gas bubbles in the capillary was noted. The MP power switch was turned off and the samples were removed from the wells of the MP apparatus.

3.8.4 Extraction of lipids and liposoluble compounds

The extraction of the milk BF was performed according to the procedure outlined in ISO 14156 | IDF 172 (2001) using 100 gm of butter sample melted in an oven maintained at 50 ± 5 ºC. Butter sample was filtered through 11cm diameter ash less whatman filter paper number 542, containing 1 g of anhydrous sodium sulphate (Na_2SO_4). The filtrate was collected in a 100 ml beaker maintained in an oven set at a temperature of 50 ± 5 ºC and filtration was performed in an oven. Care was taken to ensure that no serum was transferred when decanting the melted butter onto the filter paper.

3.9 Preparation of Fatty Acid Methyl Esters

3.9.1 Chemical preparation

Preparation of fatty acid methyl esters (FAMEs) was performed according to the procedure outlined in ISO 15884 | IDF 182 (2002) using transesterification reagent of approximately 2 mol/litre prepared by dissolving 11.2 g of Potassium Hydroxide (KOH) in 100 ml of Methanol (analytical grade).

3.9.2 Methylation of fatty acids

About 100 mg of the filtered butter was weighed in a test tube of diameter 16 mm, and length of 100 mm, fitted with PTFE-lined screw cap. Five ml of hexane was

added to the sample and mixed then 0.2 ml of the transesterification reagent was added and the tube was capped. The content of the tube was mixed vigorously using a vortex mixer for 1 min. After an additional reaction time of 5 min, 0.5 g of solid sodium hydrogen sulphate monohydrate ($NaHSO_4.H_2O$) was added into the content of the tube and mixed again. The test tubes with the fat sample were placed in the Joan S. A. centrifuge (St. Herblain-France) and operated at 786,320 rpm (± 10976) and centrifuged for 3 min at room temperature. After centrifuging, an aliquot from the clear supernatant of the test portion was taken for the GC analysis.

3.10 Fatty Acid Analysis

The FA composition of the milk BF was analyzed according to the procedure outlined in ISO 15885 │IDF 184 (2002) using GC (Varian 3400 chromatograph; Centrifugeur – B4i) equipped with a 60-m DB-wax semi polar glass capillary column (internal diameter: 0.32 mm; 0.25 µm film thickness; filled with Carbonwax® polyethylene glyco (PEG); (J and W Scientific, Folsom, CA) and a flame ionization detector (FID).

At the time of the sample injection, the column temperature was 130 °C, and it was then ramped up at 5 °/min to 240 °C. Inlet and detector temperatures were 250 and 300 °C, respectively. The split ratio was 100:1. The flame was made of hydrogen plus air and the flow rate for Nitrogen carrier gas (Praxair Inc, Vanier, Canada) was 5 ml/min. Peak area was measured using a Nelson Analytic System 2600 (version 5; PE Nelson, Cupertino, CA). Each peak was identified with the use of FAME standard mixture (Supelco™ 37 Component FAME Mix) on the basis of their retention times (Appendix 2). The area-to-concentration ratio for all identified FAs was used to

determine their respective concentrations after adjustment for the difference in molecular mass between the FAs and their methyl esters (Appendix 3).

3.11 Calculation of the Peak Area of Standard Fatty Acid Methyl Esters

The percentage, P_{Ai}, of the total peak area represented by the peak of component *i*, was calculated using the following equation:

$P_{Ai} = (Ai / \sum Ai)\ 100\%$, *where;*

Ai = the numerical value of the sum of all peak areas corresponding to the component *i.*

$\sum Ai$ = the numerical value of the sum of all peak areas corresponding to the component FAMEs.

3.12 Determination and Standardization (Calculation) of Correction Factors

3.12.1 Determination of correction factors

The correction factor *fi*, was determined by using the following equation:

$fi = (\omega_i / Ai) = (\omega_i \sum Ai)/(\sum \omega_i . Ai)$, where*:*

ω_i = the mass fraction of component *i*, in the reference milk fat expressed as FA;

$\sum \omega_i$ = the sum of the mass fractions of the various components in the reference milk fat expressed as free FA.

3.12.2 Standardization (calculation) of correction factors

The value of individual correction factor with respect to the correction factor of methyl palmitate was standardized to obtain the relative correction factor component *i*, *fi*, by using the following equation:

$f_i = (f_i/f_p)$, where:

f_p = the value of correction factor for methyl palmitate (f_i for methyl palmitate = 1).

3.12.3 Determination of methyl esters for fat sample

The transmethylated butter oil (fat sample) was analysed and its peak area was determined by using the following equation:

$P_{Ai} = (Ai/\sum Ai)$ x 100%, where:

P_{Ai} = the numerical value of the peak area corresponding to component *i*,

$\sum Ai$ = the numerical value of the sum of all peak area corresponding to the component FAMEs in the fat sample (Appendix 2).

3.13 Calculation of Results

The mass fraction of each individual component *i* in the test sample, $\omega_{i,s}$, was calculated by using the following equation:

$\omega_{i,s} = (f_i . p_{Ai,s})/\sum(f_i p_{Ai,s})100\%$, where:

$\omega_{i,s}$ = the mass fraction, in percent, of the individual component *i* in the test sample, determined as grams of free FA per 100 g of total FA;

$p_{Ai,s}$ = the percentage of the peak area representing component *i* in the test sample.

3.14 Statistical Analysis

The collected data were analyzed using Statistical Analysis System (SAS) where General Linear Model (GLM) was employed. The following model was employed:

Yijk =U + Bi +Pi+ Ti + b (Xij – X) + eijk

Where:

Yijk = the response produced by diet i in cow j;

U = the mean response;

Bi = breed effect;

Pi = Period;

Ti = the effect of diet i;

Xij = individual response;

X = mean of the sample;

b (Xij – X) = covariate/ correction factor for initial milk yield;

b = regression coefficient for Yij on Xij;

eijk = The error assumed to be normally and independently distributed with a mean zero and a constant variance.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Chemical Composition of the Forages and Treatment Diets

The results of chemical composition of the forages and the treatment diets are presented in Table 7 and 8 respectively. The CP percent in pastures ranged from 2.59 to 11.55% whereas in legume species was 8.55 to 15.4%. The result for Napier grass of 11.51% was within the reported CP percent of Napier grass (9.8 to 12.1%) (Table 4). The variation in chemical composition and the nutrient of the grasses and legume species might be due to species, ecological zones and processing technique, let alone variations that occur between laboratories during analysis as reported by Doto *et al.* (2004). Previous researchers reported that variation in nutritive value (chemical composition) in forages might be due to its stage of growth, season and management practices (Lyimo, 2004). Amina and Chen (1989) reported a low range of CP percent in tropical pastures 3 to 8% while in legume species the CP percent was ranged between 5.6 to 30%.

Table 7: Chemical composition of the grasses and legumes

		DM%				
SAMPLE	**DM%**	**ASH%**	**EE%**	**NDF%**	**ADF%**	**CP%**
Grasses						
Urochloa mozambiensis	25.5	16.1	1.16	76.5	47.6	4.40
Heteropogon contortus	28.7	12.6	1.15	76.8	49.9	3.70
Chloris gayana	20.0	8.67	1.12	80.2	52.2	5.00
Cynodon nemfuensis	19.1	10.9	1.62	81.1	52.3	11.6
Hyperrhenia rufa	21.7	13.1	1.15	77.5	51.3	2.59
Brachiaria SPP	20.3	12.0	1.24	75.5	44.3	5.47
Panicum maximum	25.0	12.7	1.55	77.6	52.5	4.30
Hay	-	11.0	0.91	75.3	50.2	3.90
Napier grass	20.0	17.7	1.62	66.8	37.5	11.5
Legumes						
Centrosema pubescence	26.0	7.62	1.84	67.4	48.3	15.4
Stylosanthes humilis	26.4	6.96	1.82	72.5	52.4	9.44
Stylosanthesguyanensis	25.9	6.10	1.88	70.5	58.0	8.55
Macroptilium atropurperium	24.0	5.71	1.55	65.5	47.2	11.7

4.2 Chemical Composition of Experimental Diets

The test diets used in the experiment differed in chemical composition reflecting the differences in chemical composition of the ingredients used in their formulation (Table 8). The energy concentration in the ration (15.2, 12.2 and 13.92) MJ, ME/kg DM was slightly higher compared to that suggested by NRC (1989) for lactating dairy cows due to the nature of the diet used in the study. The diets were limited to one energy source and two oil seed cakes due to constrain of budget. The CP percent in the diet was within the accepted range except for control diet where protein content was lower (11.1%) due to low CP percent in maize bran (MB). The CP percent in CSC and SSC based diets were (19.3 and 17.2%) (Table 8). Ruminants need a minimum of 5% CP in their food to maintain body weight, whereas potential milk yield in dairy cattle is limited when the CP percent content in the feed fall below 12% (Pando, 1999).

Table 8: Chemical composition of experimental diets

Parameters	Control diet	CSC (Diet 1)	SSC (Diet 2)
DM%	97.6	97.9	97.6
CP%	11.1	19.3	17.2
Ash%	4.46	6.50	4.95
EE%	5.88	8.11	7.38
NDF%	32.3	50.4	54.6
ADF%	8.25	32.6	43.6
MJ (ME/kg)	15.2	12.2	13.9

4.3 Effect of Breed and Treatment on Dry Matter Intake, Milk Yield and Composition

4.3.1 Effect of breed on dry matter intake, milk yield and composition

No difference in DMI was observed during feeding Napier grass (*Pennisetum*

purperium) (Table 9). Even though the animals were fed Napier grass to supply 1% they only consumed 0.7% BW**.** The possible reason may be due to lower DM content in Napier grass during rain season when the grass was lush with low DM content and also the refusal were mainly stems that were left behind due to improper chopping and low palatability. The intake of concentrate was similar between treatments. This was expected because they were offered an allowance to meet 1.2% BW and they consumed all of it.

The mean milk yields was significantly ($P<0.05$) higher in Friesian cows (8.36 kg/day) than in Ayrshire cows (5.85 kg/day) whereas the milk from Ayrshire breed on average had significantly ($P<0.05$) higher BF and CP percent, and at the same time insignificant higher SNF and TS percent ($P>0.05$). The lower value of milk yields in Friesian and Ayrshire compared to what has been reported (Hatungumukama *et al.*, 2008) was contributed to the low DMI from Napier grass. Some of the animals were probably not fed enough concentrates to meet their potential production because they were restricted to blanket concentrates supplementation. Within the same treatment diets as well as in the control, daily average milk yield of cows differed significantly ($P<0.05$) between the two breeds. The higher milk yield by Friesian cows compared to Ayrshire cows and low percentage of fat, protein, SNF and TS in Friesian breed is supported by the study conducted by Galina *et al*. (1994) between Friesian and Jersey breed. The lower value of CP percent in milk from Friesian breed might be due to high volume of milk that contributed to lower values of milk component as reported by Aloka (1997). Galina *et al*. (1994) also showed that Ayrshire breed produced milk of higher fat content than Friesian cows.

The average fat content of the milk, 2.99% in Friesian and 3.67% in Ayrshire was within normal range of 3% BF as reported by Harvey and Hills (1967). The results were consistent with those of Aloka (1997) who reported that Friesian produce milk with low BF content. Higher BF value in Ayrshire breed might be due to greater uptake of LCFAs by the mammary gland and lower compensatory reduction in *de novo* FA synthesis as reported by Aldrich *et al*. (1997) and Avila *et al*. (2000). The mean value of TS percent in Friesian and Ayrshire cows were 10.13 and 11.28 % respectively. The results are similar to 11.19 ± 1.43% as reported by Sezgin and Kocak (1982) and Dogan *et al.* (2002) for Brown-Swiss.

Table 9: Effect of breed and treatments on dry matter intake, milk yield (kg/ day) and milk composition

Dry matter Intake (DMI) (kg/day) for Napier grass

Breed	MB (control)	CSC (diet I)	SSC (diet II)	Mean breed
Friesian	5.47 ± 0.33	5.48 ± 0.33	5.55 ± 0.33	5.50 ± 0.33
Ayrshire	5.58 ± 0.33	5.47 ± 0.33	5.51 ± 0.33	5.49 ± 0.33
Mean diet	5.48 ± 0.23	5.48 ± 0.23	5.43 ± 0.23	

Dry matter Intake (DMI) (kg/day) for concentrates

Friesian	5.77± 0.18	5.95± 0.19	5.81± 0.19	5.84± 0.19
Ayrshire	5.80± 0.18	5.80± 0.19	5.87± 0.19	5.84± 0.19
Mean diet	5.79± 0.18	5.90± 0.19	5.84± 0.19	

Milk yield (kg/day)

Friesian	5.78^{b} ± 1.29	9.67^{a} ± 1.29	9.62^{a} ± 1.29	8.36^{a} ±1.29
Ayrshire	4.95^{b} ± 1.29	6.83^{ab} ± 1.29	5.77^{b} ± 1.35	5.85^{b} ± 1.31
Mean diet	5.37^{b} ± 1.29	7.99^{a} ± 1.29	7.70^{a} ± 1.32	

Milk density (LR)

Friesian	27.87 ± 2.81	28.06 ± 2.81	27.67 ± 2.81	27.55 ± 2.81
Ayrshire	28.25 ± 2.81	28.16 ± 2.81	27.93 ± 2.07	28.11 ± .56
Mean diet	28.06 ± 2.81	28.11 ± 2.81	27.80 ± 2.44	

Milk protein (CP%)

Friesian	2.11^{b} ± 0.35	2.96^{a} ± 0.35	2.47^{ab} ± 0.35	2.51^{b} ± 0.35
Ayrshire	2.93^{a} ± 0.35	3.39^{a} ± 0.35	3.18^{a} ± 0.36	3.17^{a} ± 0.35
Mean diet	2.52± 0.35	3.18 ± 0.35	2.83 ± 0.35	

Butter fat (BF%)

Friesian	2.71^{b} ± 0.41	3.34^{a} ± 0.41	2.92^{ab} ± 0.41	2.99^{b} ± 0.41
Ayrshire	3.44^{a} ± 0.41	4.02^{a} ± 0.41	3.55^{a} ± 0.43	3.67^{a} ± 0.41
Mean diet	3.08 ± 0.41	3.68 ± 0.41	3.24 ± 0.42	

Solid not fat percentage (SNF%)

Friesian	6.39±0.85	7.00±0.85	7.74±0.85	7.04±0.85
Ayrshire	7.19±0.85	8.03±0.85	7.64±0.89	7.64±0.86
Mean diet	6.79±0.85	7.52±0.85	7.69±0.87	

Total solid percentage (TS%)

Friesian	9.23 ± 1.25	11.23 ± 1.25	9.94 ± 1.25	10.13 ± 1.25
Ayrshire	10.51 ± 1.25	12.11 ± 1.25	11.23 ± 1.31	11.28 ± 1.27
Mean diet	9.87 ± 1.25	11.67 ± 1.25	10.09 ± 1.28	

Note: a,b,c,- Means with different superscript within the same row are significantly different ($P < 0.05$) a,b,c

4.3.2 Effect of dietary treatment on milk yield and composition

Results in Table 9 showed that between treatments, cows fed diets containing CSC and SSC had significant higher milk yield ($P<0.05$) than those fed diets containing MB (7.99 and 7.70 vs 5.37 kg/day) and insignificant higher milk fat percent (3.68 and 3.24 versus 3.08%). Similar results have been reported earlier by Jenkins (1993) and Palmqust *et al.* (1993) who observed a depression in milk fat of cows fed diets containing high amount of fermentable starch. The study shows that supplementing dairy cow ration with high energy concentration yielded milk with low BF percent. The results were not in agreement with those obtained by Lalman *et al*. (2000) who observed an increased in BF percent when energy allowances in ration of dairy cows was increased. Other workers reported no differences in dietary energy on milk composition (Lowman *et al*., 1979). The results obtained from the study indicated that variation in milk composition due to dietary energy supplement might be due to the source of energy in cow's diet, chewing time, particle size and buffers as reported by Emery (1988) and Jelec (1990).

Supplementation with oil seed cakes produced milk with slightly higher CP percent but did not affect SNF percent. The increased milk yield as well as CP percent with dietary oil seed cake supplementation could be due to the fat and CP content in the oil seed cake which ranged from 2.8 to 18.8% and 14.1 to 46.2% respectively (Table 4). Study conducted by DePeter *et al.* (1989) indicated that adding fat to dairy ration reduce the energy available to the microbial population for growth which alter microbial N utilization in the rumen.

Supplementing dairy cows with CSC based diet yielded milk with slightly higher values of CP (3.18%) compared to SSC based diet (2.83%) (Table 9). Increased in milk protein percentage has been reported earlier by previous researchers. Henderson *et al*. (1985) observed an increased in milk protein percentage when dairy cows were supplemented with diet rich in soy bean meal. Bonsi and Osuji (1997) observed better combination of CSC and Sesbania in terms of roughage intake and N utilization respectively than in leucaena alone. Therefore high protein level in milk from cows that received diet containing CSC might be due to better combination of CSC and MB compared to SSC and MB. The increased CP percent in milk might be due to the differences in protein flow to the small intestine between the protein sources (CSC and SSC) which may account for differences in response to milk protein percentage as observed by Spain *et al*. (1984) between corn gluteal meal and fish meal. The level of anti-nutritional factors in these two oil seed cake, gossypol in CSC, high CP and chlorogenic acid in SSC; and the methods of extraction could also account for the differences in flow of N in small intestine. Gossypol inhibits the activity of digestive enzymes and reduces the palatability of diet whereas chlorogenic acid inhibits the activity of hydrolytic enzymes. Therefore further study concerning oil seed cake and flow of its protein in the small intestine should be conducted. The lower CP percent in diet rich in SSC was observed earlier by researchers. Zhan *et al*. (2005) observed a depression in CP, BF and TS percent in lactating dairy cows fed diet rich in SSC while there was no effect on milk yield.

4.3.3 Interaction effect of breed and treatment on milk yield and composition

Supplementation with CSC and SSC based diet to Friesian breed significantly increased ($P<0.05$) milk yield while supplementation to Ayrshire breed had no significant effects. Supplementation of CSC based diet produced milk that had significantly higher CP and BF percent ($P<0.05$) in Friesian breed than those supplemented with maize bran alone while there was no effect in Ayrshire breed. Amount of nutrients in SSC could account for the quantity of milk produced and also Ayrshire cows may have reached the potential for milk production such that above the quantity of feed offered no more milk could be produced. It was apparent that the two breeds had almost the same DMI (CSC and SSC) but Friesian breed produced more milk probably due to differences in heritability between and within the animals that may account for the differences in animal efficiency as observed by Koots *et al.* (1994b) in beef cattle. This is in agreement with other workers who reported that on average Friesian breed have more milk than Ayrshire breed (Galina *et al*., 1994; Beaulieu and Palmquist, 1995).

Interaction between breed x treatments was not statistically significant ($P>0.05$) in terms of Lactometer reading (LR), SNF and TS percent. Harvey and Hills (1967) reported that milk with LR below 27 (1.027 specific gravity) might be adulterated with water. Basing on Harvey's LR standard, the values of all milk samples obtained were above the standard although milk from Friesian fed SSC had values close to the lower limit 27 (1.027 specific gravity). Normal milk has been reported to contain SNF 8.5% (Harvey and Hills, 1967). The mean values of SNF from test diets were

within the accepted range (6.82 to 11.6%) except for Friesian breed (6.39%) fed MB based diet.

4.4 Effect of Breed and Treatment on Fat Acid Composition Percent in Butter Oil

4.4.1 Effect of breed on fat acid composition percent in butter oil

The MFFA composition from the two breeds is shown in Table 10 and in Appendix I. Regardless of the diet the content of SCFAs were slightly higher (4.94%) for Ayrshire than that of Friesian cows (4.34%). This might be due to higher values of C4:0 in Ayrshire breed than in Friesian breed in all test diet with lower values of C6:0 to C10:0 as reported by Glass *et al*. (1967). Friesian breed had slightly lower values (12.73 vs 14.61 %) of MCFAs and slightly higher values (82.98 versus 80.43%) of LCFAs than Ayrshire breed but the difference was not statistically significant (P>0.05). Study conducted by Reklewska *et al.* (2005) indicated that milk from Black and White, Polish Red and Simmental breeds contained higher proportion of butyric acid. German (2002) identified butyric acid as one of the functional milk components due to its potent antimicrobial and anti-carcinogenic effect. Butyric acid is a unique feature of milk fat from ruminant animals; it inhibits cell growth and reduces differentiation in a wide-spectrum of cancer cell lines including those of the breast and colon in humans (Parodi, 1999).

