MAKERERE UNIVERSITY

FATTY ACID PROFILES OF HUMP, KIDNEY AND BRISKET FAT AND THEIR CORRELATION WITH SENSORY PROPERTIES OF BEEF SALAMI

By:

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DECLARATION

I, RUGAMBA Eddy Frank, hereby declare that this work is my own original work and that I have not previously in its entirety or in part submitted it at any university for a degree.

This work has been written and submitted under supervision and approval of my Supervisor.

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Signature……..Date…….

DEDICATION

I dedicate this work to everyone who has supported me during my studies.

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My thankful heartfelt is extended to the Almighty God for his love, guidance, protection and care.

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List of Abbreviation and Acronyms

ALA: Alpha Linolenic Acid

AMSA: American Meat Science Association

ANOVA: Analysis of Variance

BFM: Body Fat Mass

CHD: Coronary Heart Disease

CLA: Conjugated Linoleic Acid

COVAB: College of Veterinary Medicine, Animal Resources and Bio-security

DHA: Docosahexaenoic acid

EFAs: Essential Fatty Acids

EPA: Eicosapentaenoic acid

FA: Fatty Acid

FAME: Fatty Acids Methyl Ester

FAMEs: Fatty Acid Methyl Esters

FAO: Food and Agriculture organization of the United Nations

FID: Flame Ion Detector

FSA: Food Standards Agency

GC: Gas Chromatography

GC-FID: Gas Chromatography Flame Ion Detector

HDL: High Density Lipoprotein

HSD: Honestly Significant Difference

IMF: Intramuscular fat

LBM: Lean Body Mass

LC: Long Chain

LDL: Low Density Lipoprotein

Lp: Lipoprotein

MAPPES: Masters in Animal Product Processing Entrepreneurship and Safety

MUFA: Monounsaturated fatty acids

P: S: The ratio of the polyunsaturated to saturated fatty acids

PEG: Polyethylene-glycol

PUFA: Poly Unsaturated Fatty Acids

QDA: Quantitative Descriptive Analysis

SFA: Saturated Fatty Acids

SPSS: Statistical Package for Social Scientists

U. S: United States

UMI: Uganda Meat Industries

WHO: World Health Organization of the United Nations

Abstract

This study examined the influence of fatty acid composition on the sensory properties of beef salami made using brisket, kidney and hump fats. Fat samples were analyzed for total saturated fatty acids (SFAs), mono-unsaturated fatty acids (MUFAs) and poly-unsaturated fatty acids (PUFAs). Total SFAs contents from the hump and the brisket fat were at 21.25% and 20.1% respectively, while kidney fat had lower content at 13.52%. Palmitic acid was the highest SFA in the hump fat with 9.35%, Behenic Acid was higher (5.73%) in kidney fat and Stearic acid was the most abundant saturated fatty acid in the brisket fat (5.41). The total SFAs of the hump fat were significantly different from those of the kidney fat at (HSD =.036).The total MUFAs of the hump fat were significantly different from those of the kidney Fat at (HSD=.007) the same as the MUFAs of the kidney fat which were significantly different from those of the brisket fat at (HSD=.004). For the levels of monounsaturated fatty acids (MUFAs); Oleic acid (C18:1) was higher in hump fat (33.02%), Eicosenoic acid (C20:1) was higher in brisket fat (34.28%) while Erucic acid (C22:1) was very high in kidney fat (33.86%) compared to the other two fat samples. Both Linoleic and alpha linolenic acids were found in higher levels in brisket fat (2.13%) and (2.08%), followed by kidney fat (0.62%) and (2.03%) and were lowest in hump fat (0.48%) and (1.18%) respectively. Sensory attributes such as appearance, taste, flavor, texture, mealiness and product consistency were marked by panelists on a 1 to 9 hedonic scale. Fatty acids compositions were found to affect the sensory attributes especially flavor, texture and appearance of beef salami while the influence of fatty acids on taste and product consistency was not well understood. The hump fat salami was the most liked product in terms of the studied sensory properties with the highest rank (7.08) on a scale of panelist (1 to 9) while the brisket and kidney fat salami ranked 6.88 and 6.68 respectively in the overall product liking. From these findings, the meat industry would make a balanced decision of providing both good quality and less harmful products to consumers and provide them with clear information about the used ingredients to guide their choice.

CHAPTER ONE

1.0 INTRODUCTION

1.1. Background

Meat is a premium, high-demand food throughout the world. Meats have high quality proteins, which contain essential amino acids, and are a very good source of vitamin B complex, dietary iron and zinc. Meat is the most valuable livestock product and for many people serves as their first-choice source of protein. In general, meat is composed of water, fat, protein, minerals and a small proportion of carbohydrate (Heinz and Hautzinger, 2007). The meat-processing industry is one of the largest agricultural and food-processing industries in the world (Gangidi and Proctor, 2008). Meat, fat and other carcass parts used as raw materials for the manufacture of processed meat products are mainly derived from the domesticated species such as cattle (*Bos primigenius),* pigs *(*Sus *domesticus)* and poultry (*Gallus gallus domesticus)* and to a lesser extend from buffaloes *(Bubalus bubalis),* sheep *(Ovis aries)* and goats *(Capra aegagrus hircus).* Other animal species such as camels (*Camelus),* yaks (*Bos grunniens),* horses (*Equus ferus caballus)* and game (*Galliformes)* can be used as meat animals but they only play a minor role in meat processing (Heinz and Hautzinger, 2007). Throughout history, meat has been the foremost food of most people of the world. Many groups of people from various parts of the world, such as the Laplanders, Eskimos and American Indians, have subsisted almost exclusively on a meat diet for many generations (Craypo, 1994).

Meat consumption in developing countries has been continuously increasing from modest average annual per capita consumption of 10 kg in the 1960s to 26 kg in 2000 and will reach 37 kg in 2030 according to FAO projections (Heinz and Hautzinger, 2007). In the last 30 years, meat products have increasingly replaced cereal products in the human diet. This is mostly observed in urban areas and it is propelled by a high income growth rate of the urban population (Monteiro *et al.*, 2010)

In terms of global meat production, an increase from the annual production of 267 million tons in 2006 to nearly 320 million tons by 2016 is expected (World Bank, 2012). Global meat production was set to expand by almost two per cent in 2012, according to the FAO's Food Outlook report for May 2012 (Naziri and Bennett, 2012). Beef production in Uganda is estimated

at an average of 160,000 metric tons per year giving the national meat consumption per capita in Uganda as 11 kg which is lower than the 40 kg recommended by the WHO (Nalubega *et al.*, 2010). Meat quality refers to the compositional quality and the palatability of meat. The major parameters considered in the assessment of meat quality are appearance, juiciness, tenderness and flavor (Muchenje *et al.*, 2009). The sensory attributes of beef have been shown to be affected by the chemical properties and lipid content (Abbas *et al.*, 2009). The Fatty Acids (FA) composition (intramuscular fat, IMF) exerts a strong influence on meat quality because of the implications of these components on organoleptic properties through the oxidative breakdown during ageing (Wood *et al.*, 2004) and the formation of volatiles during cooking. In other words, the fatty acid composition influences both the nutritive value of beef and also the organoleptic properties and in particular flavor.

Besides adding flavor, appetite appeal and satiety to foods, fats provide essential fatty acids and aid in the absorption of fat-soluble vitamins A, D, E, and K and carotenoids. Meat healthiness is largely related to its fat content and its fatty acid composition. The absolute amounts of dietary fatty acid intake are important to calculate the physiological effect on cardiovascular risk factors and chronic diseases (Begg *et al*., 2007). In addition, modern consumers are increasingly concerned about production of safe meat with no undesirable effects on their health (Andersen and Newman, 2005). The consumption of dietary fats have long been associated to chronic diseases such as obesity, diabetes, cancer, arthritis, asthma, and cardiovascular disease (Ruiz-Rodriguez *et al.*, 2010).

Beef fat is a significant source of saturated fatty acids in the human diet because red meat has a relatively high ratio of saturated to unsaturated fatty acids in its lipids, which is a risk factor for the development of vascular and coronary diseases (Muchenje *et al.*, 2009). Fat content varies widely depending on the cut and degree of trimming (Abbas *et al.*, 2009) and the fatty acid composition of the fatty tissues vary with different locations of the cattle's body (body fat, hump fat, kidney fat and brisket fat). These fats are not equally distributed on the beef carcass; it is sometimes difficult to get enough of them to be used in processing of different beef products (e.g. the hump fat). Some of the beef products where fat is added during processing are salami. Salami is a large, thick, highly seasoned, and often cured type of sausage. It is of Italian origin and

usually served cold in thin slices.