Table 10: Effect of breed on fatty acid composition percent in butter oil

Fatty acids	Friesian	Ayrshire	P
Short chain fatty acids (C4:0 to C10:0)	4.34±0.91	4.94±0.91	NS
Medium-chain fatty acids (C11:0 to C16:1)	12.73±1.78	14.61±1.78	NS
Long-chain fatty acids (C17:0 to C20:5n3)	82.95±2.55	80.43±2.55	NS
Saturated fatty acids	42.48±5.26	53.91±5.26	NS
Unsaturated fatty acids	57.52±5.26	46.08±5.26	NS
Mono-unsaturated fatty acids (MUFA)	31.06±4.81	26.84±4.81	NS
Poly-unsaturated fatty acids (PUFA)	26.46±4.95	19.23±4.95	NS

Note: NS-Not significant (P>0.05)

The differences in proportion of SCFAs and MCFAs in both breeds agreed with other comparisons between Holstein and Jersey breeds (Beaulieu and Palmquist, 1995; and Morales *et al*., 2000). Milk fat from Ayrshire cows contains more SCFAs synthesized *de novo* within the mammary gland and fewer LCFAs that are taken up directly from non-essential FAs compared to Friesian as reported by Stull and Brown (1964). Walstra and Jenness (1974) showed that all SCFAs and MCFAs (C4:0 to C14:0), and half of the palmitc acid (C16:0) in milk fat are synthesized *de novo* in the mammary gland. Ayrshire breeds have slightly higher proportions ($P<0.05$) of MCFAs (14.61%) compared to Friesian breed (12.73%) but the fraction of C16:1 was lower in Friesian breed than in Ayrshire breed probably due to mammary enzyme Stearoyl Co-A desaturase being higher in Ayrshire breed compared to Fresian breed as reported by Bitman *et al.* (1996); DePeters *et al*. (1995); and Drackley *et al*. (2001) between Friesian and Jersey cows. Preliminary work done by Medrano *et al*. (2003) showed differences between breeds with respect to the activity of the mammary enzyme stearoyl Co-A desaturase. Stearoyl Co-A desaturase acts on a trans-fatty acid (vaccenic) and oxidizes C16:0 and C18:0 to C16:1 and C18:1; and it is involved in CLA production. Mammary desaturase activity *in vitro* was reported to use both C16:0 and C18:0, but not C14:0, as substrates (Moore and Christie, 1990).

Mean proportions of linoleic acid (C18:2n6t) for Friesian was significantly higher ($P<0.05$) compared to Ayrshire when supplemented with CSC and MB whereas supplementing Ayrshire breed with SSC resulted in a slightly higher proportion of C18:2n6t compared to Friesian (Appendix 1). The results agree with that of Morales *et al*. (2000) who reported higher values of linoleic acid (C18:2n6t) in Ayrshire

breed. Higher proportion of α-linolenic acid in the milk fat from Friesian was observed earlier by Avila *et al.* (2000). Komprada *et al.* (2000) observed higher levels (1 to 1.3%) from Czech Pied × Ayrshire × Red Holstein cattle. Although C20:5n3 were present in milk fat in negligible amount (Appendix 1) they can not be ignored because they are important components of the cell membrane associated process. However, it should be noted that an uncontrolled increased intake of these FAs may potentially result in an elevated risk of exposure of PUFAs to auto oxidation toxic products (Valivet, 1997).

4.4.2 Effect of treatment diet on fatty acid composition percent in butter oil

The mean proportion of SCFAs (C4:0 to C10:0) in butter oil from diet based on CSC, SSC and MB are shown in Table 11. The mean proportion of SCFAs in CSC based diet was slightly lower compared to SSC and MB based diets. The lower SCFAs in CSC based diet might be due to reduction in *de novo* FA synthesis in the mammary gland caused by presence of high linoleic acid and UFAs in CSC as reported by Chouinard *et al.* (2001). Similar observations were reported by Tyrrell (2004) and Reveneau *et al*. (2005) in grazing dairy cows.

The mean proportion of MCFAs was slightly lower in CSC based diet (12.46%) compared to SSC (14.11%) and MB (14.45%) based diet but was not significantly different ($P>0.05$) (Table 11). The higher value of MCFAs in SSC and in MB than in CSC diet was contributed by reduced *de novo* FA synthesis when feeding CSC based diet. Study conducted by Chouinard *et al.* (2001) revealed that CSC oils are rich in linoleic acids which decrease *de novo* FA synthesis in the mammary gland. Dietary

seed cake supplementation provides additional LCFAs for milk fat synthesis; increased uptake of LCFAs by mammary glands inhibits the synthesis of SCFAs and MCFAs because of negative feedback inhibitions on Acetyl CoA carboxylase and increased incorporation of LCFAs into milk fat. This inhibition may be due to the formation of *trans*-isomers resulting from biohydrogenation of LCFAs in the rumen. In terms of human health, decreased SCFAs and MCFAs could improve milk FA profiles due to the potential effects of these FAs on increased total and LDL cholesterol concentrations in plasma as reported by Kris-Etherton (1997).

Lowering C14:0 and C16:0 in milk fat by supplementing dairy cow rations with long chain UFAs is reported to add healthy benefits to dairy products. The results from the current experiment produced similar changes in the FA profile and therefore are in agreement with scientific effort to improve the nutrient value of dairy products (Mansbridge and Blake, 1997; Maijala, 2000).

The proportion of C18:1 was variable in both the *cis* and *trans* isomers except for MB based diet. The proportion of *trans* C18:1 and C18:2 (Appendix 1) were highest in SSC as reported by Kelly *et al*. (1998). Increases in milk C18:1 and C18:2 can partly be explained by a reduction in ruminal biohydrogenation suggesting that both SSC and MB were inert in the rumen as reported by Fellner and Spears (2005). These proportions were desirable and were consistent with improved nutritional qualities associated with increased consumption of milk and milk products (Mansbridge and Blake, 1997; Maijala, 2000). Feeding MB alone led to higher linolenic acid content in milk fat (Appendix 1). The PUFAs, especially linolenic

acids in the oils are responsible for the objectionable flavour and poor stability (Evans *et al.,* 1971). Corn oil (maize) has 1% linolenic acid. Reports indicated that, high level of linolenic acid in soybean oil (7 – 8%) causes poor flavour stability in comparison to corn oil (maize) which has 1% of linolenic acid (Hammond *et al.,* 1972). The flavour and stability of the oil increases as the linolenic acid level decreases (Evans *et al.,* 1971). The low level of linolenic acid in corn oil (MB) which is responsible for good flavor might be the possible cause of the animal to consume more MB hence high level of linolenic acid in milk fat. Similar findings were reported by other workers (Gaynor *et al*., 1994; Jenkins and Lundy, 2001).

Dairy products are rich in CLA, a product synthesized in the rumen during biohydrogenation of linoleic acid. It is possible to influence the extent of ruminal biohydrogenation, the concentration of CLA absorbed and incorporated into milk fat by dietary manipulation as reported by Beaulieu and Drackley (2004) who observed a two fold increased CLA levels in milk fat when dairy cows were supplemented with diets rich in linoleic acid.

The mean proportion of LCFAs (84.53%) was not significantly higher in CSC based diet compared to SSC (80.09%) and MB (80.45%). Oil seed cake supplement yielded a slightly lower proportion of SFAs compared to MB supplement and at the same time the proportion of UFAs was higher. Addition of oil seed cake to the ration of dairy cows altered the FA profile in milk indicating the presence of the FAs in seed cakes as reported earlier by Grant and Kubik (1990) and Kelly *et al.* (1998). Previous study shows that CSC and SSC contain 15 to 20% and 49 to 58%; and 13.5 and 75%

of oleic acid and linoleic acid respectively (Palmquist, 1988 and Kelly *et al.*, 1998). The increase though not statistically significant in UFAs was due to the presence of high level of UFA in oil seed cake based diet that led to an increase in the amount of unsaturated LCFAs in milk fat as observed by Nielsen *et al*. (2005a).

Table 11: Effect of treatment diet on fatty acid composition percent in butter oil

Fatty acids (chain length)	MB (control)	CSC (diet I)	SSC (diet II)	Significant
Short chain fatty acids (C4:0 to C10:0)	5.12±1.12	3.00±1.12	5.81±1.12	NS
Medium-chain fatty acids (C11:0 to 16:1)	14.45±2.18	12.46±2.18	14.11±2.18	NS
Long-chain fatty acids (C17:0 to C20:5n3)	80.45±3.13	84.53±3.13	80.09±3.13	NS
Saturated fatty acids	56.20±6.44	43.19±6.44	45.20±6.44	NS
Unsaturated fatty acids	43.80±6.44	56.80±6.44	54.80±6.44	NS
Mono-unsaturated fatty acids (MUFA)	28.81±5.89	26.69±5.89	31.36±5.89	NS
Poly-unsaturated fatty acids (PUFA)	14.99±6.06	30.11±6.06	23.43±6.06	NS

Note: NS-Not significant (P >0.05)

Supplementing dairy cows with SSC resulted in a non significant higher proportion of MUFA (31.36%) and slightly lower proportion of PUFA (23.43%) compared to CSC whereas CSC supplement yielded a slightly lower proportion of MUFAs (26.69%) and slightly higher proportion of PUFA (30.11%) as compared to MB 28 and 14.99% in MUFAs and PUFAs respectively. The results were similar to previous reports except for CSC and MB supplement. Work done by Contarin *et al.* (1990) indicated an increase in concentration of MUFAs, PUFAs and LCFAs in milk when lactating dairy cows were supplemented with oil seed cakes. The lower proportion of MUFAs in CSC based diet might be due to presence of cyclopropenoic FAs in CSC which are strong inhibitors of mammary delta-desaturase activity which convert

trans 11 C18: to *cis-9, trans-11* 18:2 and also convert C18:0 to C18:1 as reported by Jensen (2002).

4.4.3 Interaction effect of breed and treatment on fatty acid composition percent in butter oil

The interaction effects of the three diets and the two breeds on the FA composition and on the yields of the major FAs percent in butter oil are given in Table 12 and Appendix 1.

4.4.3.1 Short-chain fatty acids

Supplementing Friesian breed with sunflower seed cake (SSC) produced milk fat with significantly higher value of SCFA (5.98%) than supplementing with CSC (3.31%) and MB (4.08%) while supplementing Ayrshire breed with MB resulted into significantly higher ($P<0.05$) values of SCFA (6.14%) than supplementing with SSC (5.63%) and CSC (3.03%) (Table 12). Reduction in SCFAs in milk fat from cows due to feeding supplemental seed cake was reported earlier by Zhan *et al*. (2005) in buffaloes fed tallow. CSC and SSC based diet containing long- chain UFAs which act as partial inhibitors of the synthesis of the SCFAs and MCFAs (Tyrrell, 2004). CSC based diet favour the activity of Steroyl CoA in both breeds since CSC is rich in palmitic acid (C16:0) and stearic acid (C18:0) and Steroyl CoA converts these acids to C16:1 and C18:1. The results indicated that the activity of Steroyl CoA increased when feeding CSC based diet to both breeds. The methods of extraction could also impart a lot of oil which have inhibitory effect on SCFA synthesis. Although SCFA and MCFA were found in milk fat their presence have certain characteristics which

can explain their presence in milk fat. SCFA and MCFA in milk fat are absorbed as non esterified FAs into portal blood stream of young animals and are metabolised rapidly in the liver (Noble, 1978). Hence they are able to make direct and rapid contribution to the energy metabolism of the new born calf.

Table 12: Effect of breed and treatment on fatty acid composition percent in butter oil (n=24)

Breed	MB (control)	CSC (Diet II)	SSC (diet II)
Short-chain fatty acids (C4:0 to C10:0)			
Friesian	4.08±1.58	3.31±1.58	5.98±1.58
Ayrshire	6.14±1.58	3.03±1.58	5.63±1.58
Medium-chain fatty acids (C11:0 to C16:1)			
Friesian	11.89±3.08	10.57±3.08	15.72±3.08
Ayrshire	17.01±3.08	14.34±3.08	12.50±3.08
Long-chain fatty acids (C17:0 to C20:5n3)			
Friesian	84.04±4.42	86.49±4.42	78.32±4.42
Ayrshire	76.86±4.42	82.57±4.42	81.87±4.42
Saturated fatty acids			
Friesian	43.1^{b} ± 9.10	35.44^{b} ± 9.87	48.89^{ab} ± 9.10
Ayrshire	69.30^{a} ± 9.10	50.93^{a} ± 9.10	41.51^{b} ± 9.10
Unsaturated fatty acids (UFA)			
Friesian	56.90^{a} ± 9.11	64.54^{a} ± 9.11	51.12^{a} ± 9.11
Ayrshire	30.70^{b} ± 9.11	49.15^{ab} ± 9.11	59.47^{a} ± 9.11
Mono-unsaturated fatty acids (MUFA)			
Friesian	33.93 ± 8.33	30.64 ± 8.33	28.61 ± 8.33
Ayrshire	23.69 ± 8.33	27.74 ± 8.33	34.10 ± 8.33
Poly-unsaturated fatty acids (PUFA)			
Friesian	22.97^{a} ± 8.57	33.92^{a} ± 8.57	22.50^{a} ± 8.57
Ayrshire	7.02^{b} ± 8.57	21.6^{ab} ± 8.57	23.63^{a} ± 8.57

Note:a,b,c,- Means with different superscript within the same row are significantly different (P < 0.05); a,b,c,- Within treatments and breed Means with different superscript within column are significantly different (P < 0.05)

4.4.3.2 Medium-chain fatty acids

CSC supplement yielded slightly lower proportion of MCFAs in Friesian (10.57%) than SSC and MB based diet (15.72 and 11.88%) respectively while SSC supplement in Ayrshire yielded lower proportion (12.50%) of these FAs compared to CSC supplement (14.34%) and MB based diet (17.01%) but the difference were not significant (P>0.05) (Table 12). The results were similar to that reported by Chouinard *et al.* (2001) who reported a lower proportion of MCFAs in milk from lactating dairy cows supplemented with CSC diets due to reduced *de novo* fatty acid synthesis. Supplementing lactating dairy cows with CSC and SSC yielded a slightly lower proportion of C13:0 and C16:1 whereas MB supplement yielded slightly higher values of these FAs at the same time SSC supplement increased the proportion of C15:0 in both breeds (Appendix 1). Preliminary work done by Zhan *et al.* (2005) indicated that oilseed cake supplement decreases concentration of saturated short chain and medium chain FAs in milk from lactating dairy cows. The study indicated that feeding Friesian and Ayrshire cows diet rich in CSC and SSC respectively influenced the activity of Steroyl CoA desaturase enzyme in both breeds.

Supplementation with CSC decreased SCFA of low MP and slightly increased LCFA of high MP compared to SSC and MB based diet (Table 12). Increased LCFAs of high MP largely cis-C18:1 originating from intestinal absorption and from increased desaturation of C18:0 might make milk fat less fluid as observed by Drackley *et al.* (2001). Friesian breed have more UFA and PUFA compared to Ayrshire breed (Table 12). Therefore butter made from Friesian breed will be more spreadable than butter from Ayrshire breed due to the presence of higher concentration of UFA in Friesian

milk which decreased the MP of butter as reported by Latha (2004).

4.4.3.3 Long-chain fatty acids

Supplementing dairy cows with CSC produce a slightly higher but not statistically significant (P>0.05) proportion (86.48%) of LCFAs in Friesian breed whereas SSC supplement and MB based diet yield a lower values (78.32 and 84.04%) (Table 12). Oil seed cake supplementation to dairy ration resulted into an increased concentration of LCFA, MUFA and PUFA and decreased proportion of SCFA in milk as reported by Zhan *et al*. (2005). Study conducted by Stryer (1985) indicated that LCFAs originated almost exclusively from the feed supplemented to cows and on mobilised of fat from body adipose tissues. The composition of the LCFAs in milk fat does not reflect the FA composition of the feed because the ingested fat undergoes considerable modifications in the animal body. In the rumen, the FAs undergo isomerisation and hydrogenation (Sejrsen *et al*., 2007). The rumen modifications depend to a large extent on the FAs composition of the fat in the ration, but other factors, such as starch and fiber content, that affect rumen fermentation also play a role. The modifications in the mammary gland are catalyzed by desaturation enzymes, especially delta-9-desaturase that converts *trans-11* C18:1 to *cis-9*, *trans-11* C18:2, the most abundant CLA and C18:0 to C18:1 (Sejrsen *et al.*, 2007).

4.4.3.4 Saturated fatty acids

Saturated fatty acids (35.44%) were lower in Friesian fed CSC based diets than those fed MB based diets (43.1%) and SSC (48.89%) whilst in Ayrshire they were lower for those fed SSC (41.51%) than for those fed CSC (50.93%) and MB based diets

(69.30%) (Table 12). The lower proportion of SFAs in Friesian and Ayrshire breed fed CSC and SSC respectively was attributed by Stearoyl CoA in small intestinal wall which might be limiting in response to oil seed cake supplement as observed by Banks (1987) when supplementing dietary lipid to dairy cows. Stearoyl CoA converts C18:0 to C18:1. The later FA is an abundant FA resulting from ruminal biohydrogenation of dietary UFAs.

The presence of SFA in milk from dairy cows was contributed by UFA in oil seed cakes which are converted into SFA by ruminal microorganism as reported by La Count *et al*. (1998). Findings from Pfeuffer and Schrezenmeir (2000) suggested that lower proportion of SFA in milk fat seems to be beneficial for the human healthy because of their negative role in arteriosclerosis. Within the group of SFAs Pentadecanoic acid (C15:0) accounted for the higher proportion in the milk fat of both breeds. However, the result shows a lower proportion ranging between 0.13 to 2 and 31% of stearic acid (C18:0) as observed by Pesek *et al*. (2005). From these results it can be concluded that in order to produce low level of SFAs, Friesian cows should be fed CSC while Ayrshire cows should be fed SSC.

Cottonseed cake based diet produced higher proportion of UFA in Friesian cows (64.54%) while SSC based diet produced higher proportion of these FAs in Ayrshire cows (59.47%). Similarly, MUFA were higher in Ayrshire cows fed SSC based diets (34.10%) while in Friesian cows it was higher (33.93%) for those fed MB based diet. CSC based diet produced higher proportion of PUFA in Friesian cows while SSC based diet produced higher proportion of these FAs in Ayrshire. The proportion of

PUFA (34.30%) was significantly higher (P<0.05) in Friesian breed when supplemented with CSC as observed by Kudrna and Marounek (2006) in animals supplemented with rape seed cakes and extruded soya bean. Within UFAs, Elaidic acid (C18:1n9t) showed a significant (P<0.05) highest respective proportion (21.91 versus 7.38%) in Friesian and Ayrshire breed when fed CSC. The proportion of oleic acid (C18:1n9c) were significant higher (P<0.05) in Ayrshire cows versus Friesian cows (12.62 vs 2.71%) when fed SSC diet. The increased Oleic acid in Ayrshire cows indicated that mammary desaturase activity is higher in Ayrshire than in Friesian cows (Banks, 1987). Steroyl CoA desaturase enzyme oxidised C16:0 and C16:1 to C18:0 and C18:1. Statistically significant differences in the proportion of oleic acid between Holstein and Jersey cows were reported earlier by White *et al.* (2001).

4.5 Melting Point Range of Butter Oil

Melting point range of the BF is the temperature at which the solid BF changes from solid to liquid (oil). In the present study the term MP range was used which is the interval from the temperature at which a solid butter sample begins to melt (to form liquid) and the temperature at which melting is complete. Results of MP range from the present study are shown in Table 13.