Dietary recommendations from many health agencies world-wide emphasize a reduction in saturated fatty acids and an increase in n-3 polyunsaturated fatty acids (PUFAs) (Kris-Etherton *et al.*, 2007). The evidence that long chain (LC) n-3 PUFA exert a preventive effect on cardiovascular disease has been extensively reviewed (Wang *et al.*, 2006), but these FA are not synthesized by humans and must be provided in the diet. Not all saturated fatty acids found in foods add to the risk of heart disease; four (caproic, caprylic, capric and stearic) appear to have a neutral effect on low density lipoprotein (LDL) cholesterol, and three (lauric, myristic and palmitic) actually have LDL increasing potential (German and Dillard, 2004). Polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA) are also found in beef, but are not as harmful as saturated fatty acids (SFA) (Zhang *et al.*, 2008).

1.2. Problem statement

All over the World, consumers are increasingly becoming concerned about production of safe meat with no undesirable effects on their health (Andersen and Newman, 2005). The consumption of dietary fats have long been associated with chronic diseases such as obesity, diabetes, cancer, arthritis, asthma, and cardiovascular disease (Ruiz-Rodriguez *et al.*, 2010).

Although fats are used widely during processing of meat products, there is scanty information on the fatty acid profiles of hump, kidney and brisket fat and their effects on sensory properties of locally processed beef products. It is in this context that a study was conducted to determine the fatty acid content of the brisket fat, the hump fat and the kidney fat that originate from the beef carcass and to compare the sensory attributes (appearance, flavor, mealiness, taste, texture and consistency) of these fats when used in beef salami production.

1.3 Justification of the study

In recent years the fat content and fatty acid composition of foods has become more important as consumers have become more aware of the relationships between dietary fat and the incidence of lifestyle diseases, notably coronary heart disease. The United States (U.S) department of health recommends a reduction in the intake of saturated fatty acids and an increase in that of polyunsaturated fatty acids (PUFA) (Kris-Etherton *et al.*, 2003). Beef contains significant levels

of PUFA and at present, within both the scientific and producer communities there is considerable interest in production methods aimed at raising the levels of these fatty acids in beef to improve meat quality (Enser, 2001a). The ability to control fatty acid content in meat will have powerful implications for human health and nutrition. Such systems will provide a product which is healthier and more attractive to the consumer.

The findings of this study will reveal which fat (i.e. hump, kidney and brisket fat) should be given preference in processing of salami regarding its saturated and unsaturated fatty acids content and the sensory attributes (appearance, flavor, mealiness, taste, texture and consistency) it gives to the finished product. Processors may use the findings of this study to select for the part of the beef carcass that contain greater monounsaturated fatty acid deposits or with lower deposits of saturated fat, and thereby produce a healthier product for the consumer.

1.4. Objectives of the study

1. 4. 1. General objective

To determine the fatty acid content of the brisket fat, the hump fat and the kidney fat from the beef carcass and to compare their effects on the sensory attributes (appearance, taste, flavor, texture and consistency) of these fats when used in making salami.

1. 4. 2. Specific objectives

1. 4. 2. 1. To compare the saturated and unsaturated fatty acids content of the brisket, hump and kidney fats from the beef carcass.

1. 4. 2. 2. To determine the relationship between fatty acid profile and sensory attributes (appearance, flavor, mealiness, taste, texture and consistency) of salami made from fats from different body sites.

1.5. Research questions

Is there a significant difference in the saturated and unsaturated fatty acid content of the brisket, kidney and hump fats of bovine meat?

Do the sensory attributes of salami vary significantly when brisket, kidney and hump fat are used separately as one of the ingredients in salami making?

CHAPTER TWO

2.0 LITERATURE REVIEW

2. 1. Composition of meat and meat products

Meat is composed of, in descending order, water, protein, fat, other water-soluble organic material and water-soluble minerals. The composition of lean meat is relatively constant over a wide range of animals. Variation is most marked in the lipid content, which may be evident as different degrees of marbling (Varnam and Sutherland, 1995).

Table 1: Chemical/nutrient composition of selected raw and processed food products

	Product	Water	Protein	Fat	Ash	Calories/100 g
FRESH	Beef (lean)	75.0	22.3	1.8	1.2	116
	Beef carcass	54.7	16.5	28.0	0.8	323
	Pork (lean)	75.1	22.8	1.2	1.0	112
	Pork carcass	41.1	11.2	47.0	0.6	472
	Chicken	75.0	22.8	0.9	1.2	105
PROCESSED	Ham sausage	68.5	16.4	11.1	0	170
	Frankfurters	63.0	14.0	19.8	0.3	240
	Lean meat mix	72.9	18.0	3.7	0	110
	Salami	33.9	24.8	37.5	0	444

Source: Heinz and Hautzinger, 2007

Water content can be decreased (dried or dehydrated meat), and it varies in many processed meats such as sausages, salamis, bacon and ham. During these processes, care must be taken to protect the nutritional and organoleptic (taste, smell, texture and appearance) properties of the meat (Heinz and Hautzinger, 2007).

Meat is considered, justifiably, as a high protein food. Of the essential amino acids, meat supplies substantial quantities of lysine and threonine and adequate quantities of methionine and tryptophan (Varnam and Sutherland, 1995). Meat is of relatively high lipid content, which is of dietary significance in provision of energy, especially for persons engaged in heavy labor. However, the lipid content of meat has been associated with obesity and atherosclerosis (Varnam and Sutherland, 1995). Muscle tissue in general is an excellent source of some of the B-complex vitamins especially thiamine, riboflavin, niacin, pyridoxine (vitamin B_6) and cyanocobalamin (vitamin B_{12}). The B-complex vitamin content of meat, however, varies according to a number of factors, including species, breed, age, sex, muscle type and general health of individual animals. Vitamin A is the most important fat-soluble vitamin in meat. Contents of vitamins D, E and K are generally low in meats although levels of vitamin E are elevated where animals are fed high tocopherol diets. Lean meat is recognized as a good source of iron and phosphorus with 2.5-4mg/100g of lean beef and 229mg/100g of lean beef respectively (Lombardi *et al.*, 2005)

2. 2. Relevance of fatty acids to human health

Fatty acids have been classified into good or bad according to their degree of unsaturation or whether they are animal fat or vegetable fat. Monounsaturated fatty acids are still considered as having a neutral status. Controversy surrounds omega-6 polyunsaturated fatty acids, because even though they lower LDL cholesterol levels, excessive intakes do not appear to be correlated with cardiovascular benefit. However, the omega-3 fatty acids are known to exert cardiovascular protective effects (Lecerf, 2009).

2. 2. 1. Association between saturated fats and human diseases

The link between diet and diseases is complex and difficult to unravel. This is because our diet is made up of lots of different foods and nutrients. Fats are a necessary part of our diet but high-fat diets can increase our risk of cancer, heart disease and other conditions (Simopoulos, 2002).

2. 2. 1. 1. Saturated fat and cancers

High intake of red meat has been associated with increased risk of colorectal cancer in Western countries. There has been much interest in the role of n-3 polyunsaturated fatty acids (PUFA) in colorectal cancer prevention, but epidemiological findings are limited and

inconsistent (Kimura *et al.*, 2007). Colorectal cancer is one of the most common cancers in the world, accounting for nearly 10% of all incident cases. In Japan, mortality from and incidence of colorectal cancer, especially of colon cancer, have increased markedly over the last decades (Dezawa *et al.*, 2004), and it has been argued that the increase is primarily due to westernization of the Japanese diet. Of the dietary factors possibly linked with colorectal cancer, fat intake has long been a matter of interest (Kimura *et al.*, 2007). It remains uncertain whether saturated or animal fat is related to increased risk of colorectal cancer. However, high intake of red meat has been implicated as being associated with an increased risk of colon or colorectal cancer (Riboli *et al.*, 2002).

Smoking is an important risk factor for pancreatic cancer. In prospective studies, neither meat nor saturated fat was linked to incidence of this cancer in women but both have been linked to cancer in men (Doyle, 2004). Prospective studies generally have not reported links between dietary fat and lung cancer among smokers or non- smokers. However, saturated fat was linked to this cancer in male smokers in one study and dietary ham and sausage were linked to cancer in nonsmoking females in another study (Doyle, 2004).

2. 2. 1. 2. Saturated fats and cardiovascular diseases

Cardiovascular disease is a three-stage process involving injury to the inner endothelial cell layer of arteries, buildup of atherosclerotic plaque at the site of injury, and formation of a blood clot that occludes the narrowed arteries. Dietary saturated fat may affect progression of plaque buildup and plaque formation. Non dietary factors that affect the atherosclerotic process include heredity, gender, and general level of aerobic fitness (Doyle, 2004).