Table 13: Melting point of butterfat of milk from cows fed CSC, SSC and MB based diets

Breed	CSC	SSC	MB
Friesian	39.5 ºC – 41.5 ºC	39.5 ºC – 40.5 ºC	39.5 ºC – 41.0 ºC
Ayrshire	39.5 ºC – 41.0 ºC	39.5 ºC – 41.0 ºC	39.5 ºC – 40.5 ºC

Note: CSC – Cotton seed cake; SSC – Sunflower seed cake and MB- Maize bran

Determination of MP was conducted in MP apparatus. The MP range of milk BF in

Friesian cows (39.5 – 41.5 ºC) and (39.5 – 41.0 ºC) was slightly higher compared to that of Ayrshire (39.5 – 41.0 ºC) and (39.5– 40.5 ºC) when supplemented with CSC based diet and MB based diet respectively (Table 13). The results agree with the previous researcher (Drackley *et al.,* 2001) who observed higher MP of milk fat in Friesian cows than in Jersey cow. The experiment revealed that, BF from Ayrshire cows will have slightly low MP range compared to BF from Friesian cows. The lower MP range in BF could be due to the presence of SCFAs and MCFAs in Ayrshire cows compared to Friesian cows (Table 12) as observed by Morales *et al.* (2000). The melting properties of milk fat revealed in this experiment shows that Ayrshire milk fat would be liquid at low (refrigerator) temperatures because of the presence of more SCFAs and MCFAs of low MP as reported by Drackley *et al.* (2001) between Jerseys and Holsteins breeds.

Milk fat from Friesian had higher concentrations of LCFAs of high MP than did milk fat from Ayrshire (Table 10 and 12). The results agree with the previous researchers. Beauleu and Palmquist (1995) and Drackley *et al*. (2001) observed a higher proportion of LCFAs in Friesian breeds compared to Jersey cows and MP of 71 to 76 ˚C. Supplementing Ayrshire breed with SSC yielded BF with slightly higher MP range (39.5 – 41.0 ºC) compared to BF from Friesian breed (39.5 – 40.5 ºC). The lower MP range between breeds when fed SSC could be due to the lower rate of conversion of C16: 0 to C16:1 and C18:0 to C18:1 in Friesian than in Ayrshire by mammary enzyme stearoyl-CoA desaturase. Stearoyl-CoA desaturase might be lower in Friesian breed than in Ayrshire breeds as reported by DePeters *et al*. (1995) between Friesian and Jersey cows.

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

1. The results from the present study show that feeding dairy cattle concentrates containing different oil seed cake supplement improved yield traits (milk yield, BF and CP percent) and the content of basic milk constituents including TS and SNF percent compared to the control diet. The study shows that fat content in Friesian and Ayrshire breeds have been significantly influenced by feeding oil seed cakes. According to the results obtained in this study, it is possible to say that when the milk fat content was higher, protein and TS contents were also higher and that milk fat, protein and TS percent were higher in Ayrshire breed than in Friesian breed.

2. It follows from the results of the present experiment that milk composition and milk FA composition are not significantly affected by breed and type of diet. The milk fat (BF) from the dairy cows has unique functional properties influenced by nutritional manipulation. The result obtained on oil seed cake supplement indicated that it is not possible to substantially alter FA profile in BF using the two oil cakes.

3 From the study it can be concluded that in order to produce low level of SFAs, Friesian cows should be fed CSC while Ayrshire cows should be fed SSC.

4 Feeding CSC to lactating dairy cows yielded milk fat with more LCFAs and PUFAs compared to cows fed MB and SSC based diet and at the same time cows

that received SSC based diet produced milk fat that have more MUFA compared to cows fed CSC and MB based diet

5 Seed cake supplementation resulted in changing FA composition that slightly affected the MP of butter directly and indirectly, by influencing the array of FAs in the various positions of the glycerol backbone, the determinant structure of crystallization behaviour and hardness of milk fat and MP in milk fat.

5.2 Recommendations

1. Further studies concerning breeding focusing on the desaturase gene to produce milk with decreased levels of SFA may be beneficial to determine if genetic differences among breeds and individual animals are translated into ratios of SFA and UFAs.

2. Further study concerning oil seed cake supplementation to dairy ration and flow of its protein in small intestinal tract should be conducted so as to compare the results obtained in this study due to feeding oil seed cake supplement.

3. Determination of FA composition of different oil seed cake so as to be able to compare their effect on the composition of milk from different *bos Taurus* since those which will contribute to high UFAs will be preferred for feeding the animals since milk fat with higher UFAs are likely to fetch higher price.

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APPENDICES

Appendix 1: Effect of breed and treatment on fat acid composition in butter oil from milk of cows fed diets with CSC, SFC and MB

Short-chain fatty acids (SCFAs)

Item		CSC		SSC		MB	
		FR	AY	FR	AY	FR	AY
Butyric	(C4:0)	2.94	3.03	3.34	4.88	4.69	5.59
Caproic	(C6:0)	0.00^{b}	0.00^{b}	0.85^{a}	0.50^{ab}	0.00^{b}	0.00^{b}
Caprylic	((C8:0)	0.00	0.01	0.00	0.00	1.29	0.10
Capric	(C10:0)	0.00^{b}	0.01^{b}	0.01^{b}	0.77^{a}	0.01^{b}	0.04^{ab}

Medium-chain fatty acids (MCFAs)

Undecanoic	(C11:0)	0.01	0.22	1.01	0.00	0.02	0.01
Lauric	(C12:0)	0.09	0.07	1.17	0.09	0.02	0.16
Tridecanoic	(C13:0)	0.00^{b}	1.13^{a}	0.36^{ab}	0.00^{b}	2.84^{a}	3.34^{a}
Myristic	(C14:0)	3.03^{a}	1.72^{a}	1.67^{a}	1.76^{a}	0.24^{b}	1.65^{ab}
Myristoleic	(C14:1)	0.06	0.20	0.22	0.49	0.00	0.20
Pentadecanoic	(C15:0)	3.2	0.51	5.62	4.59	4.23	4.94
cis-Pentadecanoic	(C15:1)	1.39^{a}	0.56^{a}	0.07^{b}	0.66^{a}	0.00^{b}	0.48^{ab}
Palmitic	(C 16:0)	0.48^{a}	0.60^{a}	1.41^{a}	0.42^{ab}	0.09^{b}	1.00^{a}
Palmitoleic	(C 16:1)	0.81	1.95	0.39	0.49	2.12	1.52

Appendix I *cont.*

Long-chain fatty acids (LCFAs)

Heptadecanoic	(C17:0)	11.28	20.56	18.83	17.69	11.43	4.62
Cis-Heptadecanoic	(C17:1)	1.56	5.04	1.84	2.88	2.11	2.79
Stearic	(C18:0)	2.31	1.81	0.13	1.26	2.81	0.80
Elaidic	(C18:1n 9t)	21.91	7.38	14.34	13.37	3.04	11.33
Oleic	(C18:1n 9c)	4.27	5.23	2.71	12.62	5.49	4.74

Linolelaidic	(C18:2n6c)	0.10	0.44	1.41	5.23	5.13	2.03
Linoleic	(C18:2n 6t)	16.63	0.44	8.54	15.02	12.07	0.59
Arachidic	(C20:0)	3.06^{b}	2.77^{b}	0.93^{b}	2.12^{b}	1.43^{b}	6.61^{a}
Y- Linoleic	(C18:3n6)	0.71^{a}	0.60^{a}	0.52^{ab}	1.14^{b}	3.44^{a}	0.17^{b}
Cis-11-eicosanoic	(C20:1)	1.09	1.29	2.47	0.69	0.61	0.78
α-Linolenic	(C 18:3n3)	0.59	2.50	6.74	0.38	1.51	8.24
Heneicosanoic	(C21:0)	1.71^{b}	8.03^{a}	1.00^{b}	0.41^{c}	4.16^{a}	2.63^{ab}
Cis-11, 14- eicosadienoic	(C20:2)	0.72^{b}	0.57^{b}	0.63^{b}	0.73^{b}	13.28^{a}	0.90^{b}
Behenic acid	(C22:0)	10.80^{a}	12.88^{a}	6.98^{a}	0.84^{b}	2.77^{b}	6.34^{ab}
Cis-8,11, 14- eicosatrienoic	(C20:3n6)	0.37	0.72	0.66	0.12	0.00	0.09
Erucic	(C22:1n9)	0.24	1.47	5.94	0.21	2.74	0.00
Cis-11, 14.17- eicosatrienoic	(C20:3n3)	2.21^{a}	.18^{ab}	0.83^{b}	0.17^{b}	4.14^{a}	0.04^{b}
Arachidonic	(C20:4n 6)	1.98^{a}	0.00^{b}	2.24^{a}	0.00^{b}	3.39^{a}	0.00^{b}
Cis-13, 16- eicosatrienoic	(C22:2)	1.73	0.00	0.00	0.00	0.48	0.00
Lignoceric	(C24:0)	0.18	2.81	0.00	0.00	1.38	2.06
Cis-5, 8, 11, .17- eicosapentaenoic	(C20:5n3)	0.37$^{a\,b}$	0.00^{b}	1.21^{a}	0.00^{b}	1.68^{a}	2.42^{a}

Note: FR –Friesian; AY – Ayrshire;

a,b,c,- Means with different superscript letters within the same row are significantly different (P<0.05)

* – Significant at (P<0.05).

1 – Fat source: CSC- Cotton seed cake, SSC-Sunflower seed cake;

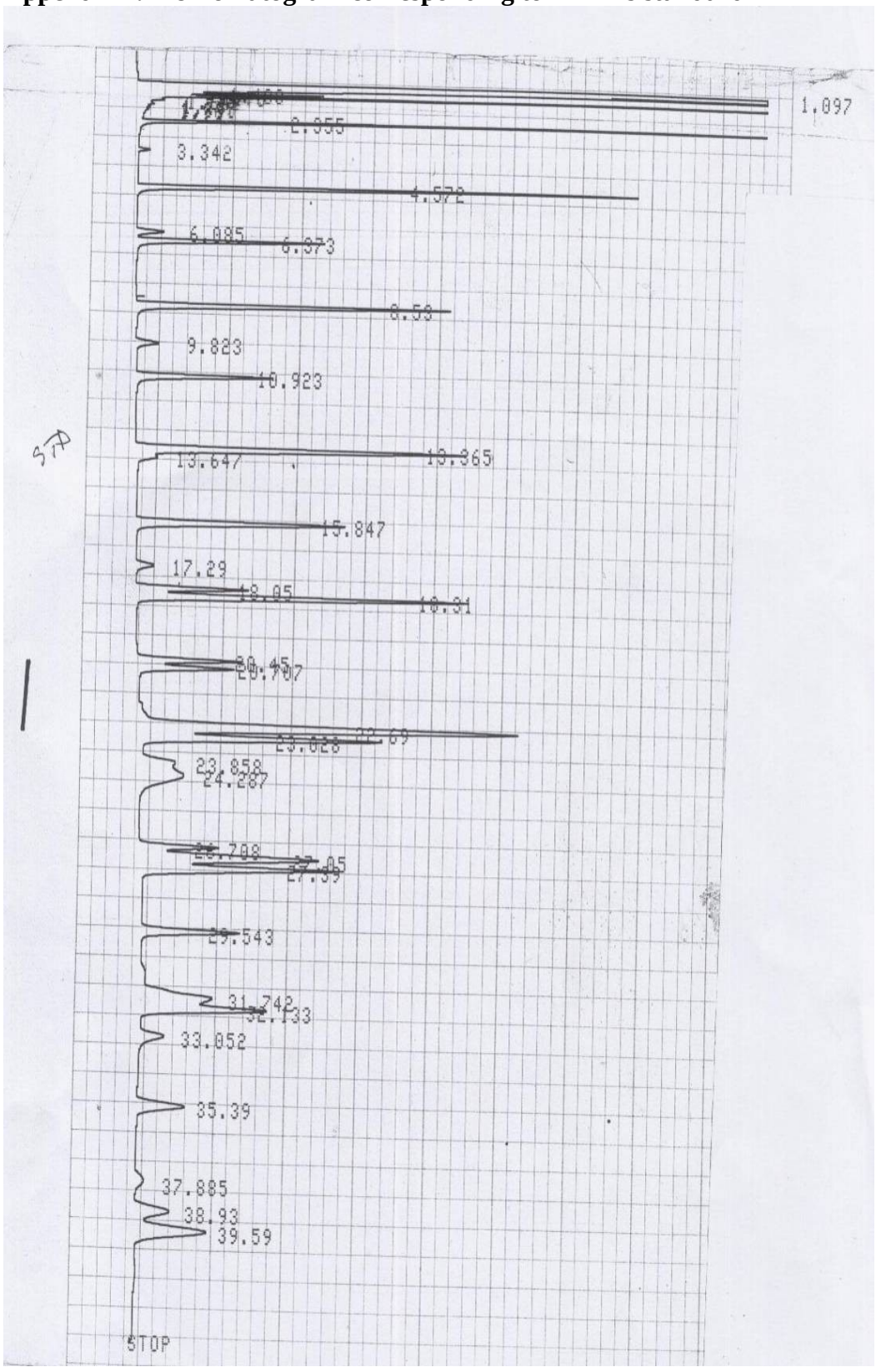
2 – C4:0, C6:0, C8:0, C10:0, C12:0, C14:0, C15:0, C16:0, C17:0, C18:0, C20:0, , C22:0, C24:0;

3 – C14:1, C15:1, C16:1, C17:1, C18:1t-9, C18:1c-9, C18:2n6c, C18:2n6t, C18:3n6, C20:1, C18:3n3, C20:2, C20:3n6, C22:1n9, C20:3n3, C20:4n6, C22:2, C20:5n3 (EPA);

4 – C14:1, C16:1, C18:1t-9, C18:1c-9, C20:1, C22:1n9,;

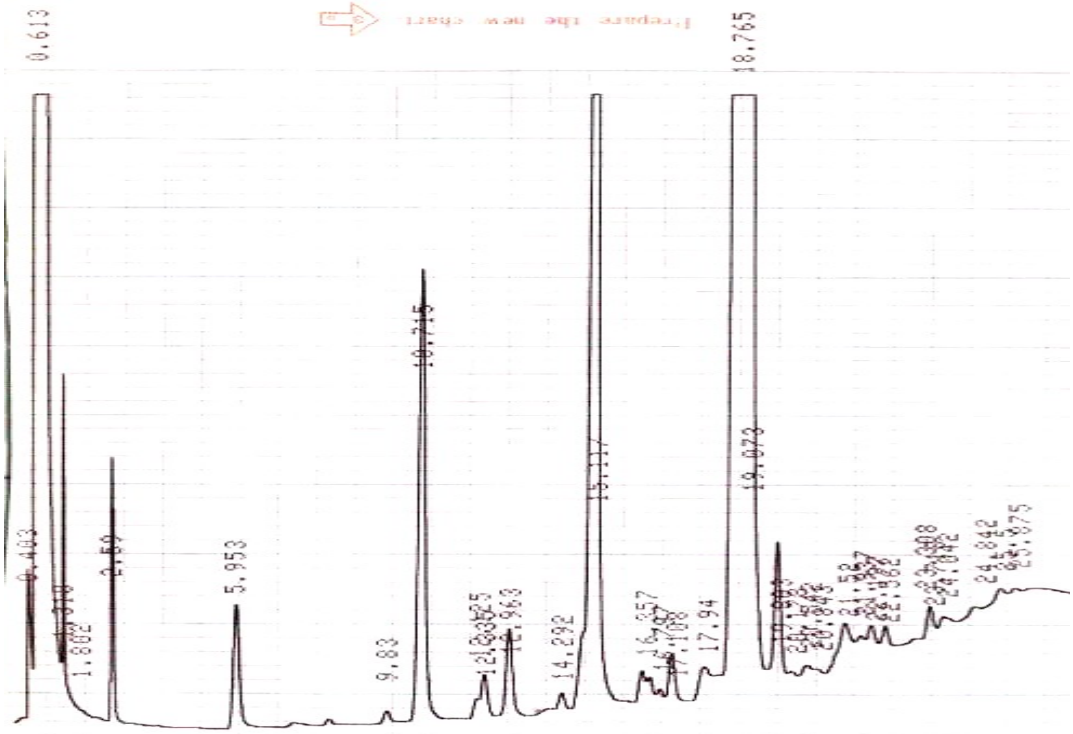
5 – C18:2n6c, C18:2n6t, C18:3n6, C18:3n3, C20:2, C20:3n6, C20:3n3, C20:4n6, C22:2, C20:5n3 (EPA)

Appendix 2: A chromatogram corresponding to FAMEs standard



A Chromatogram corresponding to the fatty acid methyl esters (FAME) standards

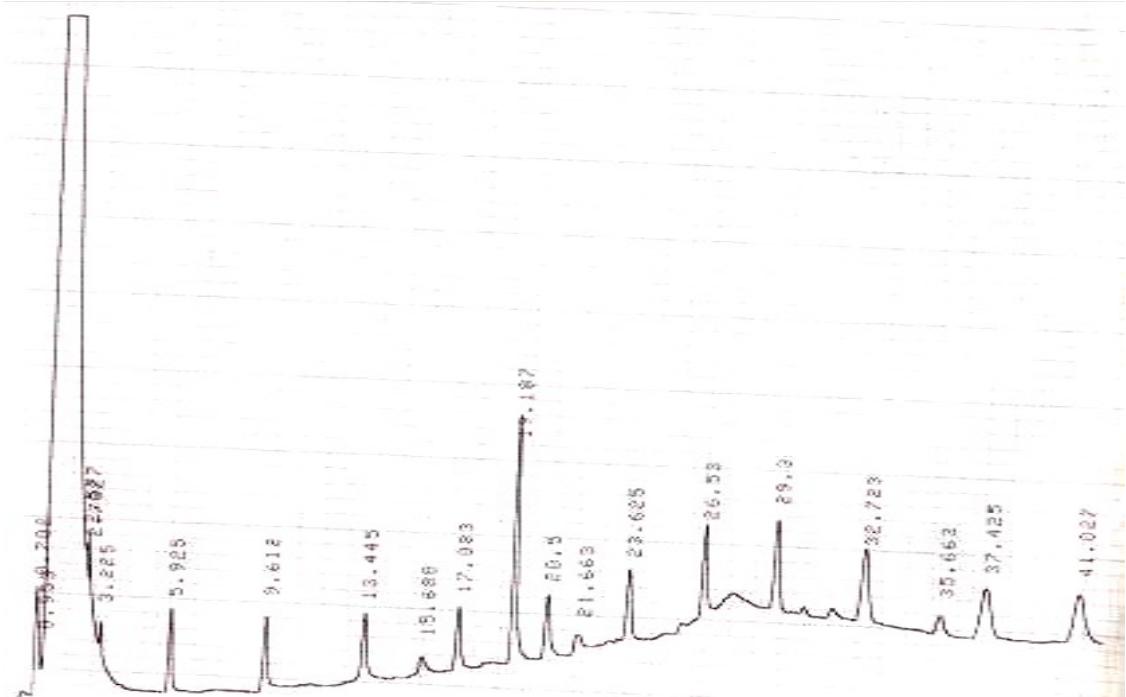
Appendix 3: Chromatograms corresponding to the FAMEs (standards) from milk of cows that received different diets.