Increased incidence of or death from heart disease is associated with a higher saturated fat intake; and a high intake of dietary fibre weakens the association with saturated fat but the results differ for males and females (Doyle, 2004).

The levels of serum low-density lipoprotein (LDL) cholesterol are correlated with dietary total or saturated fat levels (Doyle, 2004). However, some individuals do not respond to low saturated fat diets with decreased levels of LDL cholesterol (Doyle, 2004). The consumption of trans fatty acids raises levels of low-density lipoprotein (LDL) cholesterol, reduces levels of high-density lipoprotein (HDL) cholesterol, and increases the ratio of total cholesterol to HDL

cholesterol, a powerful predictor of the risk of Coronary Heart Disease (CHD) (Mozaffarian *et al*., 2006). Trans fats also increase the blood levels of triglycerides as compared with the intake of other fats (Mensink *et al*., 2003), increase levels of lipoprotein (Lp), and reduce the particle size of LDL cholesterol (Mauger *et al.*, 2003), each of which may further raise the risk of CHD.

2. 2. 1. 3. Saturated fats and obesity

Certainly excess calories from fats will cause weight gain if there is no compensatory increase in exercise to utilize this energy intake. Dietary fat may also affect food intake and fat deposition (Doyle, 2004) but the physiological effects of a high fat diet are complex and depend on an individual's activity level and genetics.

2. 2. 1. 4. Saturated fats and diabetes

A high fat diet generally has a negative effect on glucose metabolism and induces diabetic changes in laboratory animals (Doyle, 2004). Epidemiological studies in humans also implicate high-fat diets in the development of diabetes (Doyle, 2004).

2. 2. 2. Beneficial effects of fats to human health

2. 2. 2. 1. Conjugated Linoleic Acid

The natural sources of conjugated linoleic acid (CLA) are red meats and fatty dairy products, which are mainly bovine derivatives (Eynard and Lopez, 2003). Conjugated linoleic acid is a collective term for a mixture of positional and geometrical isomers of linoleic acid, in which the two double bonds are conjugated (Kritchevsky, 2000). Unlike the non-conjugated *trans* polyunsaturated fatty acids, CLA is recognized as possessing health benefits (Scimeca and Miller, 2000). This has been found to contain both anti-atherogenic and anti-carcinogenic properties (Wilson *et al.*, 2000). Numerous beneficial effects are attributed to CLA, as in slowing down or even preventing tumor development (Eynard and Lopez, 2003). CLA decreases body fat storage in animal models and promotes cardiovascular protection against atherosclerosis (DeLany and West, 2000). CLA mainly *cis*-9, *trans*-11, 18:2 n-6 derivatives, are shown to consistently produce antitumor effects (Rudel, 1999).

Dietary CLA can inhibit 1, 2-dimethylhydrazine-induced colon carcinogenesis by a mechanism probably involving increased apoptosis (Park *et al.*, 2001). There is a possible chemo-preventive activity of CLA in the early phase of colon tumorigenesis through modulation of cryptal cell proliferation activity and apoptosis. It is also possible that one of the antitumoral effects ascribed to CLA may be linked to its capability of weight reduction (Eynard and Lopez, 2003). In a short-term trials, conjugated linoleic acid (CLA) was shown to reduce body fat mass (BFM) and increase lean body mass (LBM), but the long-term effect of CLA was not examined (Gaullier *et al.*, 2004).

2. 2. 2. 2. Polyunsaturated fatty acids (n-3 and n-6 PUFAs)

There are two classes of PUFA: omega-3 (n-3) and omega-6 (n-6) fatty acids. The distinction between n-6 and n-3 fatty acids is based on the location of the first double bond, counting from the methyl end (CH_3^-) of the fatty acid molecule. In the n-6 fatty acids, the first double bond is between the 6^{th} and 7th carbon atoms and in the n-3 fatty acids the first double bond is between the 3^{rd} and 4^{th} carbon atoms (Clandinin *et al.*, 2000).

Omega-6 (n-6) and omega-3 (n-3) fatty acids are known as essential fatty acids (EFAs) because humans like all mammals, cannot make them and must obtain them from their diet. The main essential omega-3 fatty acid is alpha-linolenic acid (ALA), and the main essential omega-6 fatty acid is linoleic acid. Specifically, their beneficial effects have been shown in the prevention and management of coronary heart disease, hypertension, type 2 diabetes, renal disease, rheumatoid arthritis, ulcerative colitis, Crohn's disease, and chronic obstructive pulmonary disease (Simopoulos, 2002). However, the beneficial effects of PUFA depend on the ratio of the fatty acid omega 6 (n-6) to omega 3 (n-3); it is generally accepted that the ideal proportion of n-6 to n-3 is around 4:1 (Schaefer, 2002). The ratio of n-6:n-3 PUFA is also a risk factor in cancers and coronary heart disease, especially the formation of blood clots leading to heart attack (Enser, 2001b) . The metabolic products of omega-6 acids promote inflammation, blood clotting and tumor growth, the omega-3 acids act entirely opposite (Simopoulos, 2002). It is important to maintain a balance of omega-3 and omega-6 in the diet as these two substances work together to promote health (Simopoulos, 2002).

2. 3. Fatty acid composition of beef

Lean beef has an intramuscular fat content of around 5% or less with approximately 47%, 42% and 4% of total fatty acids as saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA), respectively (French *et al.*, 2000). The ratio of the polyunsaturated to saturated fatty acids (P:S) for beef is typically low at around 0.1 (Scollan *et al.*, 2001), except for double muscled animals which are very lean (<1% intramuscular fat) where the P:S ratio is typically 0.5-0.7 (Raes *et al.*, 2004).

The n-6: n-3 ratio for beef is beneficially low, typically less than 3. This reflects the considerable amounts of n-3 PUFA in beef, particularly alpha-linolenic acid (18:3n -3) and the long chain PUFA, eicosapentaenoic acid (20:5n-3, EPA) and, docosahexaenoic acid (22:6n -3, DHA). The predominant SFA are 14:0, 16:0 and 18:0. Of the total SFA, 0.3 are represented by stearic acid (18:0) (Wood *et al.,* 2008). SFA are recognized to influence plasma cholesterol, though 18:0 is regarded as neutral in this regard and 16:0 is not hyper cholesterolemic if the diet contains high levels of linoleic acid (18:2n-6) (Clandinin *et al.,* 2000). Myristic acid (14:0) is regarded as more potent than palmitic acid (16:0) in raising plasma lipids (Zock *et al.,* 1994). Linoleic and α-linolenic acids are the main PUFA while oleic acid (18:1n-9) is the most prominent MUFA (Wood *et al.,* 2008). The PUFA and MUFA are generally regarded as beneficial for human health and there is recent evidence of beneficial effects of 18:1 trans-11 (Corl *et al.,* 2003), though other work suggests negative effects (Clifton *et al.,* 2004). The main CLA isomer in beef is cis-9, trans-11 representing 72-90 % of total CLA isomers. Biological effects of two isomers, cis-9, trans-11 and trans-10, cis-12 CLA, have been extensively investigated and anti-carcinogenic and anti-atherogenic effects of cis-9, trans-11 and the anti-obesity effects of trans-10, cis-12 have been documented (Dannenberger *et al.*, 2004).

2. 4. Effect of breed and diet on fat deposition in beef muscle and adipose tissue

Intramuscular fat (IMF) as well as fatty acid composition are affected by numerous factors including age, breed, gender and in particular nutrition (Nürnberg *et al.*, 2011). These factors are discussed below in details.

2. 4. 1. Effects of breed on fatty acids in beef

Breeds may differ in their fat composition (both total intramuscular fat and individual fatty acids) of beef. The fatty acid composition of beef is influenced by genetic factors although to a lower extent than dietary factors. Though these breed differences are generally small, they nevertheless reflect differences in underlying gene expression or enzymes involved in fatty acid synthesis (Wood *et al.,* 2008). There is a strong negative exponential relationship between fatness and P:S ratio (Luan *et al.,* 2001*).* As the content of SFA and MUFA increase faster with increasing fatness than does the content of PUFA, the relative proportion of PUFA and the P: S ratio decrease with increasing fatness. To some extent fatty acid composition of deposited fat is related to breed partly because of stage of maturity (Pitchford *et al.,* 2002). The early maturing breeds deposit fat earlier in life and at maturity contain more fat throughout the body (including the edible muscles) than the late maturing breeds (Wheeler *et al.,* 2005). Dairy breeds deposit more intramuscular fat in relation to total fat while having a learner carcass than more traditional beef breeds (Scollan *et al.,* 2001).