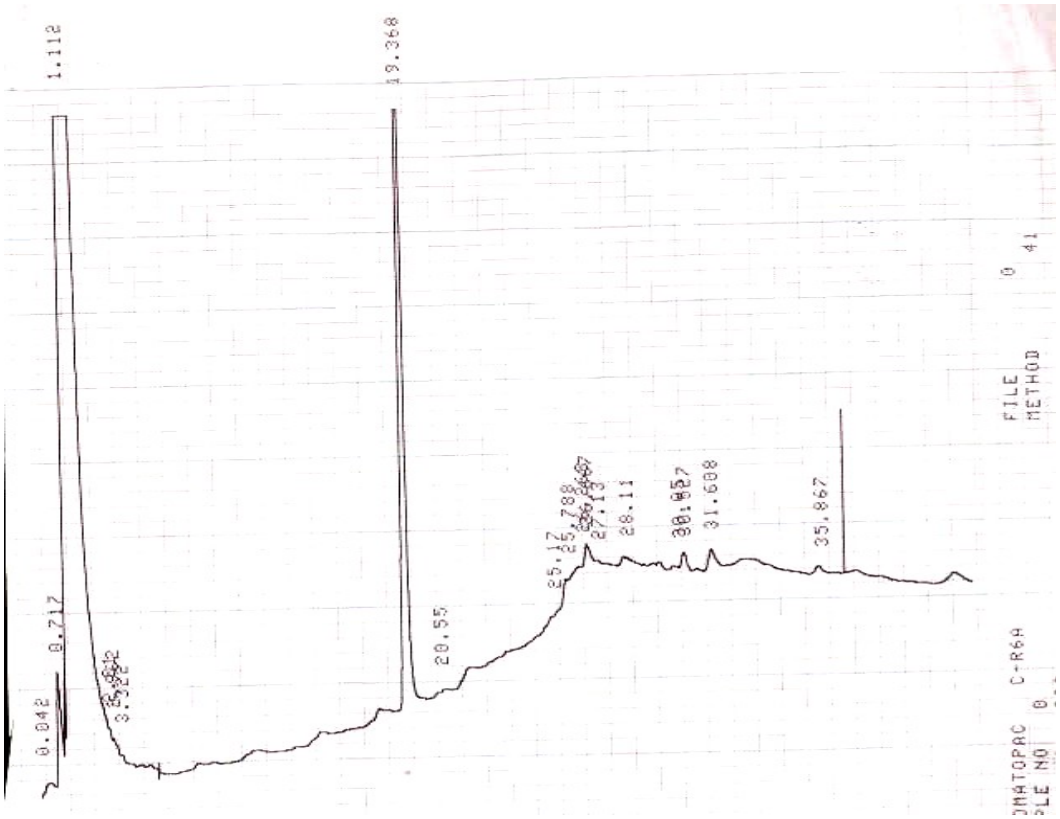


A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

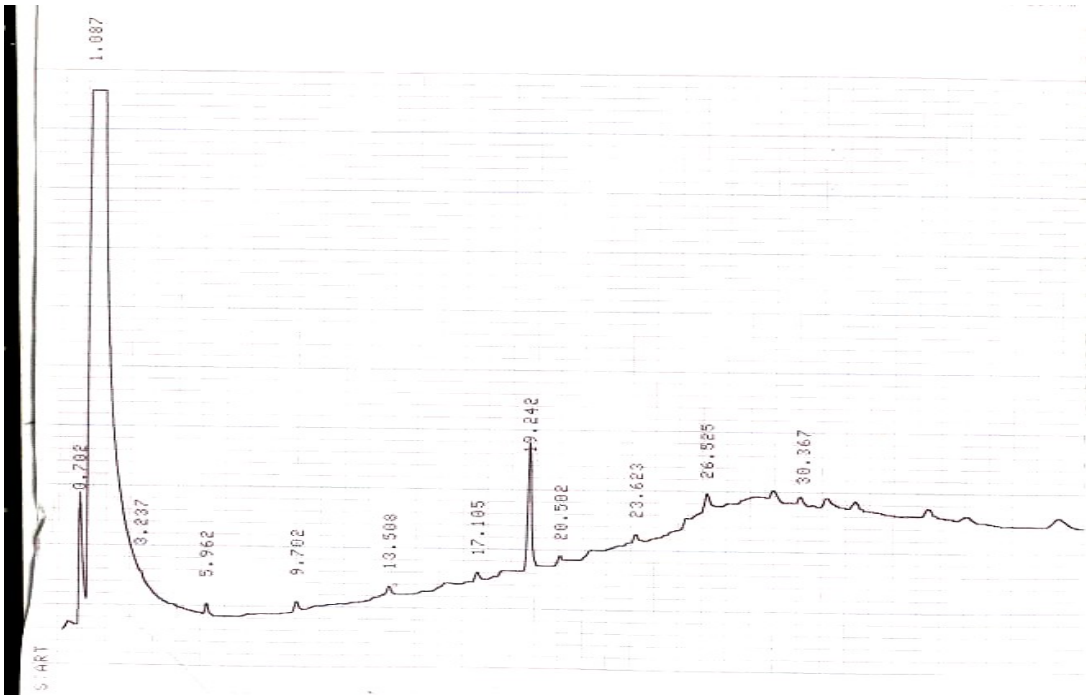
A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

Appendix 3 cont.



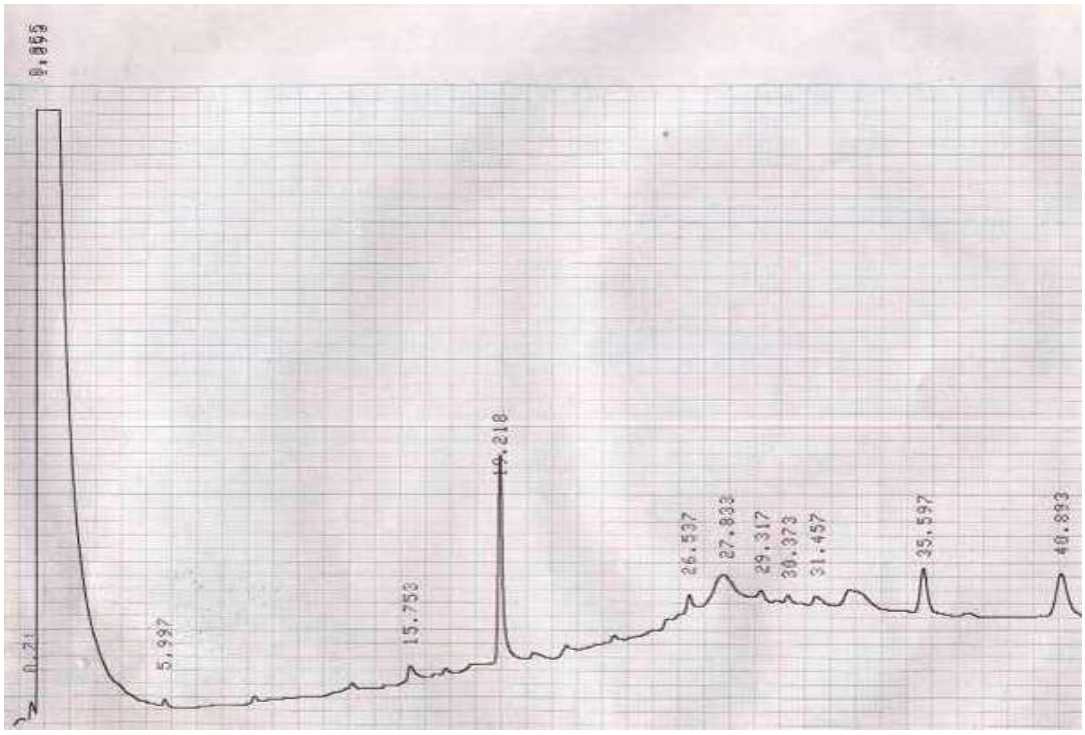


A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

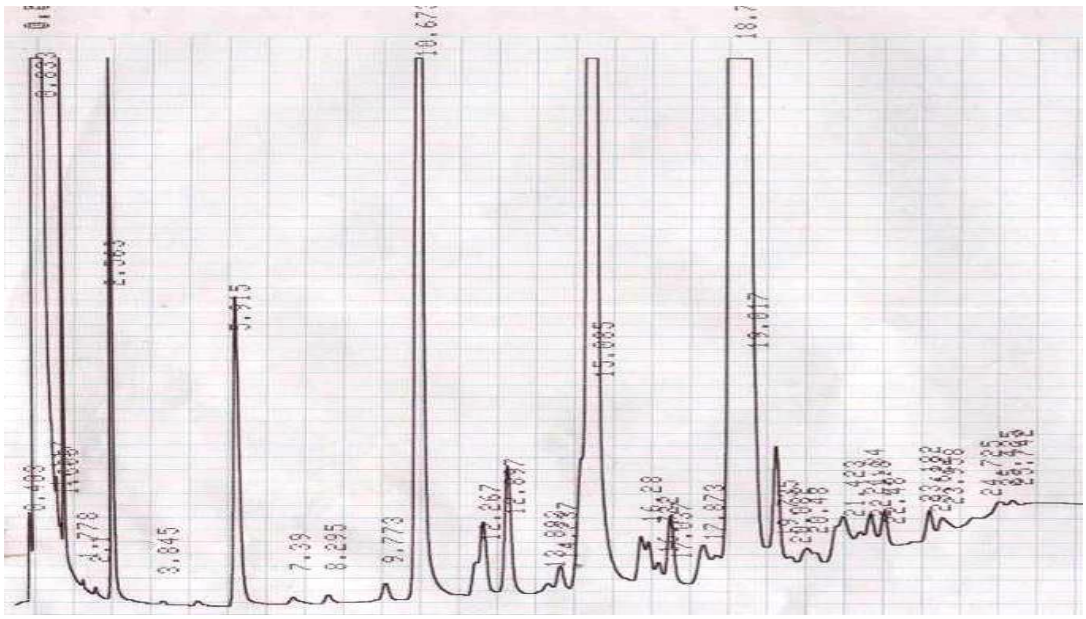


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

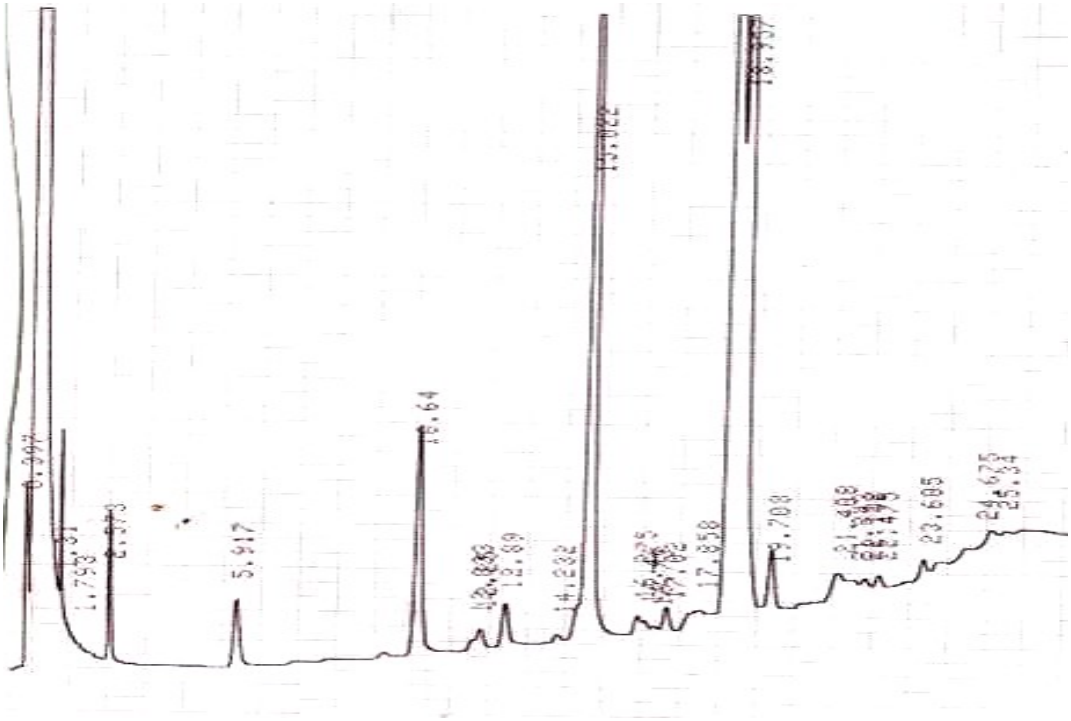
Appendix 3 cont.



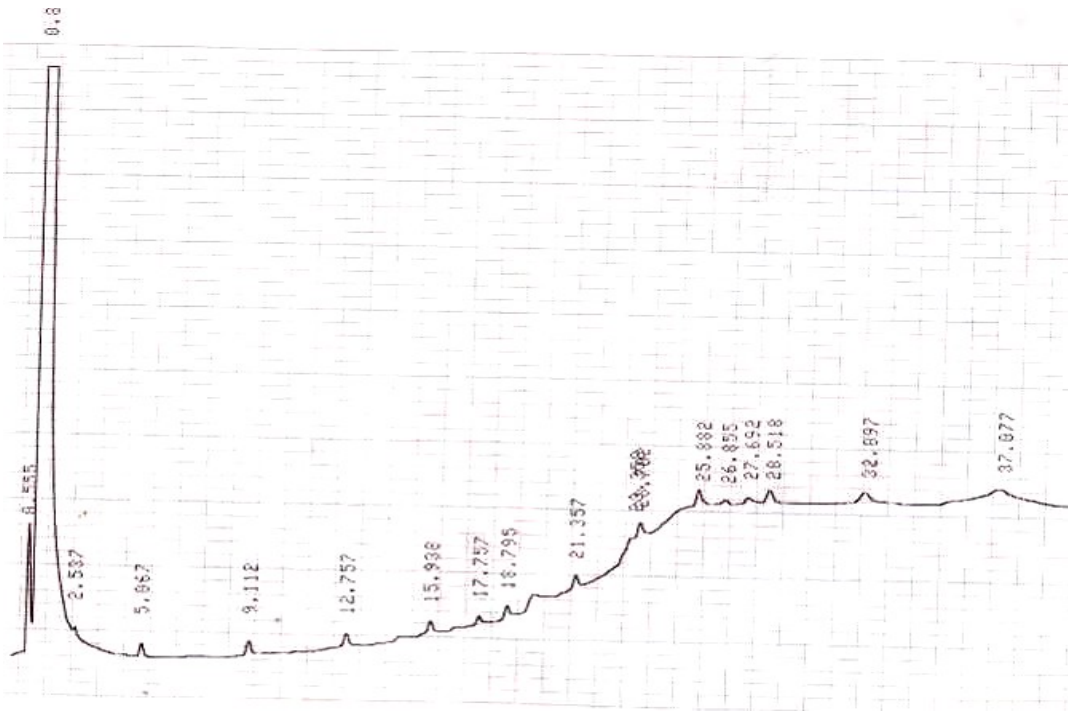
A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB



A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB



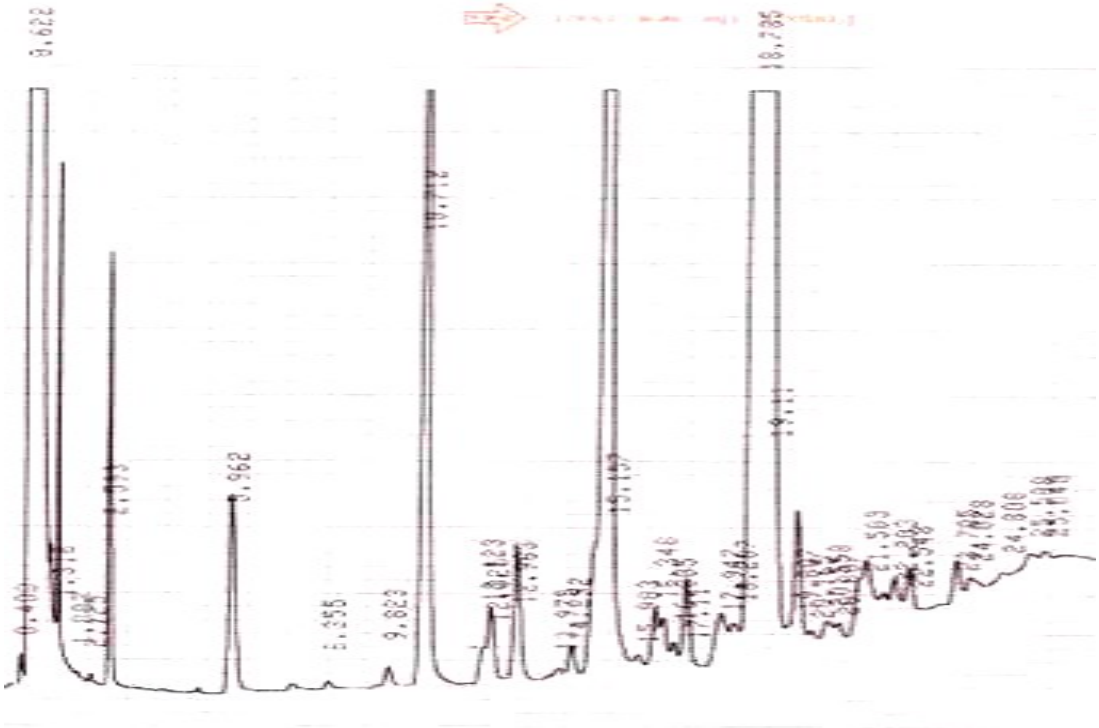
Appendix 3 cont.



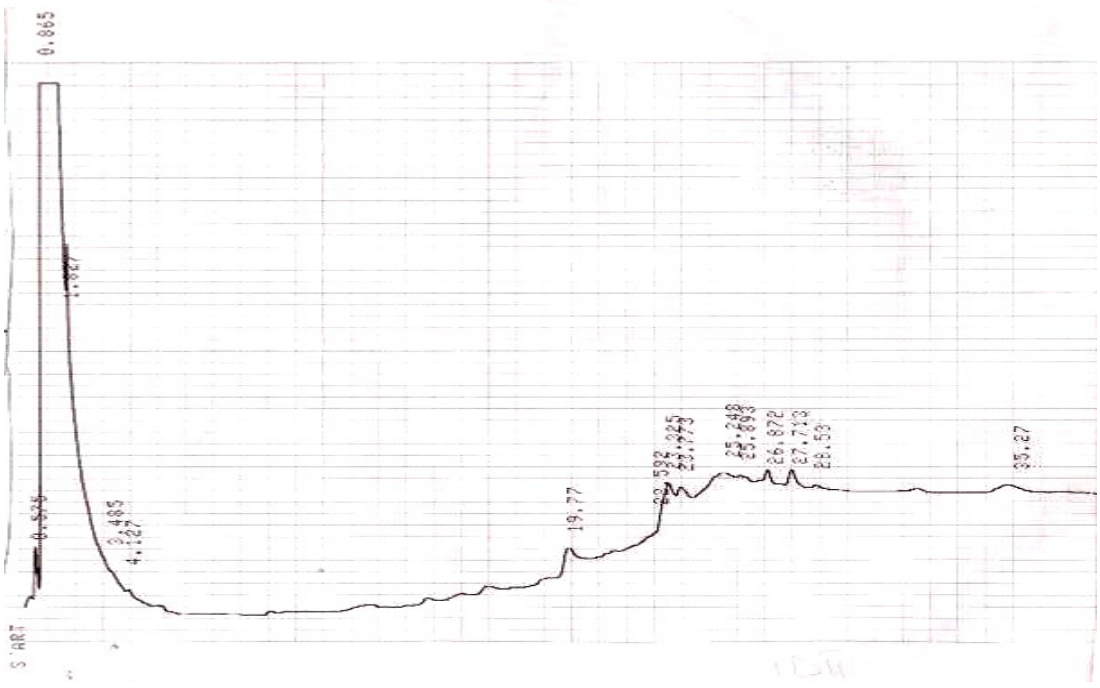
A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