2. 4. 2. Effects of diet on fatty acids in beef

Dietary effects are relatively well recognized and in general the composition of the beef lipids reflects the fatty acid composition of the diet. For example, giving animals diets rich in C18: 3 n-3 or C20: 5 n-3 and C22:6 n-3, such as linseed and fish oil/meal, respectively increases the levels of these PUFA in the meat (Scollan *et al.,* 2001). Feeding pasture rich in C18:3n-3 relative to concentrates rich in C18:2n-6, results in higher concentrations of n-3 PUFA in muscle lipids (Dannenberger *et al.*, 2004). Grass relative to concentrate feeding not only increases C18:3n-3 in muscle phospholipids but also eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Nuernberg *et al*., 2005). Concentrates rich in C18:2n-6 lead to higher concentrations of C18:2n-6 and associated longer chain derivatives (C20:4n-6) (Realini *et al.,* 2004). An increase in the proportion of grass in the diet decreases the concentration of SFA, increases P:S and n-3 PUFA concentration and decreases n-6:n-3 PUFA (French *et al.*, 2000).

Pasture feeding does result in higher concentrations of more oxidizable n-3 PUFA in muscle lipids, but the meat is more resistant to lipid oxidation than concentrate (grain) fed-beef (Warren *et al.*, 2002). The differences in the fatty acid composition of meat induced by feeding grass

compared to concentrates have been reported to affect beef flavour (Larick and Turner, 1990). The scores for flavour descriptors changed when cattle previously grass-fed were changed to a maize diet in a feedlot. Comparison of the effects of forage-based diets with concentrate (usually grain) based diets showed little difference in marbling between grain-fed and grass-fed beef at the same carcass weight (Muir *et al.*, 1998).

2. 5. Salami processing and role of different ingredients

Salami is a large, thick, highly seasoned, and often cured type of sausage, Italian in origin and usually served cold in thin slices. Salami is processed using lean minced beef, Ice, Sodium Nitrate, phosphate, salt, Spanish paprika, spice mix, garlic and beef fat. In Uganda it's not highly marketed as a lot of consumers are not aware of it compared to the consumption of sausage (Ayo *et al.*, 2012).

2. 5. 1. Phosphate

Processors add phosphate to meat for many reasons, but especially to increase the water holding capacity by forcing the protein molecules apart, which in turn allows water to move in between them, for preservation of natural flavours in processed meats, as a very good chelating agent and lend freeze/thaw stability(Knipe *et al.*, 2006). Added benefits of phosphate include: Increased moisture retention, which reduces cook-cool losses (decreasing shrinkage) thereby improving yield without a wet glossy appearance; developing an intense cured pigment by reducing nitrite residuals and inhibiting rancidity of fat and warmed-over flavour development in re-heated product. Grinding or chopping meat can form a "meat emulsion" that needs to hold up during cooking and processing. Adding phosphates enhances stability in finely chopped meat systems, such as salami or hot dogs, by influencing the pH, ionic strength, protein extraction, divalent cation binding and viscosity (Knipe *et al.*, 2006).

2. 5. 2. Garlic

Garlic (*Allium sativum* L.) has been widely used as a spice since antiquity and has acquired a reputation in the folklore of many cultures as a therapeutic agent. It has been known as an herbal remedy to prevent and treat a variety of heart diseases and metabolic diseases, such as atherosclerosis, thrombosis, hypertension, dementia, cancer, and diabetes (Amagase *et*

al., 2001). Garlic has rich organosulfur compounds and precursors (allicin, diallyl sulfide, and diallyl trisulfide). These compounds provide garlic its characteristic odor and flavor as well as most of its biological properties and have been identified as having the hypocholesterolemic effect in human and animal products (Chowdhury *et al.*, 2002).

During storage, quality attributes of the food products deteriorate due to lipid oxidation and microbial growth. Lipids oxidation is responsible for reduction in nutritional quality as well as changes in flavor (Aguirrezábal *et al.*, 2000), while microbial contamination can precipitate major public health hazards and economic loss in terms of food poisoning and meat spoilage. Thus, application of suitable agents possessing both antioxidant and antimicrobial activities may be useful for maintaining meat quality, extending shelf-life and preventing economic loss (Yin and Cheng, 2003).

Research has indicated that lipid oxidation and microbial growth in meat products can be controlled or minimized by using either synthetic or natural food additives (Mielnik *et al.*, 2003). However, consumers are concerned about the safety of synthetic food additives. This concern has led to a great interest in natural additives. Natural agents i.e. garlic possessing antioxidant and antimicrobial properties have the advantage of being readily accepted by consumers, as they are considered natural. Garlic is one of the most commonly used ingredients as a flavor enhancer. In addition to flavoring the foods, garlic is appreciated for its medicinal properties. Garlic has a wide spectrum of actions; not only antibacterial, antiviral, antifungal and antiprotozoal, but also has beneficial effects on the cardiovascular and immune systems (Harris *et al.*, 2001). During the last decade, the antimicrobial activity of garlic and garlic-derived organosulfur compounds was widely investigated against both food spoilage bacteria and food-borne pathogens (Leuschner and Ielsch, 2003). Besides its antimicrobial effect, garlic showed effective antioxidant activity (Jackson *et al.*, 2002).

2. 5. 3. Paprika spice

Paprika is a red powder that is made from grinding the dried pods of mild varieties of the pepper plant known as *Capsicum annuum (L.*). The most commonly produced paprika is made from the sweet red pepper also called the tomato pepper. The spice is used to add color and flavor to meat products. It is often used for garnishing purposes. Paprika is rich in vitamin C and other

antioxidants and hence it is beneficial to the body as it improves the immune system (Kaur and Kapoor, 2008). It has some therapeutic qualities. The hotter the paprika, the higher the amount of capsaicin, a highly beneficial and medicinal component of the spice, whose anti-inflammatory and antioxidant effects may lower the risk of cancer (Kaur and Kapoor, 2008).

2. 5. 4. Salt

Salt is added to meat products for a variety of technical reasons; it has an essential function in terms of flavour, texture, shelf-life and microbiological safety (Desmond, 2006) but the main function of salt in cured products is to add flavor. However; even at low concentrations salt has some preservative action. Salt levels are dependent on consumer's taste, but a two to three percent concentration in the product is about right (Albarracín *et al.*, 2011). Processed meat products comprise one of the major sources of sodium in the form of sodium chloride (salt). The food industry has been under pressure from the Food Standards Agency (FSA) to deliver reductions in the salt intake of the population through the introduction of lower salt levels in processed foods (Gilbert and Heisler, 2004). Intake of dietary sodium has been linked to hypertension and consequently increased risk of cardiovascular disease (Desmond, 2006).

2. 5. 5. Meat curing salt

Curing is a food preservation and flavoring process, especially of meat or fish, done by the addition of a combination of salt, nitrates, nitrite or sugar. Nitrates and Nitrites are fundamental components of the global nitrogen cycle and are therefore found throughout the environment. Nitrates and nitrites are compounds that contain a nitrogen atom joined to oxygen atoms, with nitrate containing three oxygen atoms and nitrite containing two. In nature, nitrates are readily converted to nitrites and vice versa. Both are anions or ions with a negative charge. They tend to associate with cations, or ions with a positive charge to achieve a neutral charge balance (Lewis, 2005). Sodium salts of the nitrate ($NaNO_3$) and nitrite ($NaNO_2$) are the most extensively used of all food additives (Stevanović and Šentjurc, 2000). Nitrates and nitrites in cured meat and meat products play a multipurpose role; in addition to effectively inhibiting the growth and toxigenic effect of *Clostridium botulinum*, nitrite is responsible for the development of typical cured-meat color and flavor, and also functions as an antioxidant (Rincón *et al.*, 2008), retarding the development of rancidity, off-odors and off-flavors during storage, inhibiting the development of

warmed-over flavor and preserving flavors of spices and smoke (Sikorski and Kołodziejska, 2002). Sodium nitrite, rather than sodium nitrate, is the most commonly used for curing. In a series of normal reactions, nitrite is converted to nitric oxide (Grigioni *et al.*, 2000). Nitric oxide combines with myoglobin, the pigment responsible for the natural red color of uncured meat forming nitric oxide myoglobin, which is a deep red color (as in uncooked dry sausage). This changes to the characteristic bright pink color normally associated with cured and smoked meat when heated during the smoking process (Feiner, 2006). When used, sodium nitrate ($NaNO_3$) does not contribute directly to the formation of the red curing color but rather, is reduced to sodium nitrite ($NaNO_2$), thus providing nitric oxide (NO) by the reactions above, which results into the formation of the characteristic pink cured meat color (Armenteros *et al.*, 2009). Examples of such cured products are; ham, corned beef, bacon, salami, and sausage.

2.6. Sensory evaluation methods

Sensory analysis is a multidisciplinary science that uses human panelists and their senses of sight, smell, taste, touch and hearing to measure the sensory characteristics and acceptability of food products (Tharani and Amirthaveni, 2013). There is no one instrument that can replicate or replace the human response, making the sensory evaluation component of any food study essential. Different sensory evaluation methods can be used depending on the type of information which is targeted, whether information on consumer likes and dislikes preferences or acceptance.

2.6.1. Preference Tests

Preference tests allow consumers to express a choice between samples; one sample is preferred and chosen over another or there is no preference. The paired-preference test is the simplest preference test but category scales and ranking tests are also often used to determine preference. Panelists are asked which of two coded samples they prefer. Panelists are instructed to choose one, even if both samples seem equal. Results are analyzed using a 2-tailed binomial test. The 2-tailed test is appropriate since either sample could be preferred; and the direction of the preference cannot be determined in advance. The number of judges preferring each sample is totaled and the totals tested for significance (Watts *et al.*, 1989).

2.6.2. Acceptance Tests

Acceptance tests are used to determine the degree of consumer acceptance for a product. Category scales, ranking tests and the paired-comparison test can all be used to assess product acceptance. Acceptance of a food product usually indicates actual use of the product (purchase and eating). Panelists are asked to rank coded samples for acceptance in order from the least acceptable to the most acceptable. Ties, where the samples are given equal acceptance ranks, are not usually allowed. For data analysis, the ranks assigned to each sample are totaled. The samples are then tested for significant differences by comparing the rank totals between all possible pairs of samples using the Friedman Test (Watts *et al.,* 1989)

2.6.3. Hedonic Tests

Hedonic tests are designed to measure degree of liking for a product. Category scales ranging from like extremely, through neither like nor dislike, to dislike extremely, with varying numbers of categories, are used. Panelists indicate their degree of liking for each sample by choosing the appropriate category. Panelists are asked to evaluate coded samples of several products for degree of liking, on a 9-point scale. They do this by checking a category on the scale which ranges from like extremely to dislike extremely. The numerical scores for each sample are tabulated and analyzed by analysis of variance (ANOVA) to determine whether significant differences in mean degree of liking scores exist among the samples (Watts *et al.,* 1989). In this study, hedonic test was used because it indirectly measures product preference.

2.7. Fatty acid composition determination method

2.7.1 Gas Chromatography-Mass Spectrometry (GCMS) method

An aliquot of lipid solution is dried with anhydrous sodium sulfate and evaporated to dryness under nitrogen in a centrifuge tube with a teflon lined screw cap. Boron fluoride-methanol reagent is added under nitrogen, in the proportions 1 ml reagent per 4-16 mg of lipid, and the tubes closed with a teflonlined screw cap. The tube is then heated in a boiling water bath for the requisite time, cooled, and opened. The esters are extracted by adding 2 volumes of hexane, then one volume of water, shaken briefly, and centrifuged until both layers are clear (Almeida *et al.,* 2006)

One μL of the mixed hexane extracts is injected splitless (the split is opened after 4 min), and chromatographed on a 25 m x 0.25 mm (i.d) fused silica column with polyethylene-glycol (PEG) as stationary phase, with a thickness of 0.2 μm (CP-WAX 52CB Chrompack) and hydrogen gas at 20 psi as mobile phase. The column is mounted in clarus 500 OR 480 gas chromatograph equipped with and a flame-ionisation detector (FID). The injector temperature is set at 260^{o}C and detector at 330^{o}C. The oven is programmed as follows: 90^{o}C for 4 min, 30^{o}C/min to 165^{o}C, then 3^{o}C/min to 225^{o}C where it will be left to isothermal for 10.5 min before cooling for the next run (Almeida *et al.*, 2006)

The FAMEs are determined through their retention times in comparison to a standard mixture. Mass spectrometry GCMS (Agilent 6890N is used for those peaks that are not identified using standard mixture). Quantification of FAME is based on the internal standard technique, using nonadecanoic acid (19:0) as internal standard. The FAMEs are expressed as percentages of total fatty acid (Almeida *et al.*, 2006)

2.7.2 Fatty acid profiles determination by Gas Chromatograph Flame Ion Detector (GC-FID)

This was the method used in this study because of the ease availability of reagents, the reputation and the reliability of the results from Chemiphar laboratory where the experiment was conducted as far as laboratory analysis is concerned. This method is clearly explained in 3.6.section of the materials and methods.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3. 1. Study location

The study was conducted in Kampala, the capital city of Uganda, which represents a fairly balanced mix of beef consumed across the country. Most of the processing of the salami was done at Uganda Meat Industries (UMI)/Top Cuts limited a private enterprise, while the laboratory analysis was conducted at Chemiphar laboratory.

3. 2. Sample collection

Fats (brisket, kidney and hump fat) were continuously collected from different cattle carcasses slaughtered at UMI/Top Cuts Ltd. abattoir. Fats were collected regardless of the breed, age, sex and diet of the cows for the study to have a clear image of the situation as it is done the same way by the meat industry in Uganda where the study was conducted. Fat free meat used in processing was also obtained from the slaughtered animals in the same abattoir.

3. 3. Sample size and sampling strategy

The sample included fats obtained from three different parts/locations of the beef carcass. These were brisket, kidney and hump fat. From different carcasses, fats were continuously collected until a total of three kilograms for each was obtained, which is the quantity required by the salami processing recipe. Top cuts Ltd abattoir was used for fat collection. The daily collected fat was vacuum packed to avoid lipid oxidation and microbial contamination. Fats were finally minced separately using a meat mincer and used to make three different salamis where each fat was added together with other ingredients used in salami processing.

3. 4. Salami Processing

In addition to fat collection, preliminary processing (mincing, chopping, vacuum packaging and storage) was done at Uganda Meat Industries/Top cuts Limited abattoir. Three different salamis were processed and the only difference between them was the source of fat used. The first batch was made using brisket fat, the second one using kidney fat and the third using hump fat; the other ingredients listed in the salami recipe remained the same. Each batch was produced in duplicate to avoid any kind of bias.

3. 4. 1. Salami ingredients and recipe

The following are the ingredients and their volumes used in salami processing: lean minced beef: 11kg, beef fat: 3 kg, ice blocks: 3 kg, sodium nitrate (NaNO3): 50 g, phosphate: 100 g, salt: 200 g, spanish paprika: 100 g, spice mix: 300 g and garlic: 500 g. A total output of 50.6 kg of salami was obtained from three runs. The brisket fat salami batch produced a total of 16 kg, the kidney fat salami batch had 18.1 kg while the hump fat salami batch produced 16.5 kg of salami. Note that this output was from the equal amount of input (ingredients) and it's the average of duplicate processes for each of the salami. The variation in salami output resulted from the difference in fats water holding capacity.

3. 4. 2. Salami processing procedure

The salami was processed using minced lean beef which was placed into a bowl cutter then the machine was run as shown in figure 1. Ice blocks were continuously added to cool the machine as it generates heat when working. Sodium nitrate (NaNO3) was added to stabilize the bright red color of cured meat. Meat curing is a preservation method where nitrate or nitrite salts are added to meat for the development of typical cured-meat color. Nitrite (NaNO2) combines with myoglobin to form nitrosomyoglobin, a bright red compound. The nitrosomyoglobin is heat stable i.e. when the meat is heat treated the bright red color remains (Rincón *et al.,* 2008). Salt was added next, the main role of salt is to add on taste, texture and firmness of the finished product. Spice mix was added and the speed of the bowl cutter increased. While the machine was running, the dough was mixed using a scrapper. Beef fat was added to the dough, followed by paprika and finally garlic. After getting a fine emulsion, the machine was stopped and salami mixture was transferred into the filling machine and stuffed into poloni casings. The stuffed salami were heated in a boiler containing warm water for two hours; the water temperature inside the boiler was monitored at 65^{o} C for water not to boil otherwise the salami could shrink. Salamis were cooled using tap water and chilled over night to stabilize. Salami were sliced the following day and vacuum packed into golden packs and then stored under refrigeration temperature (0-4^{o}C).