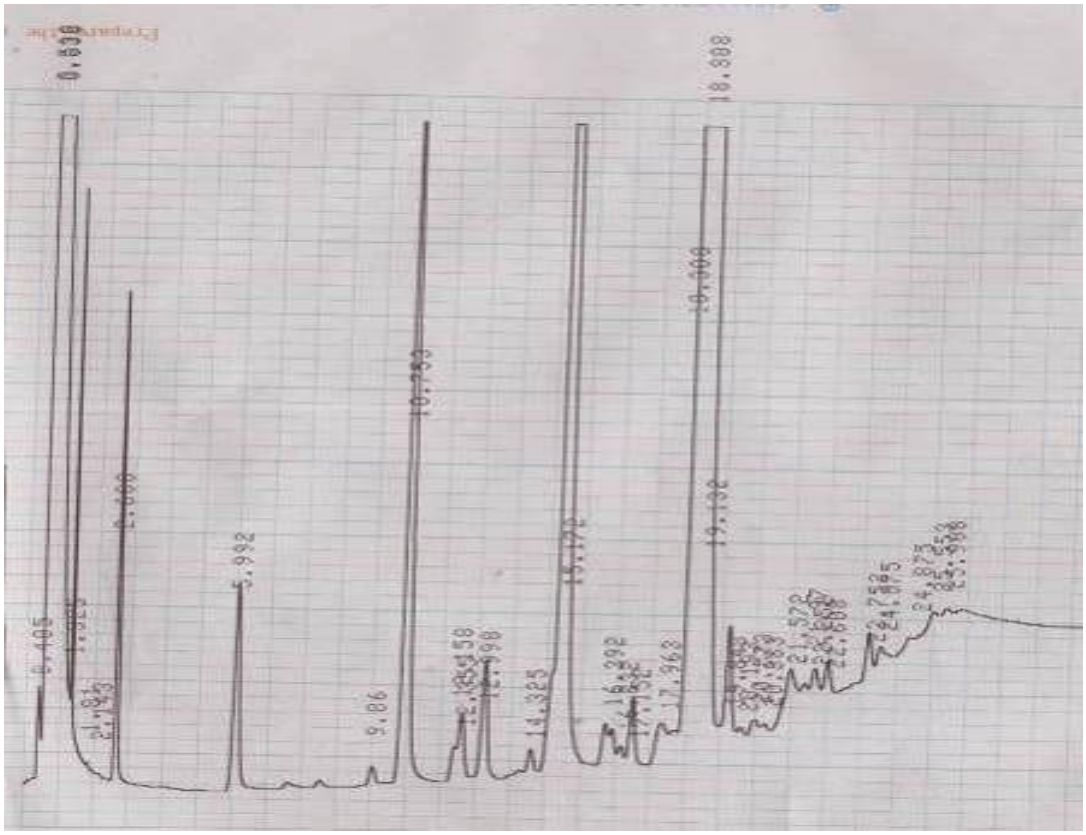
Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC



A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

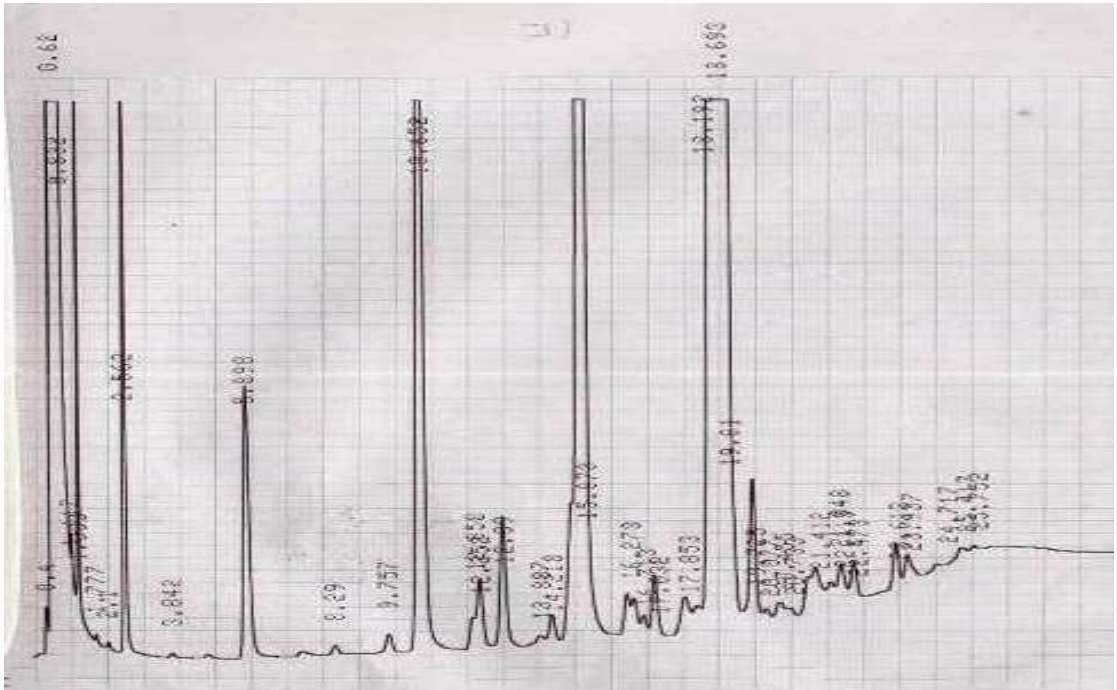


Appendix 3 cont.

A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

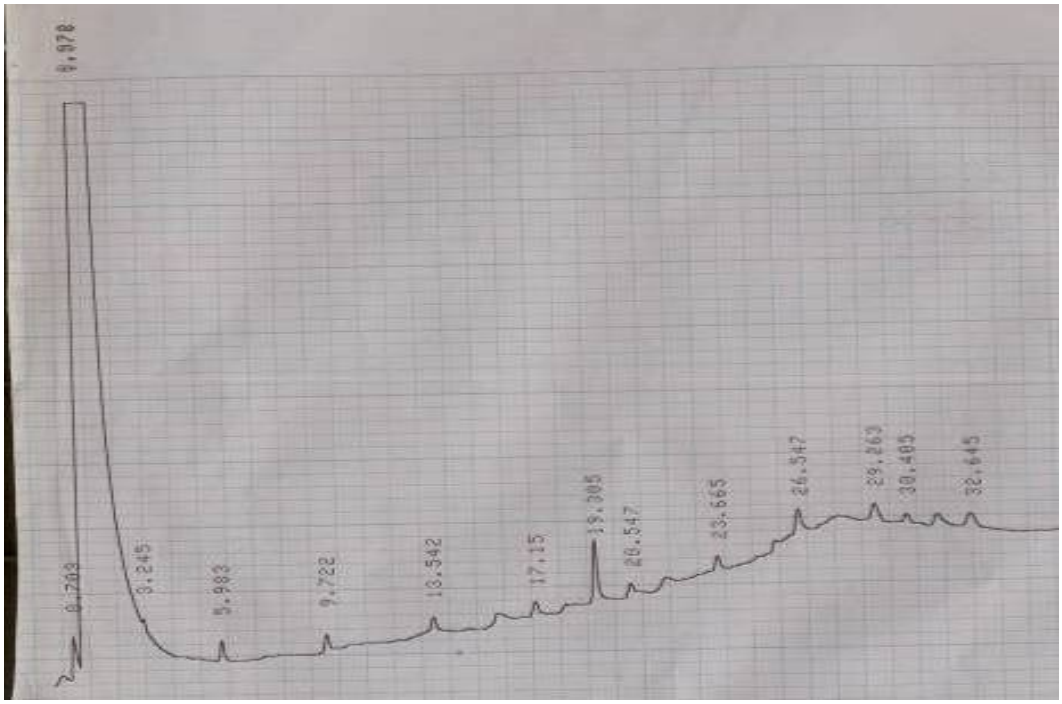
Appendix 3 cont.

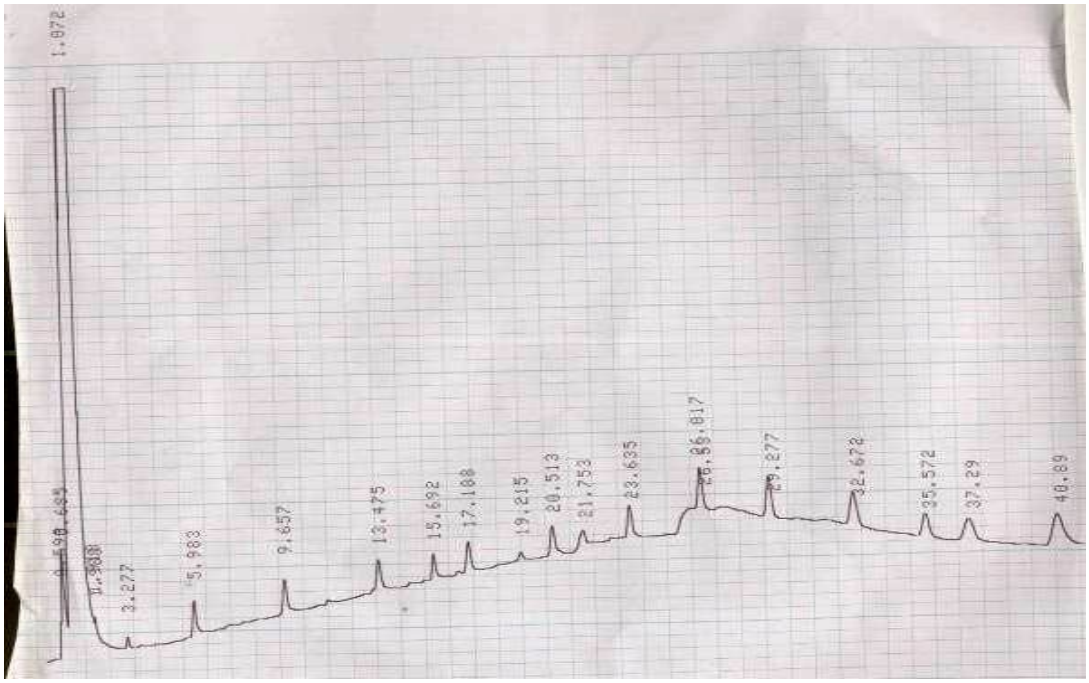


A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

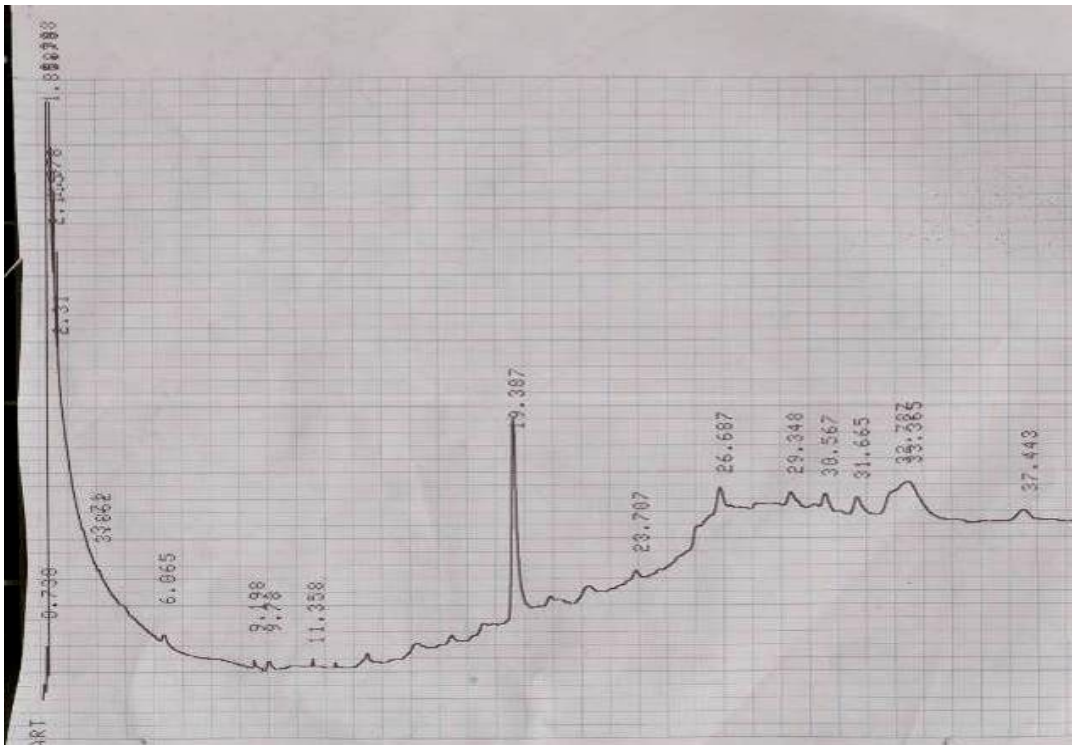
Appendix 3 cont.



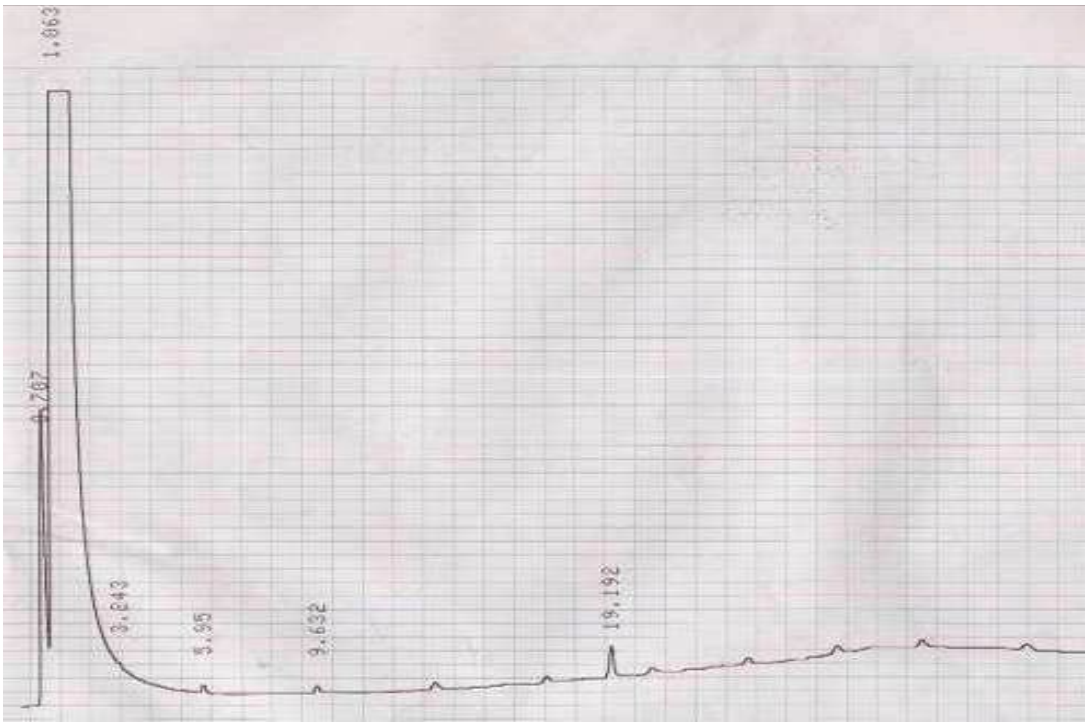


A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB



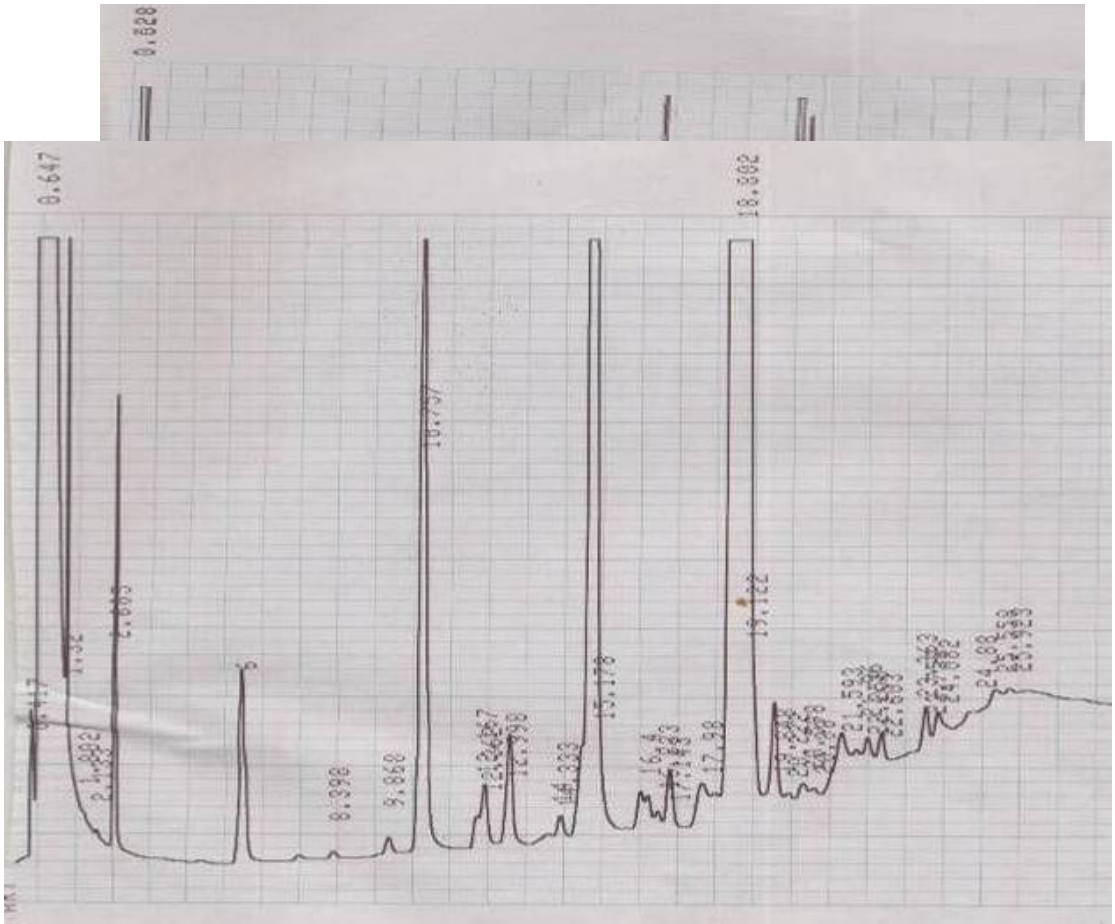
Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

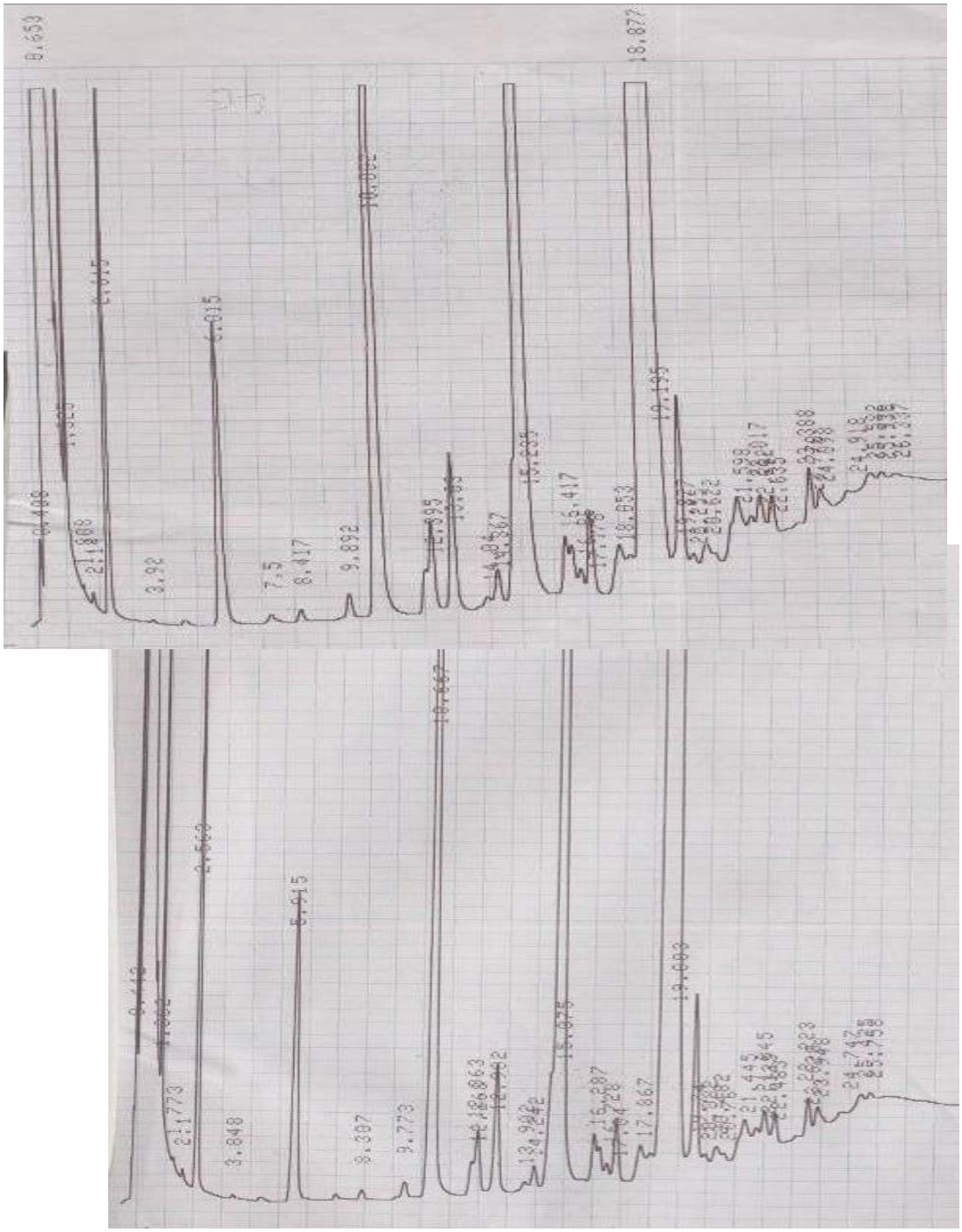
Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