3.4.3 Salami processing flow diagram

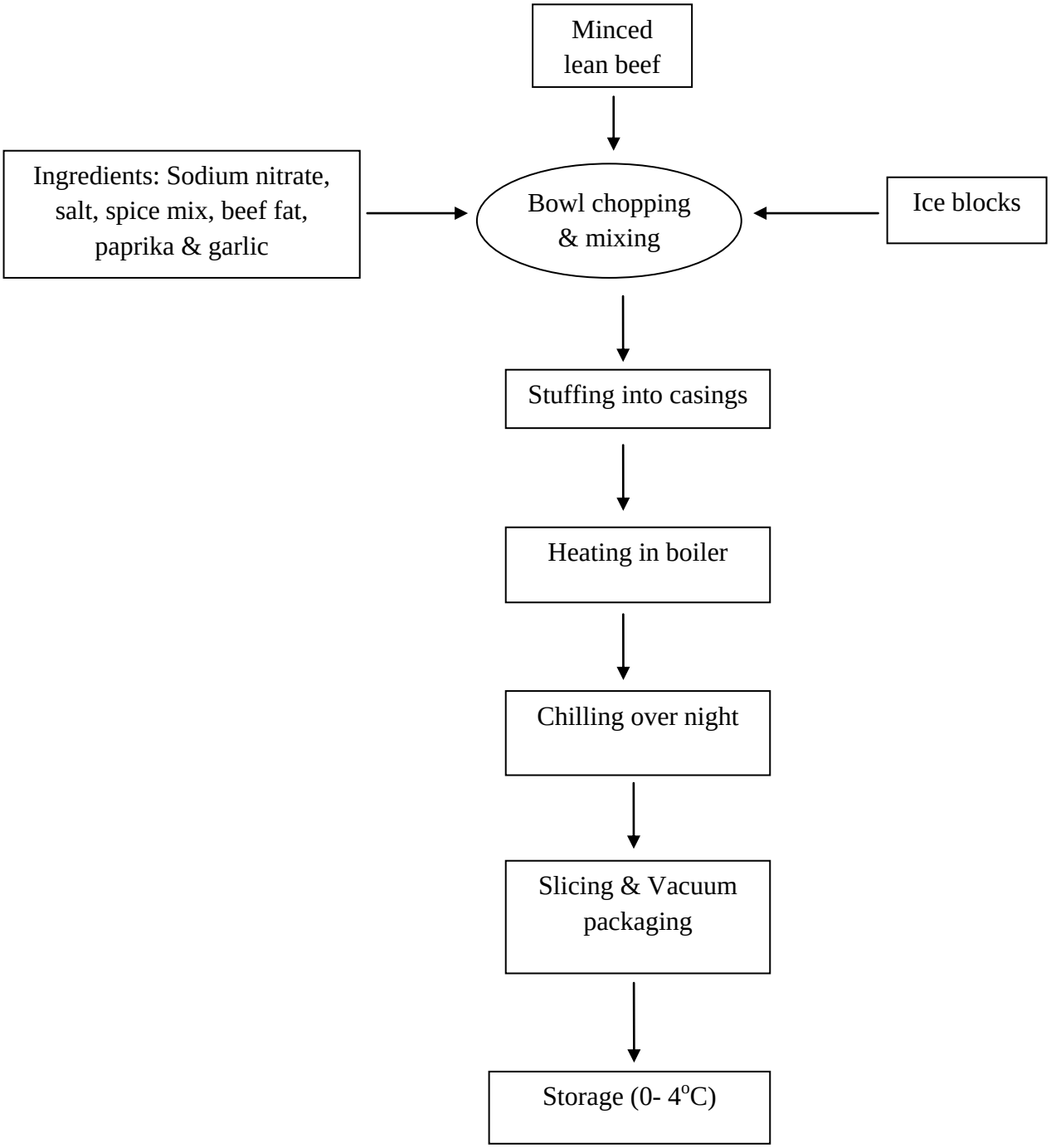

Figure 1: Salami process flowchart

3. 5. Sensory evaluation

The three processed salami samples e.g. hump, kidney and brisket fat salami were evaluated for the degree of product liking using hedonic test, one of the consumer-oriented sensory evaluation tests. Sensory attributes such as appearance, taste, flavor, texture, mealiness and product consistency were marked by ten panelists on a 1 to 9 hedonic scale (where 1 is dislike extremely and 9 is like extremely) (Raharjo, 1996). The panelists were selected from Top cuts staff members, College of Veterinary Medicine, Animal Resources and Bio-security (COVAB) lecturers and the students of Masters in Animal Product Processing Entrepreneurship and Safety (MAPPES). The panelists were first taken through the meaning of the sensory attributes before the actual sensory evaluation took place. Each panelist was given three slices of each sample (one after another) and asked to taste one sample at a time and recorded the response. Between each sample panelists were required to rinse their mouths with mineral water to avoid interference of samples. The numerical scores for each sample were tabulated and analyzed by analysis of variance (ANOVA) to determine whether significant differences in mean degree of liking exist among the samples.

The sensory attributes evaluated in this study are defined in table 2:

Table 2: Sensory attributes definitions

Sensory attribute	Definition
Appearance	Food product appearance is the visual identification based on color, size, shape and form which affect its expected pleasantness and acceptability of food products (Hutchings, 2003). For this study, appearance referred to the product color uniformity.
Flavor	Flavor refers to a combination of taste, smell, and mouthfeel with the volatile components perceived when the food is placed in the mouth mainly contributing to the overall perception of food (Dainty and Blom, 1995). For this study, flavor focused mainly on the perception of smell form the different evaluated salami (hump, kidney, and brisket fat salami).
Texture	Texture is the sensory manifestation of the structure or inner makeup of the food in terms of their reactions to the applied forces, which are measured as hardness, firmness, tenderness, juiciness, crispness, and wateriness by the kinesthetic sense in the muscles of hands, finders, tongue, jaw, or lips (Meilgaard *et al.*, 2007). In this study, texture referred to the panelists' perception of firmness when the product was eaten.
Taste	Taste relies on the perception of the four basic sensations of salty, sweet, sour and bitter on the tongue (Carterette, 1978). For this study salami were ranked according to their levels of salt and the good taste was the one which was not much salty, not sweet, not sour neither bitter.
Mealiness	Product mealiness refers to the ease of chewing. It consisted in whether the salami was hard to break or breaks up too much in the mouth (Watson *et al.*, 2008). The same meaning applied for this study.
Consistency	Meat product consistency refers to the property of holding/sticking together and retaining the shape (Issanchou, 1996). For this study, salami consistency referred to the harmonious uniformity among the product particles, in other words the way different used ingredients were able to stick together leaving no empty spaces in the final product.

3. 6. Determination of the fatty acid profile

Fatty acid profiles of the brisket, kidney and hump fat were determined using GC-FID. GC 3800 was the model used by the GC-FID for FAs determination and the analysis was done in duplicate.

3. 6. 1. Test procedure

3. 6. 1. 1. Extraction of the fat

Samples were extracted with chloroform. Ten grams of sample were accurately weighed into a 250 ml flat bottomed flask and homogenized. One hundred milliliters of 99.5% chloroform were added and mixed for 90-120 seconds with the Ultra turrax. The mixture was allowed to settle and separate for 10 minutes. The chloroform was filtered over a 1.5 µ Whatman filter paper with 99% Sodium sulphate. Ten milliliters of filtrate were drawn out with a pipette into a test tube and evaporated to dryness under nitrogen at 40oC (Kirk and Sawyer, 1991). The reagents used were acquired from Sigma Aldrich Ltd.

3. 6. 1. 2. Derivatization

After extraction of the fat, 2 ml of 98% diethyl ether was added to the same test tube from the concentration block (evaporator) and 0.5 ml of 5% potassium hydroxide (KOH) in methanol (W/V) solution was added. The mixture was shaken with a vortexor until all fats were completely dissolved. The reaction was left for 15 minutes, during this period the solution became a little cloudy, because of soap forming (Horwitz and Latimer, 2000).

3. 6. 1. 3. Washing

The procedure was carried out according to the one described by Horwitz and Latimer (2000) in which 2 ml of demineralized water were added and the test tube filled by about 10 ml of 95% n-hexane, the solution was shaken and the phases let to separate. The top layer (the hexane layer) was transferred into a second test tube and 2 ml of water were added to it, shaken to mix and the phases let to separate. The top layer was transferred to a third test tube and again 2ml of water were added to it, shaken and the phases let to separate. The top layer was transferred to a fourth test tube and this time 99% Sodium sulphate (Na_2SO_4) was added to the test tube to remove all

the remaining residues of KOH and diethyl ether. The solution was shaken and allowed to rest for a few minutes. One ml of the sample was transferred to a GC-auto sampler vial.

3. 6. 1. 4. Gas Chromatograph (GC) Analysis

The sample was ready for GC-analysis. The HT8, 25 m X 0.22 mm ID X 0.25 μm film column was installed. The oven temperature was set first at 140^{o}C and allowed to rise up 240^{o}C and kept for 5 minutes. The carrier gas used was helium, the FID (Flame Ion Detector) and the injector temperatures were set at 260^{o}C. After the GC was set; the samples, the procedure blank and the control standards were run on the GC. The samples were integrated and peaks assigned names according to the control standard and the retention times. Each peak represented a fatty acid.

FAME Mix rapeseed oil and Supelco^{TM} 37 component FAME Mix control standards from Sigma Aldrich, USA were used to assign names to the fatty acid peaks and to show their retention times. After integration and assigning names to the peaks, the different compositions of the FAs (SFA, MUFA and PUFA) were calculated automatically on the computer. The results were reported in percentages on a weight basis.

3. 7. Data management and analysis

Following the laboratory analysis and the sensory evaluation, data were obtained and stored in excel sheet. Data from sensory evaluation were later analyzed for significant difference in mean of each of the studied sensory attributes using SPSS data analysis software.

Total saturated, monounsaturated and polyunsaturated fatty acids composition of the hump, kidney and brisket fat were statistically analyzed for significant difference using multiple comparison. A mean for each total was generated to enable the comparison possible. The level of significance was considered to be significant different for values <.05 and not significant different for values >.05.

CHAPTER FOUR

4.0 RESULTS

4.1. Comparison of sample sensory properties' mean ranks

Results from sensory evaluation showed that the hump fat salami appearance ranked highest (M=7.5±0.7). There was no significant difference between the appearance of the hump and the one of the kidney fat salami with (PV =.634>.05). No significant difference was noted between the hump and brisket fat salami at (PV =.662>.05). Further results, revealed no significant difference between the kidney and the brisket fat salami appearance with (PV =.716>.05). Hump fat salami flavor ranked highest with (M=6.7±1.16). No significant difference was found between the hump and kidney, hump and brisket, kidney and brisket fat salami flavor with (PV =.484>.05), (PV= .225>.05) and (PV =.202>.05) respectively. In terms of mealiness, kidney and brisket fat salami ranked equally with (M=6.9±0.738). A significant difference was noted in mealiness of the fats with (P=.033<.05). Results also showed that hump fat salami ranked highest in taste with (M=7.5±0.527). When it came to texture, brisket fat salami ranked the highest with (M=7.2±0.632). As far as product consistency was concerned, hump and brisket fat salami ranked equally with (M=7.3±0.949). There was no significant difference found in the taste, texture and consistency of the hump, kidney and brisket fat salami.

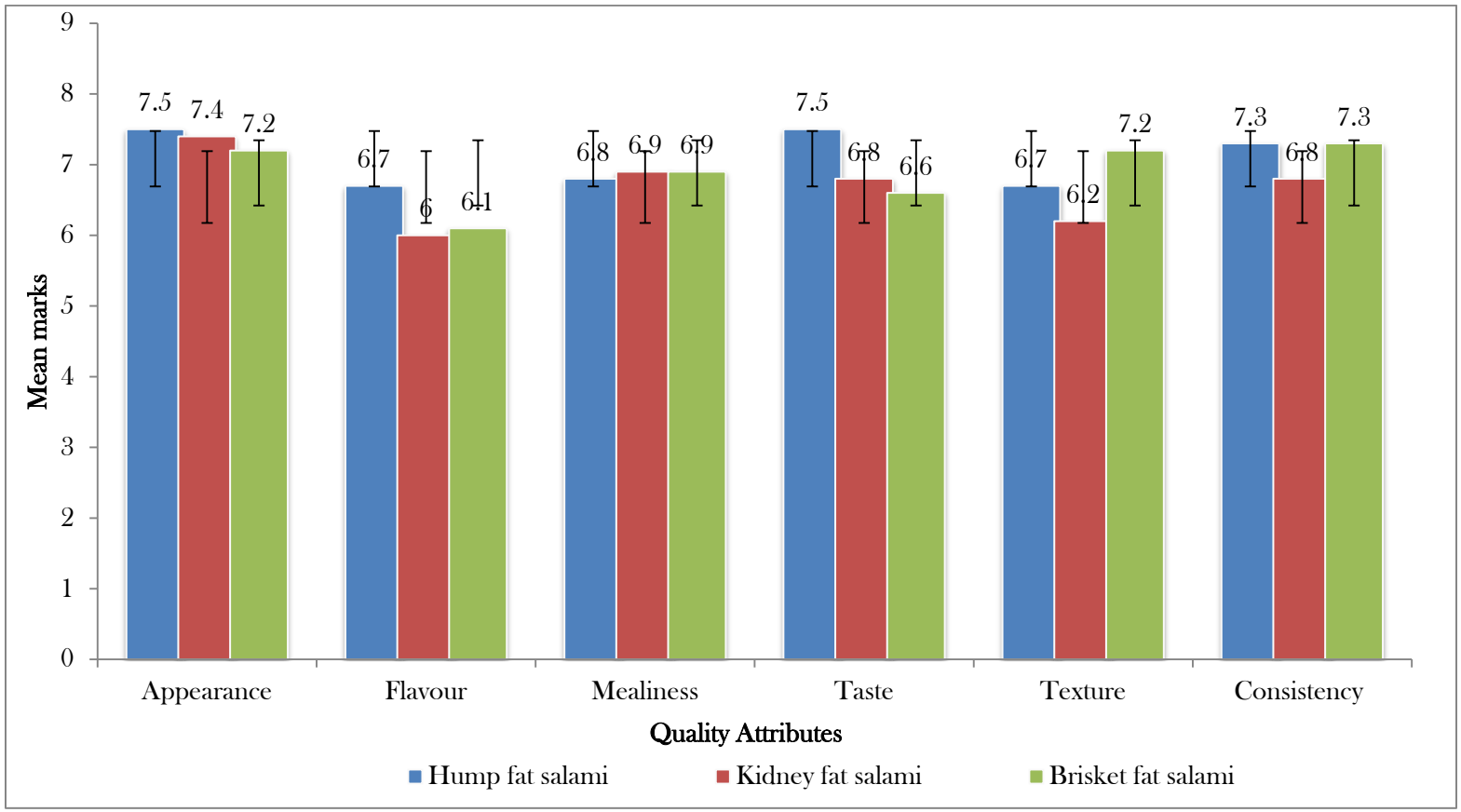

Figure 2: Sensory properties mean ranks of hump, kidney and brisket fat salami

4.1.1 Overall product liking

Means in Figure 3 show the hump fat salami with the highest rank of (M=7.08) which rates as like moderate on the used sensory evaluation form. This signified that Hump fat salami was the most liked product among others (kidney and brisket fat salami) in terms of the studied sensory properties. Brisket and kidney fat salami were liked slightly on the scale of panelists with (M=6.88) and (M=6.68) respectively.

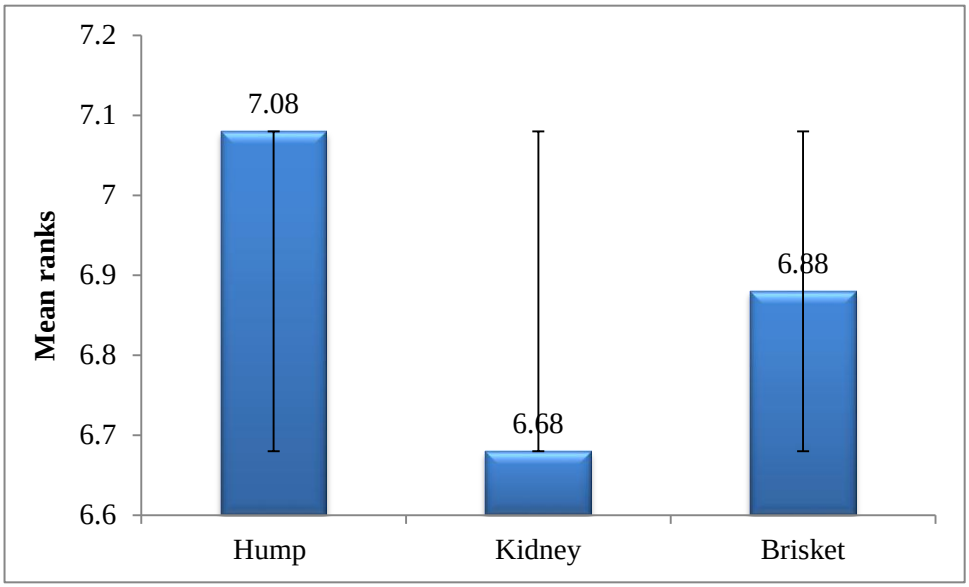

Figure 3: Most liked salami

4. 2. Fatty acids methyl ester composition of the hump, kidney and brisket fat

4. 2. 1. Saturated fatty acids (SFAs)

Six saturated fatty acids were present in all the three fat samples: myristic (C14:0), palmitic (C16:0), stearic acid (C18:0), arachidic (C20:0), behenic (C22:0) and lignoceric acid (C24:0). Their compositions are clearly described in Table 2. Of the three analyzed fats, the hump fat was found to have a highest composition of SFAs with a total of 21.25%, followed closely by the brisket fat with 20.10% total SFA, while kidney fat had far lower levels of SFA with a total of 13.52% SFA. The total SFAs of the hump fat were significantly different from those of the kidney fat with Highest Significant Difference (HSD = 0.036 <.05). However, hump and Brisket total SFAs were not significantly different with (HSD = 0.777>.05). The same was observed in the total SFAs of the Kidney which were not significantly different from those of the brisket fat with (HSD = 0.054>.05).