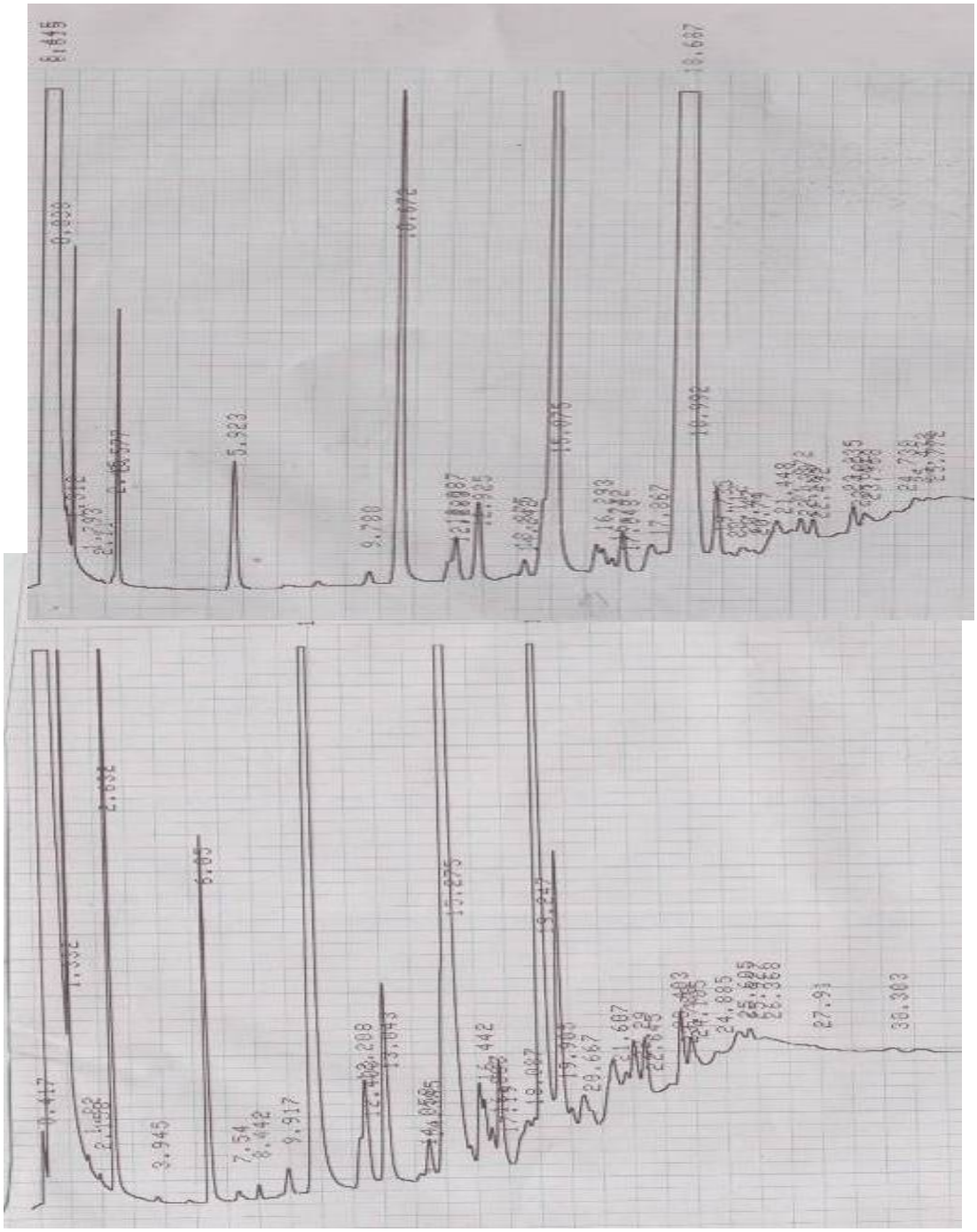
Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

Appendix 3 cont.

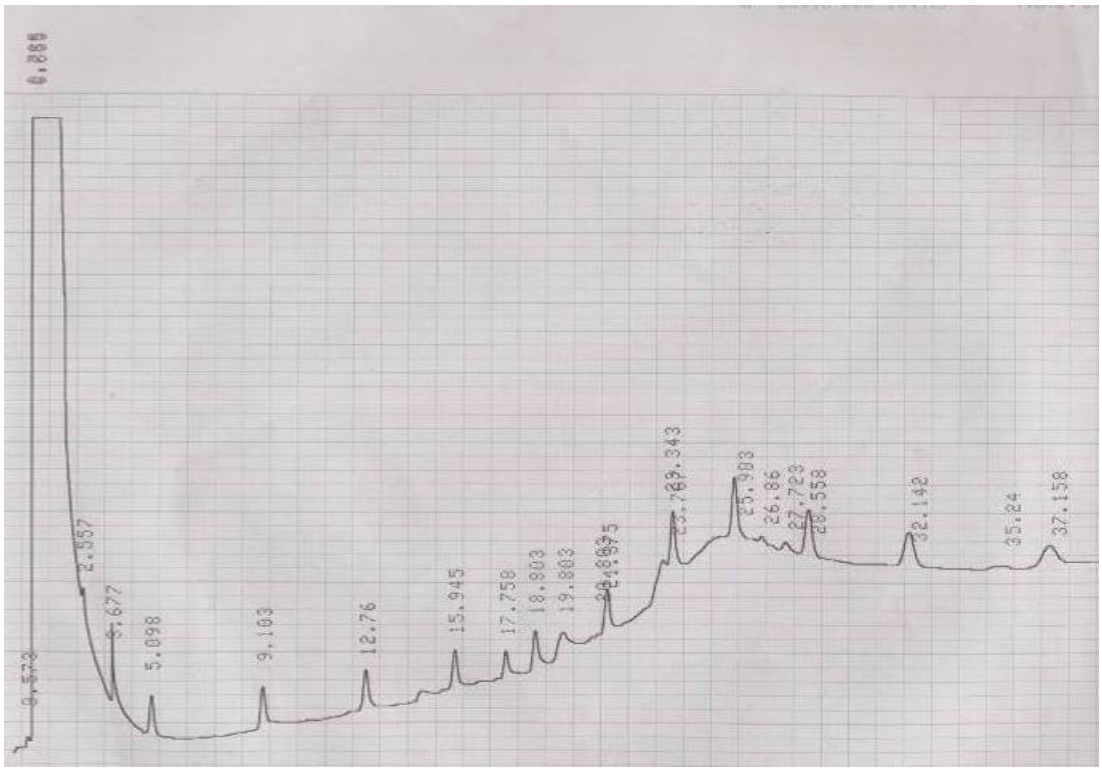
A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

Appendix 3 cont.

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB



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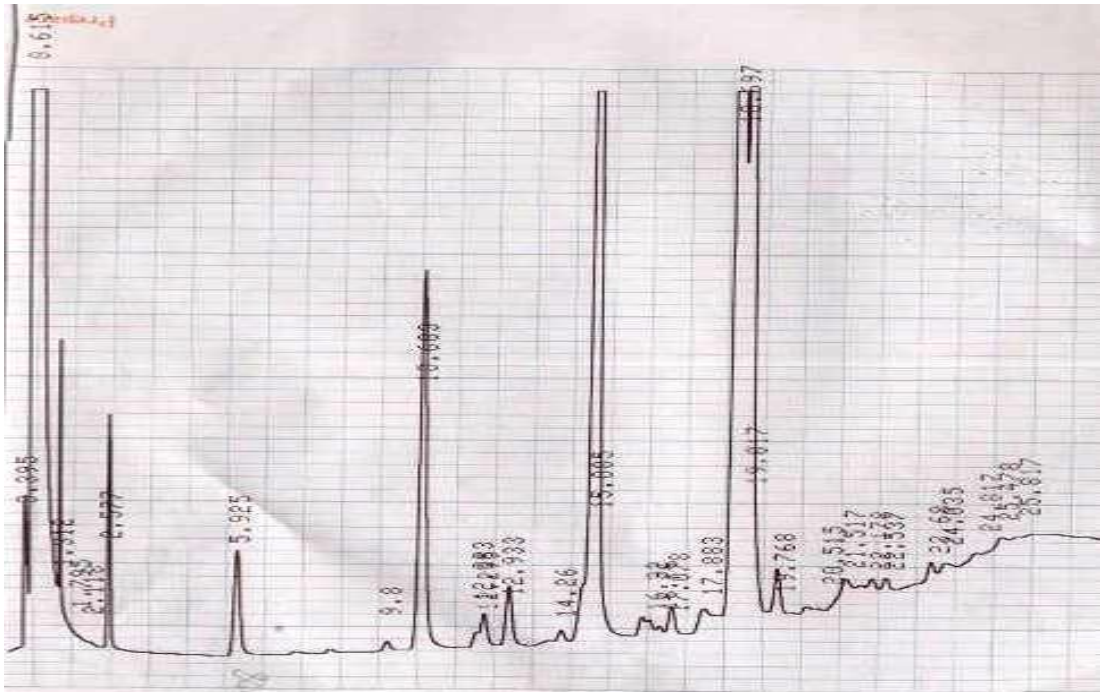
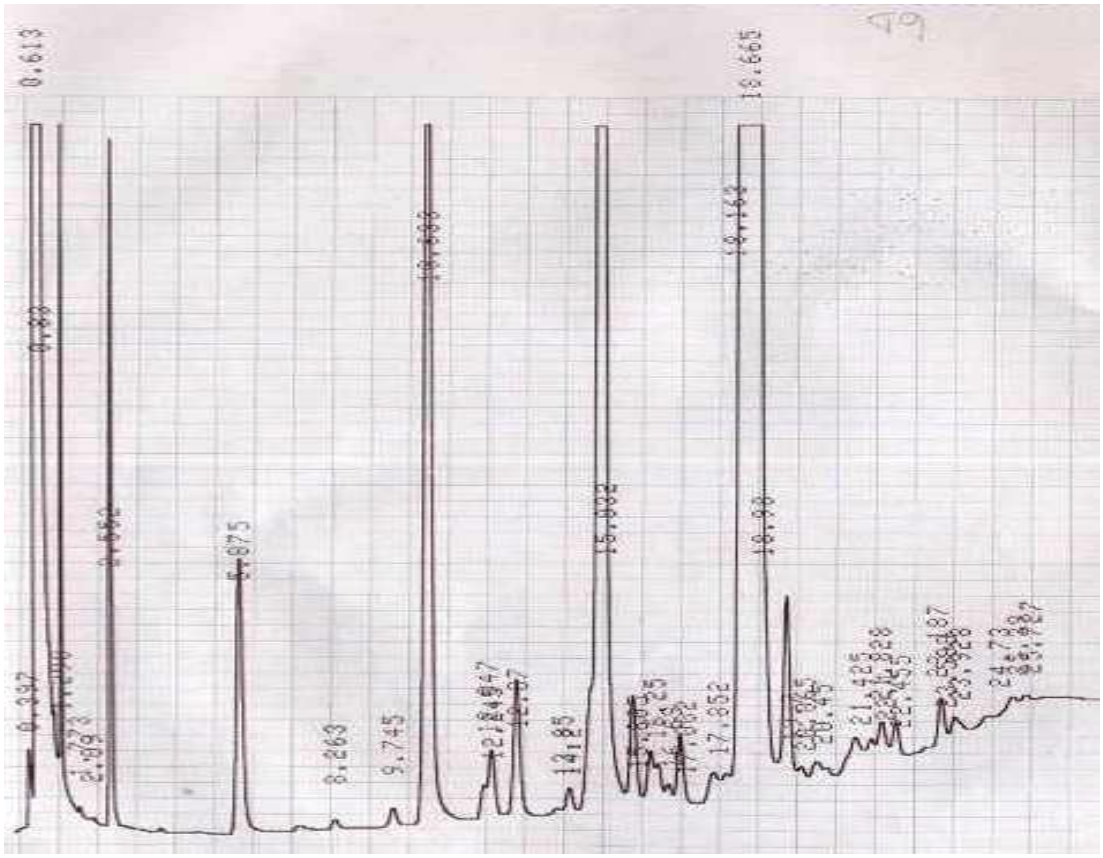
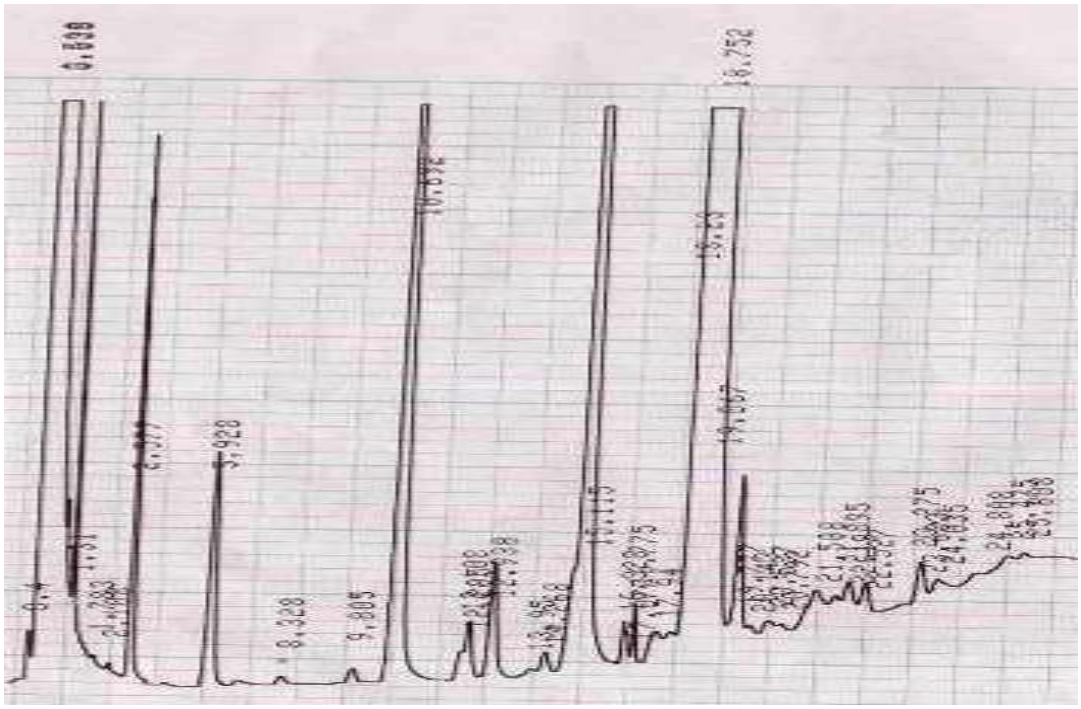


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

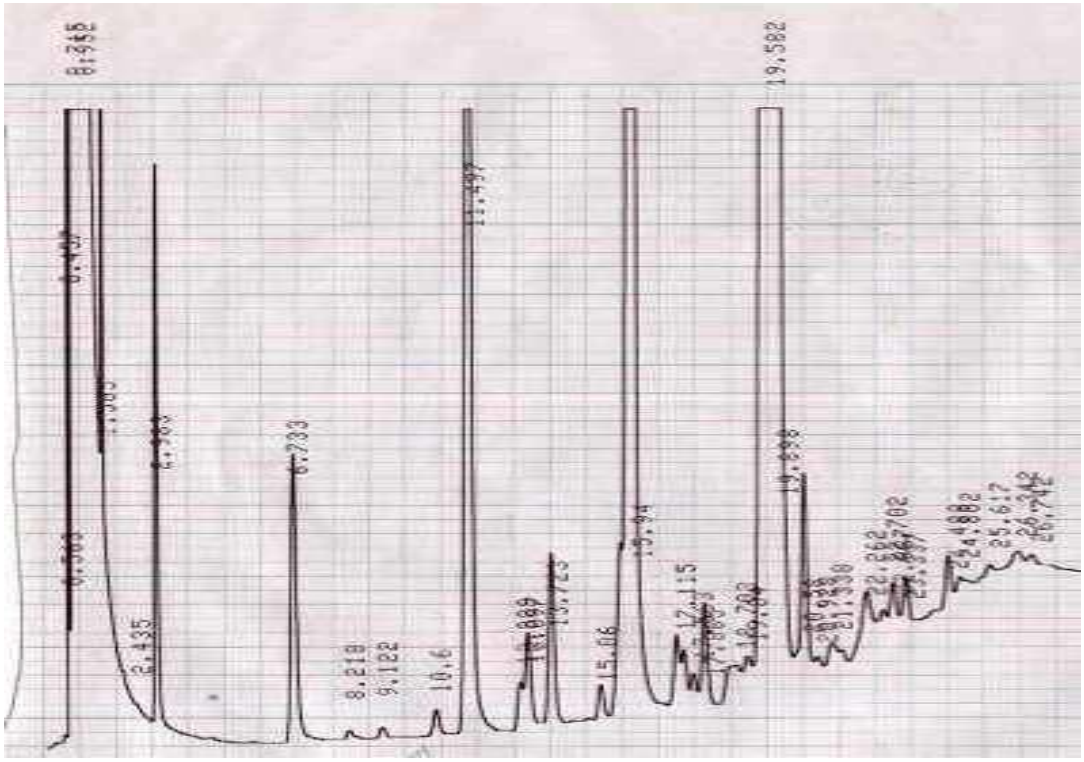


A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

Appendix 3 cont.

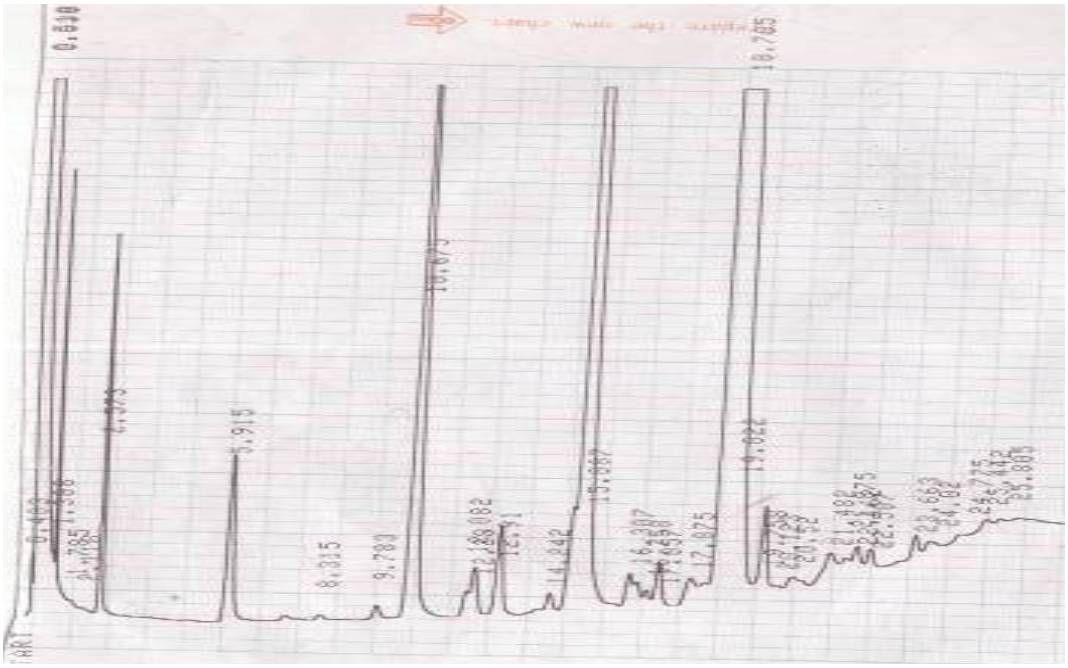


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

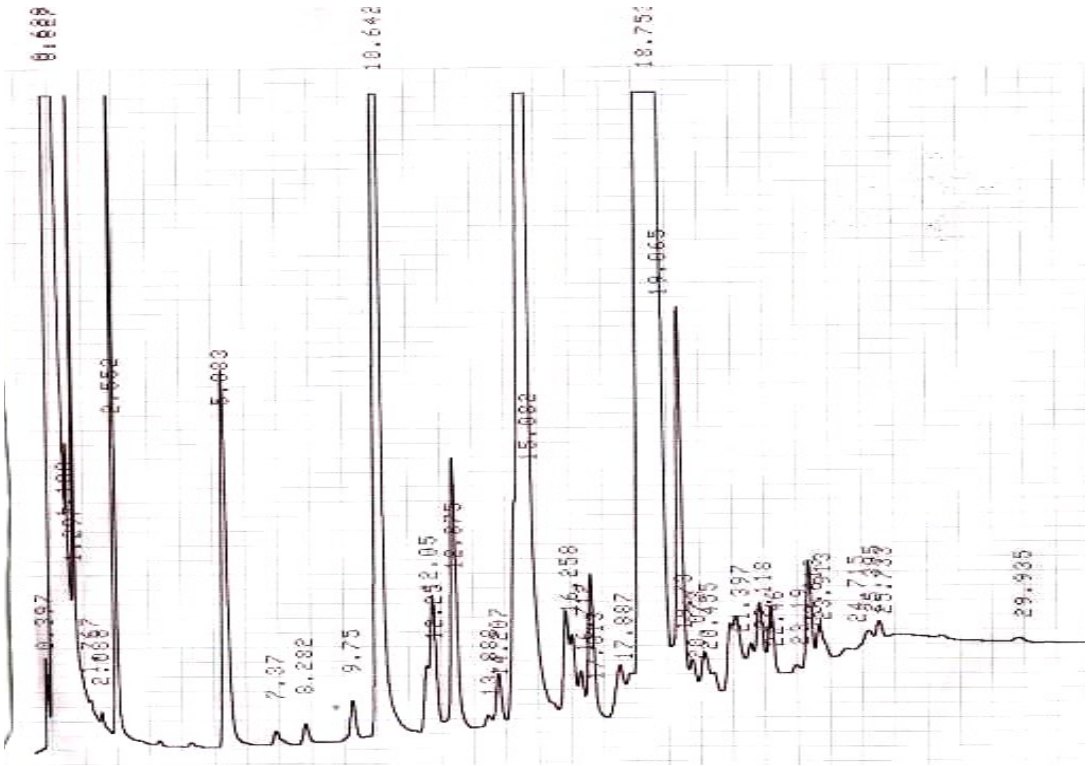


A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

Appendix 3 cont.

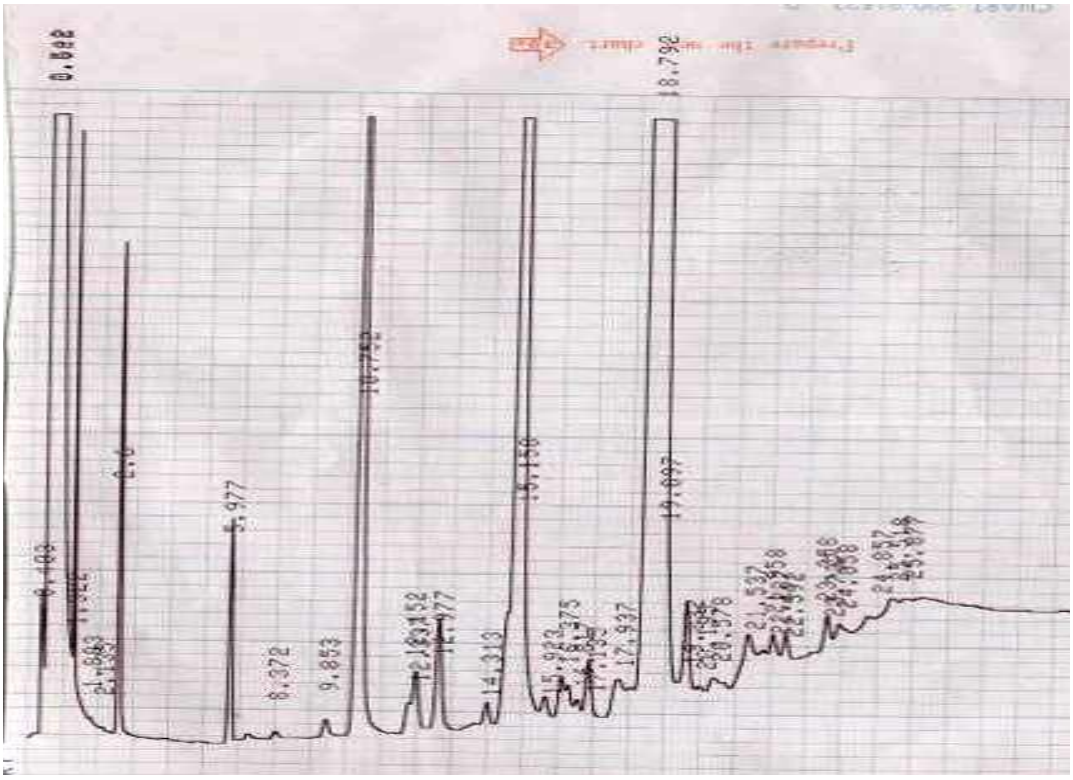


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

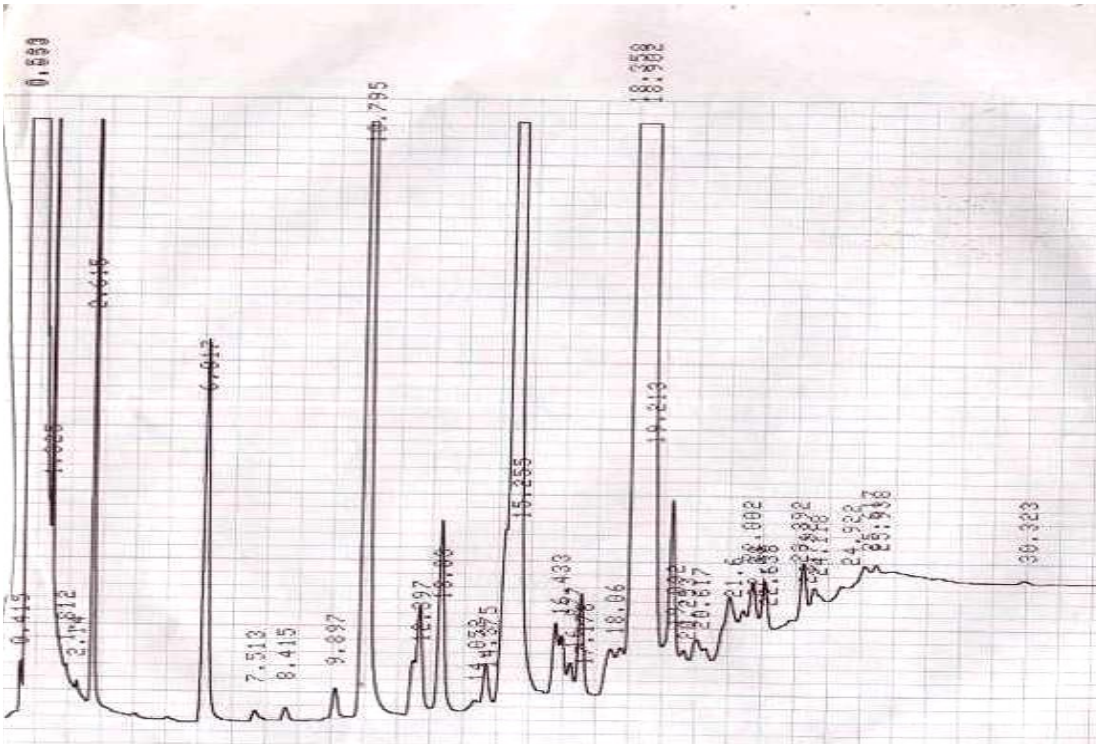


A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

Appendix 3 cont.

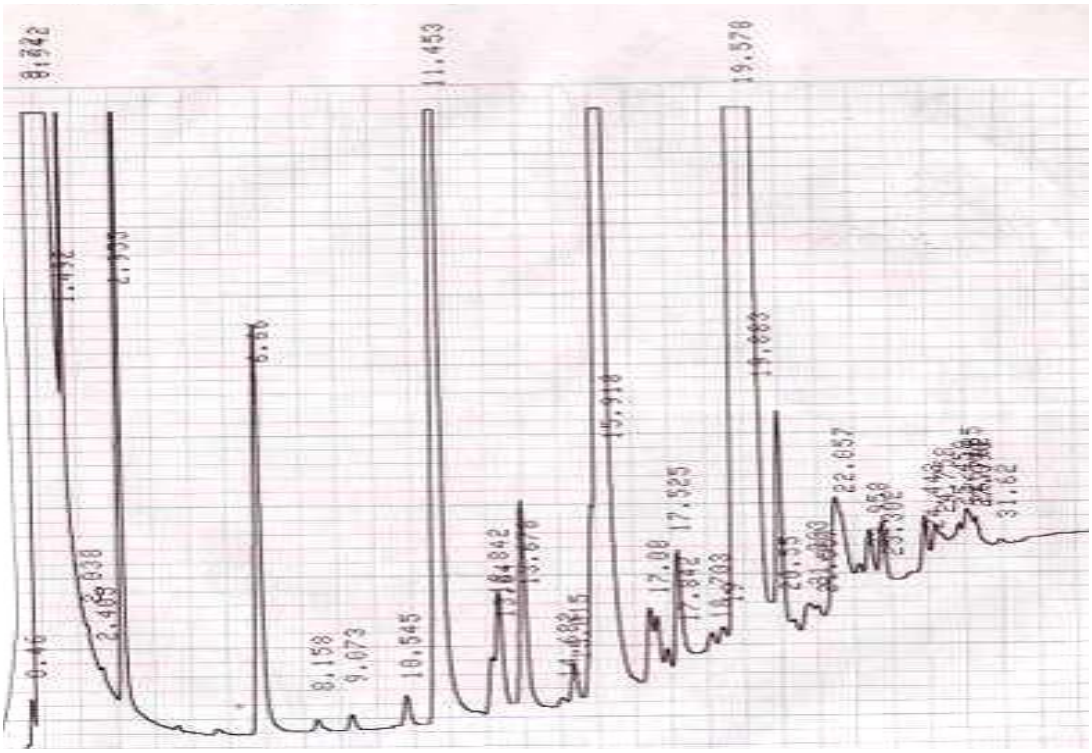


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

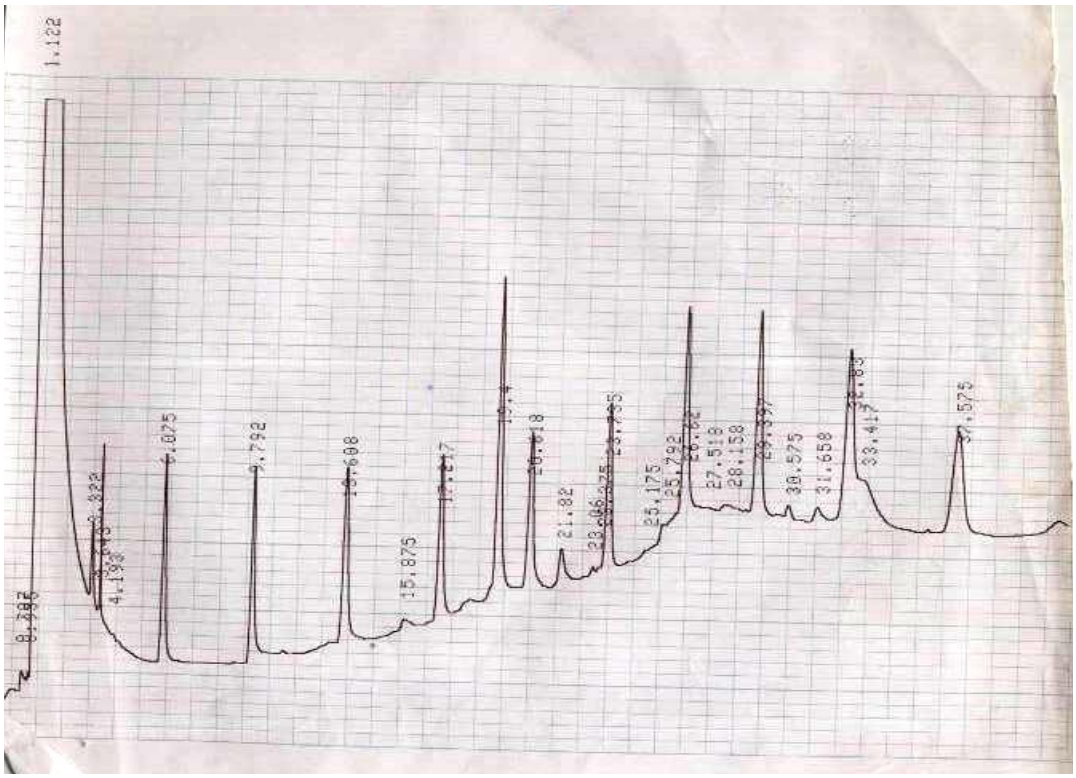


A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

Appendix 3 cont.

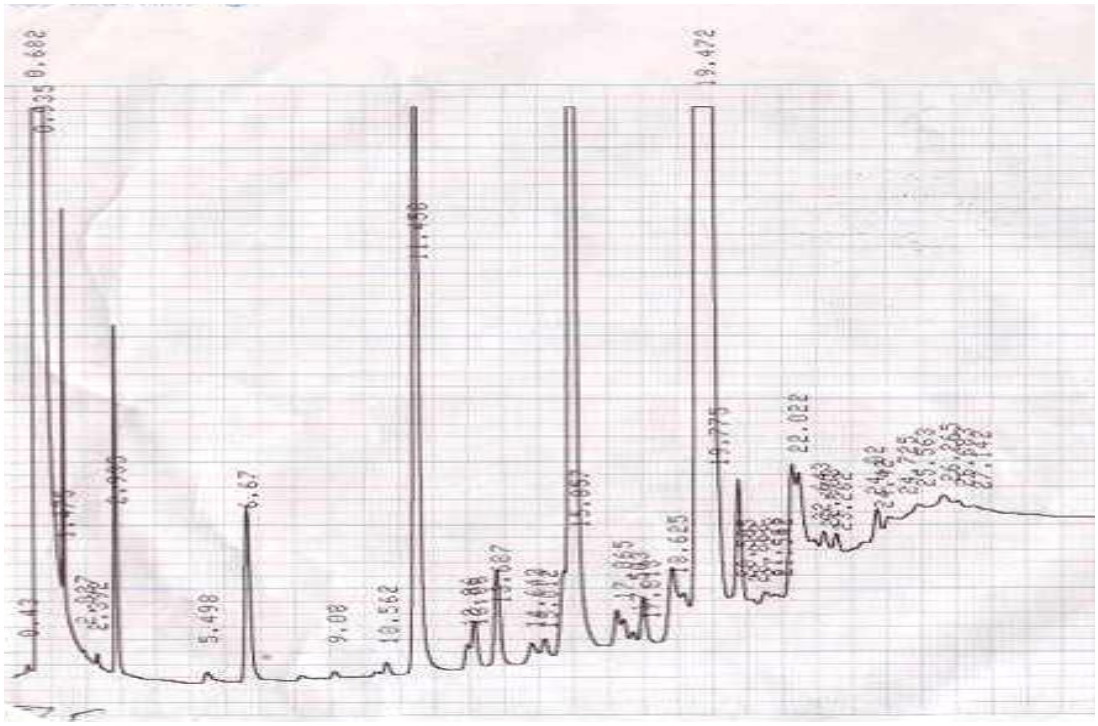


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

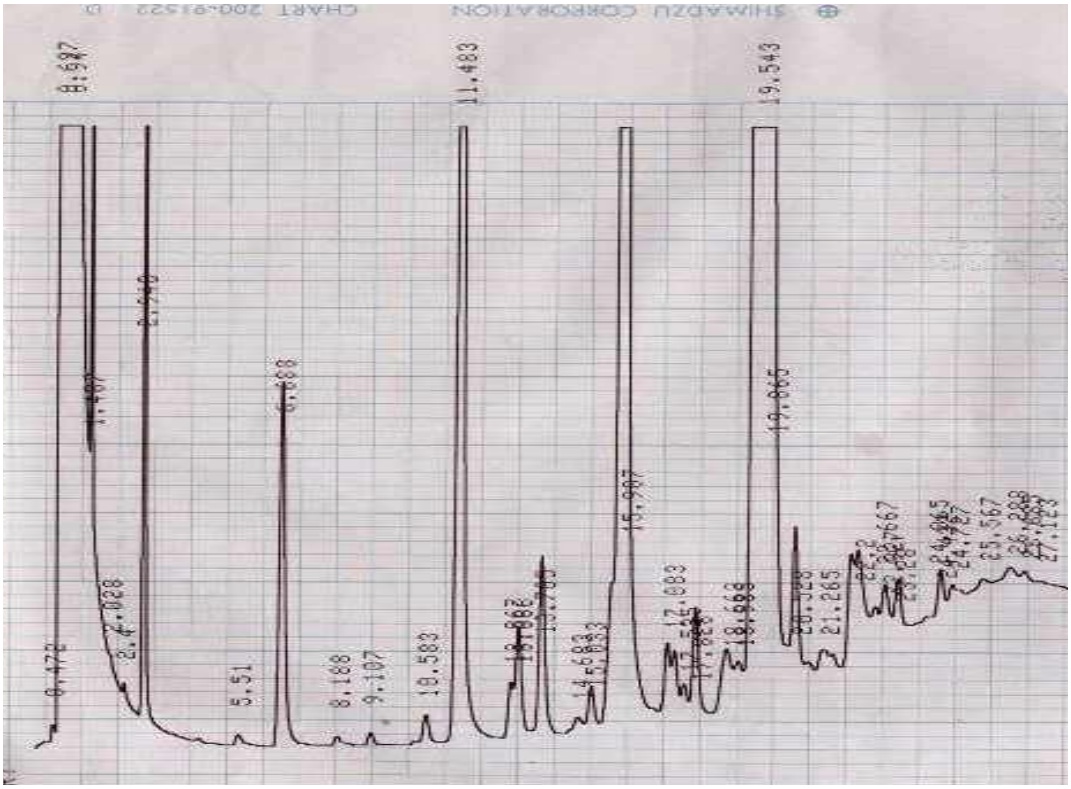


A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

Appendix 3 cont

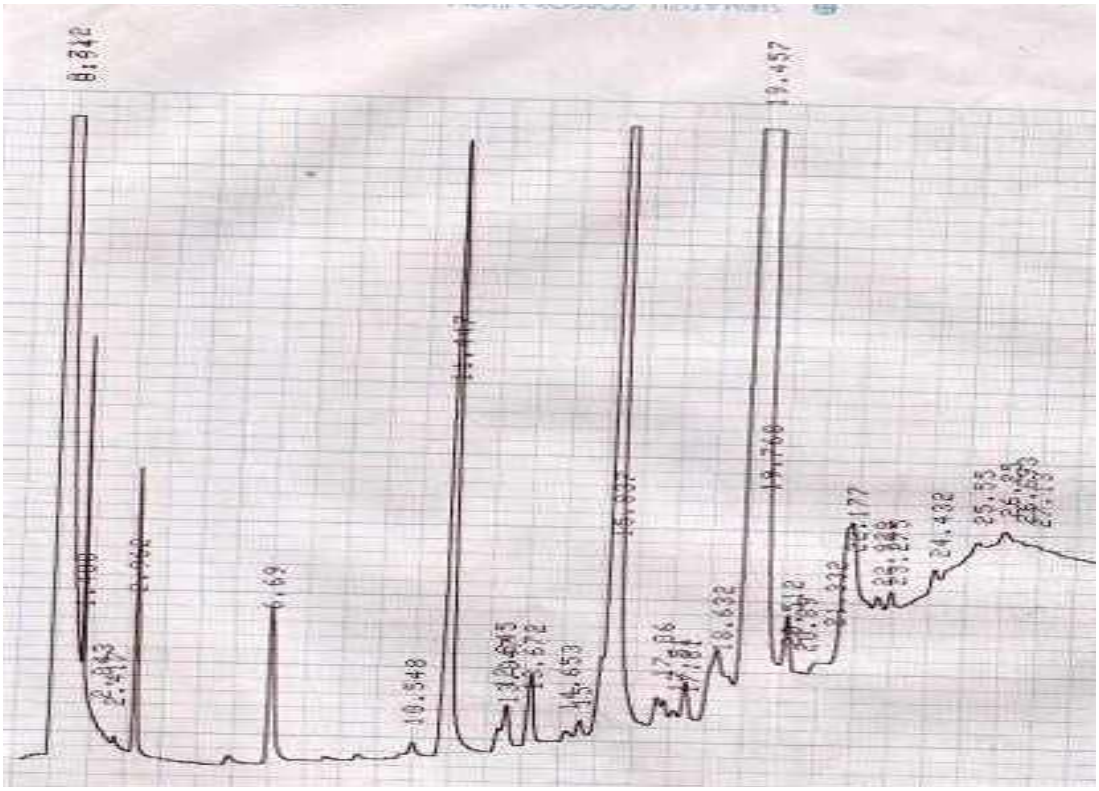


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

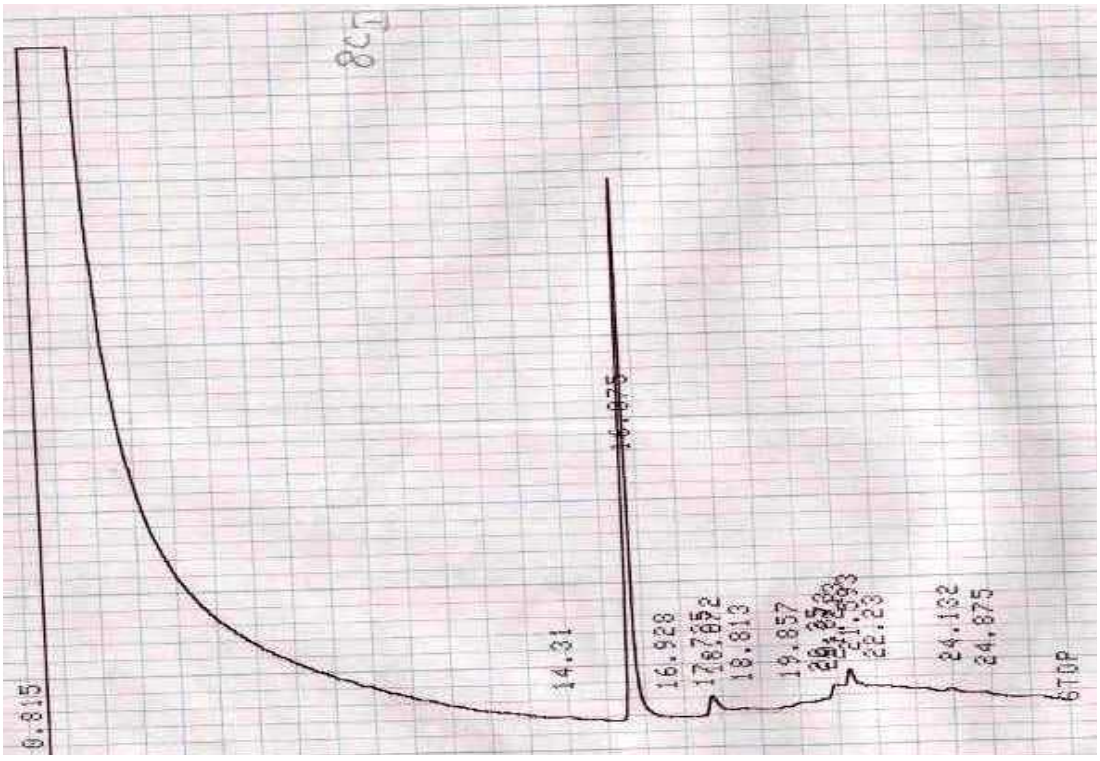


A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

Appendix 3 cont.

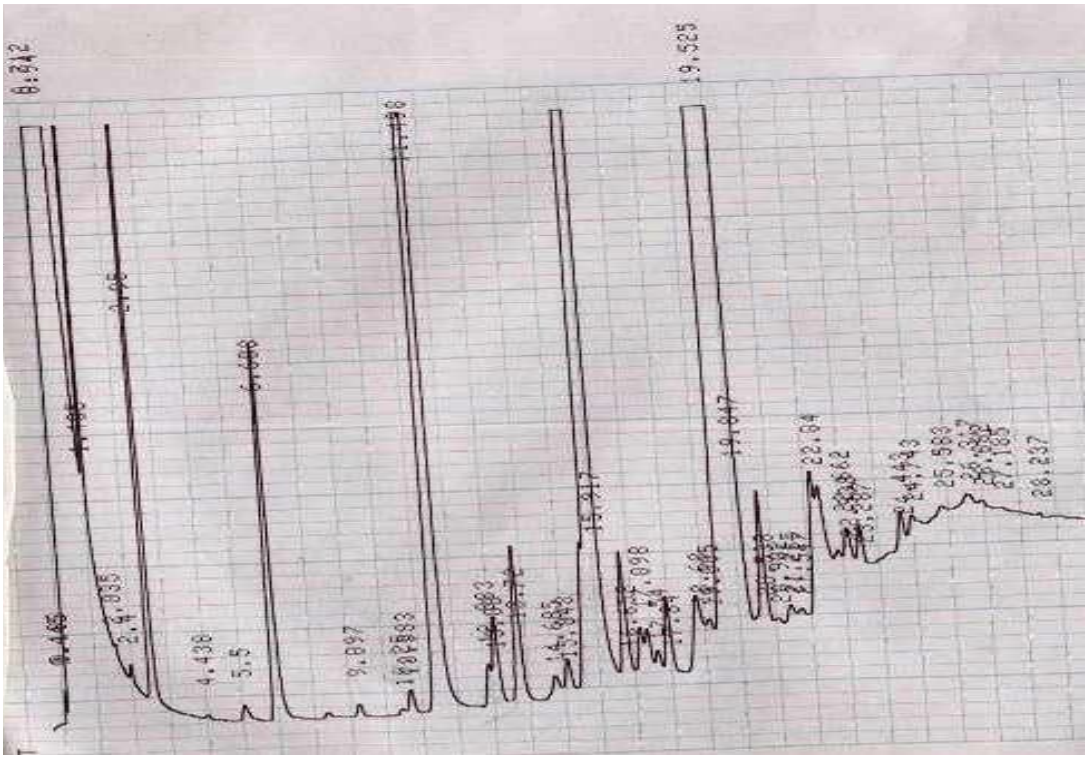


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC

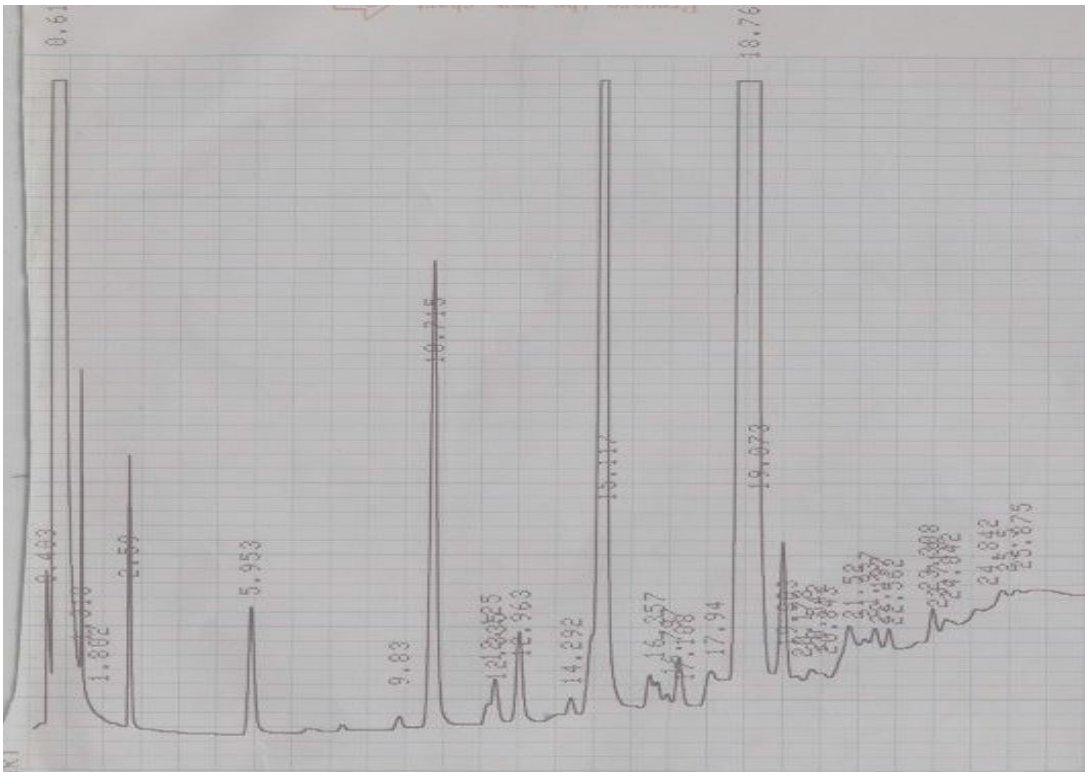


A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC

Appendix 3 cont.

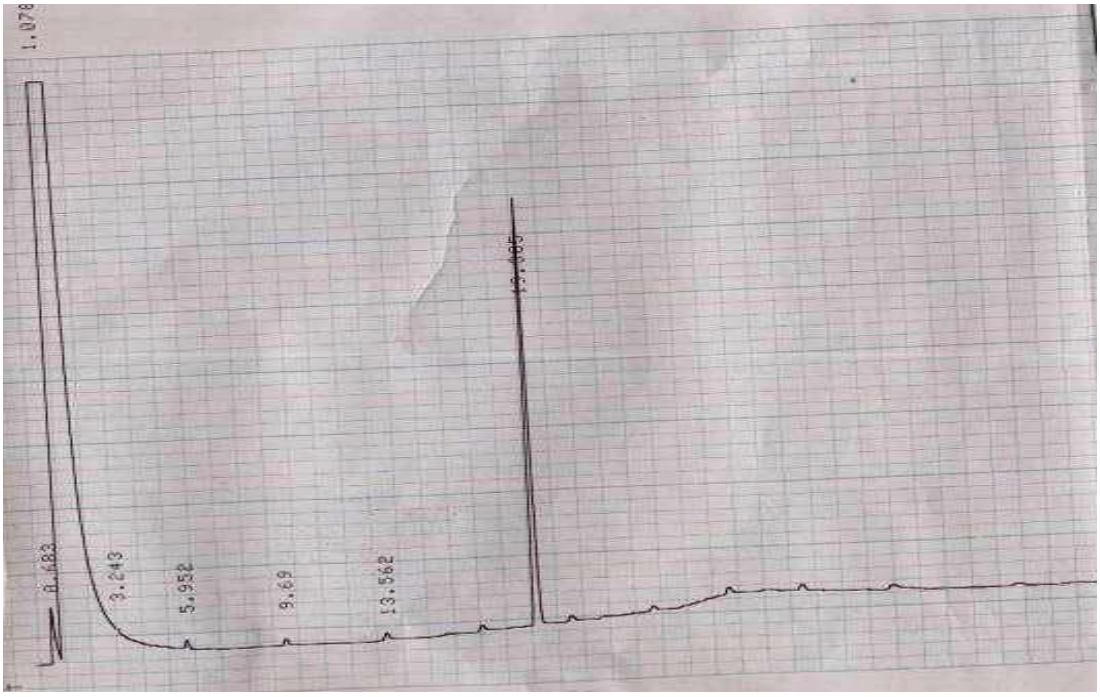


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed SSC

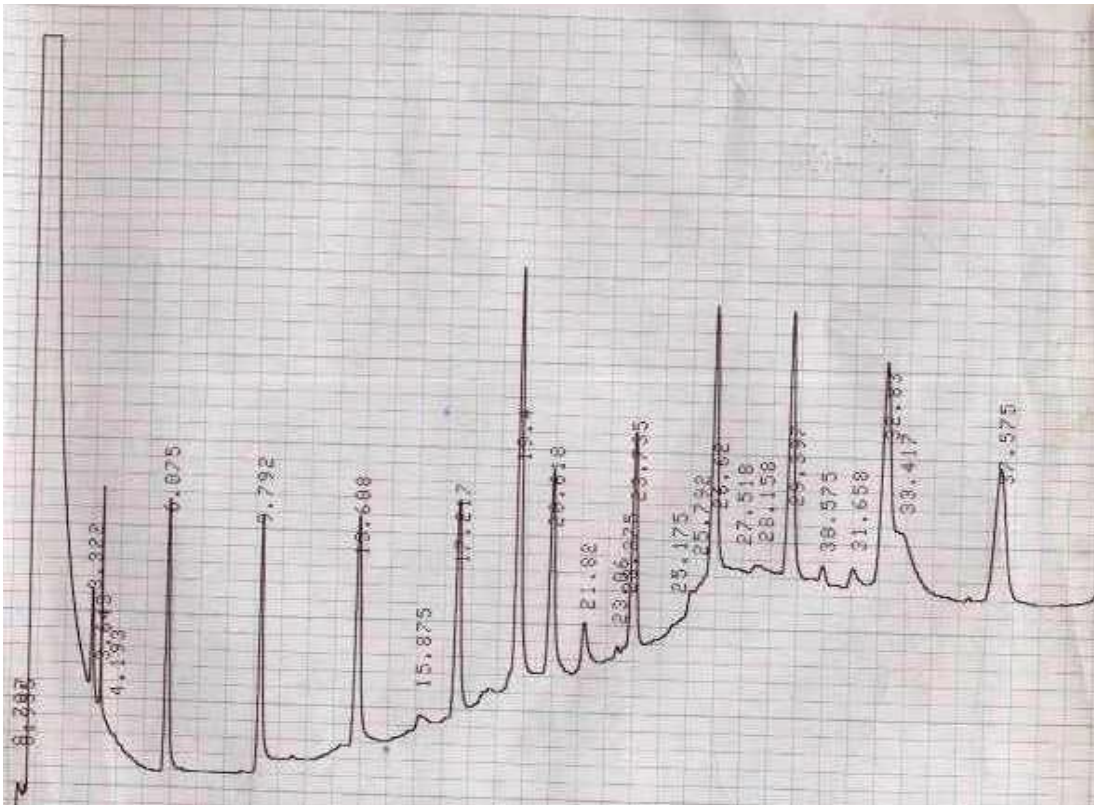


A chromatogram corresponding to the FA profile of milk BF from Friesian fed MB

Appendix 3 cont.

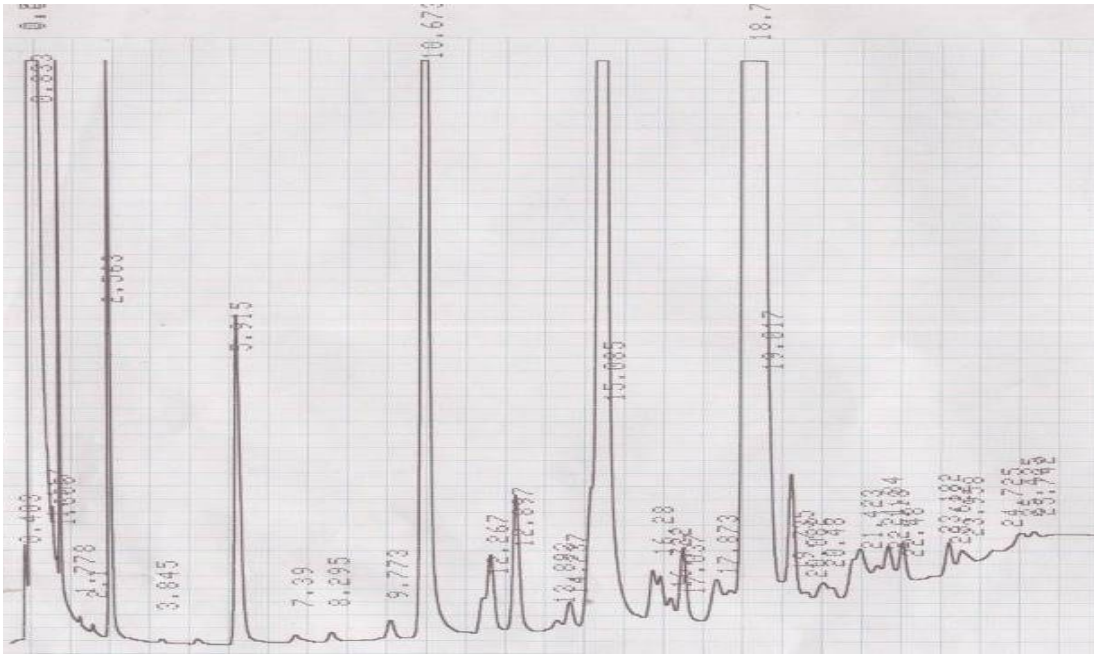


A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed MB

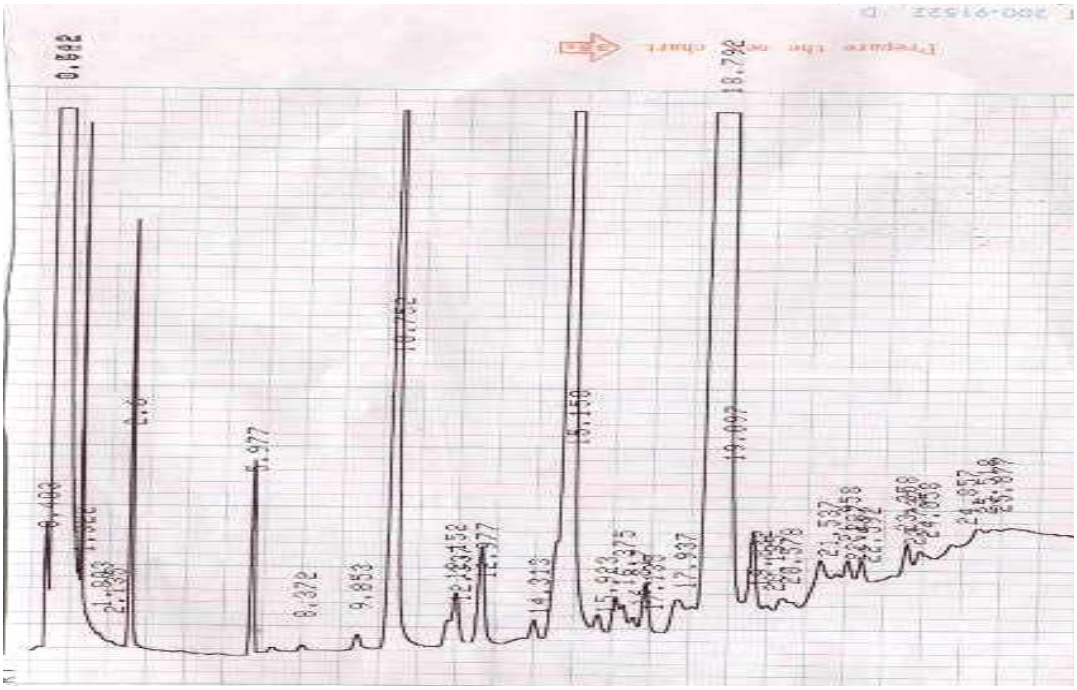


A chromatogram corresponding to the FA profile of milk BF from Friesian fed CSC

Appendix 3 cont.



A chromatogram corresponding to the FA profile of milk BF from Ayrshire fed CSC



A chromatogram corresponding to the FA profile of milk BF from Friesian fed SSC