Table 3: Saturated fatty acids composition of the hump, kidney and brisket fat

Individual SFA % Composition out of total fatty acids			
SFA	**Hump fat**	**Kidney fat**	**Brisket fat**
Myristic Acid (C14:0)	1.78	0.45	3.69
Palmitic Acid (C16:0)	9.35	4.49	3.54
Stearic Acid (C18:0)	3.95	1.53	5.41
Arachidic Acid (C20:0)	1.94	0.49	1.07
Behenic Acid (C22:0)	3.28	5.73	3.75
Lignoceric Acid (C24:0)	0.95	0.83	2.64
Total SFAs	**21.25**	**13.52**	**20.1**

At the individual fatty acids levels, the concentration of myristic, stearic and lignoceric acids were highest in brisket fat, followed by hump fat, and least in kidney fat. On the other hand, palmitic acid was more found in highest concentration in hump fat, followed by kidney fat and least in brisket fat, while arachidic acid was more concentrated in hump fat, followed by brisket fat and least in kidney fat. Only behenic acid showed a unique pattern, being highest in kidney fat, followed by brisket and hump fat.

4. 2. 2. Monounsaturated fatty acids (MUFAs)

The monounsaturated fatty acids found in the samples included: Oleic (C18:1), Eicosenoic (C20:1) and Erucic Acid (C22:1). Kidney fat samples were found to contain the highest total amount of the MUFAs, at 83.82% of the total MUFAs, followed by hump fat at 77.07% and then brisket fat at 75.69% total MUFAs. There was a significant difference between the total MUFAs of the hump and the ones of the kidney fat at the HSD Value (.007<.05). The same result was observed between the total MUFAs of the kidney and the brisket fat at HSD value (.004<.05). However, no significant difference was found in the total MUFAs of the hump and the brisket fat at HSD value (.339>.05).

Table 4: Monounsaturated fatty acids composition of the hump, kidney and brisket fat

Individual MUFA % Composition out of total fatty acids			
MUFA	**Hump fat**	**Kidney fat**	**Brisket fat**
Oleic Acid (C18:1)	33.02	31.12	23.84
Eicosenoic Acid (C20:1)	25.14	18.84	34.28
Erucic Acid (C22:1)	18.91	33.86	17.57
Total MUFAs	**77.07**	**83.82**	**75.69**

The profiles for individual MUFAs showed that oleic acid was highest in hump fat, followed closely by kidney fat, and a distant amount in brisket fat. Eicosenoic acid was most concentrated in brisket fat, followed by hump fat and lowest in kidney fat. Erucic acid was found to be highest in kidney fat, followed by at a distant amount by hump and brisket fat.

4. 2. 3. Polyunsaturated fatty acids (PUFAs)

Only two polyunsaturated fatty acids linoleic acid (C18:2) and alpha linolenic acids (C18:3) were present in the fat samples analyzed as shown in Table 4. Their concentrations were not as much as MUFAs and SFAs. Of the three fat samples, the brisket fat had the highest total PUFAs (4.21%) as shown in table 4. Both linoleic and alpha linolenic acids were found to be highest in brisket fat with 2.13% and 2.08% respectively. A significant difference was observed between the total PUFAs of the hump and those of the kidney fat with HSD Value (.008<.05). In addition, a significant difference was found between the total PUFAs of the hump and the ones of the

brisket fat at HSD value (.000<0.05). Finally, there was a significant difference between the PUFA of the kidney and brisket fat with HSD value (.002<.05).

Table 5: Polyunsaturated fatty acids composition of the hump, kidney and brisket fat

Individual PUFA % Composition out of total fatty acids			
PUFA	**Hump fat**	**Kidney fat**	**Brisket fat**
Linoleic Acid (C18:2)	0.48	0.62	2.13
Alpha Linolenic Acid (C18:3)	1.18	2.03	2.08
Total PUFAs	**1.66**	**2.65**	**4.21**

CHAPTER FIVE

5.0 DISCUSSION

A good meat product appearance i.e. color is determined mainly by the persistence of the bright red color of oxymyoglobin on the product surface. No significant difference in the appearance of the three different samples (hump, kidney and brisket fat salami) were noted although hump fat salami ranked highest with (M=7.5±0.707). Several factors including free radicals produced from oxidation of unsaturated fatty acids can accelerate the change from the bright red to a brown colour, denoting the appearance of metmyoglobin (Wassell and Young, 2007). The hump fat had a lower composition of unsaturated fatty acids (UFAs) with 78.73% total UFAs compared to 86.47% and 79.9% total UFAs of the kidney and the brisket fat respectively. Lipid peroxidation depends on the degree of unsaturation of the fatty acids and the levels of the antioxidant vitamin E (tocopherol) and pro-oxidants such as free iron (Moreno and Mitjavila, 2003). Increasing the degree of unsaturation of the fatty acids results in a decrease in colour and oxidative shelf-life (Enser, 2001b). This shows that the hump fat salami was subjected to a reduced oxidation as it had a lower concentration of UFAs and explains why it retained its color compared to the other samples (kidney and brisket fat salami) which were highly concentrated in UFAs and hence highly subjected to oxidation and color changes. Elmore *et al.*, 2000 cooked muscle samples from the lambs fed the megalac, linseed, fish oil and fish oil/linseed diets and found that lipid oxidation products were at much higher concentrations in the more unsaturated samples, especially fish oil. These results were similar to those in beef cattle fed similar diets.

The flavor of the kidney fat salami was different from the other two salami samples and was the lowest ranked salami in flavor with a mean of 6±0.667. This different flavor of the kidney fat salami may probably be associated to the ammonical smell of urine given the fact that the kidney is involved in the ultra filtration of the urine (Fend, 2004). Altering the fatty acid composition of beef muscle can affect its flavor characteristics (Mottram, 1998). The C20 and C22 MUFAs are deposited in the phospholipids of ruminants (Wood and Enser, 1997), which are important sources of lipid derived flavor compounds during cooking (Mottram, 1998). Due to the low oxidative stability of these fatty acids, it seems likely that changes in their concentration, although small, would result in alterations to the composition of the aroma

volatiles produced during cooking. During cooking, chemical reactions occur between fatty acids, amino acids and carbohydrates and their degradation products such as aldehydes and ketones, ammonia and hydrogen sulphide to give a large number of compounds that can contribute to meat flavour (Mottram, 1998). In general, the main sources of volatiles in cooked meat are the thermal degradation of lipid and the Maillard reaction , which occurs between amino acids and sugars (Mottram, 1998). Flavor in meat derives from volatile compounds produced during cooking. Some of these are products of Maillard reactions between sugars and amino acids and some are derived from fatty acid oxidation. Also, the fatty acid oxidation products interact with products from Maillard reactions to produce a further range of odor and flavor compounds (Elmore *et al.*, 1999). Heat-induced oxidation of fatty acids, particularly unsaturated fatty acids, produces degradation products, such as aliphatic aldehydes, ketones, and alcohols, which may have intrinsic flavors. Flavor can largely be affected by the breed of cattle from which the meat is derived. Nitrogen and sulfur-compounds, free amino acids, alcohols, aldehydes and ketones in the flavor volatiles differ among beef from different breeds of cattle (Insausti et al., 2005).

Results also revealed no significant difference between mealiness of the hump and brisket fat salami with (PV =.875>.05). This may probably be associated to the saturated fats containing long-chain fatty acids which solidify easily upon cooling thus influencing the chewiness of salami (Webb and O'Neill, 2008), but these are more likely to be affected by the total amount of fatty acids rather than individual fatty acids. Statistical findings showed a significant difference in mealiness between the hump and kidney fat salami with (PV =.033<.05). This difference was also observed between kidney and brisket fat salami mealiness with (PV =.034<.05). This may also be explained by the effect of fatty acids on tenderness which is said to be due to the different melting points of individual fatty acids, especially stearic and linoleic acids (Wood *et al.*, 2008), thus stearic and linoleic acids contribute to tenderness and chewiness of meat products.

The oral perception of fat has traditionally been considered to rely mainly on texture and olfaction, but recent findings suggest that taste may also play a role in the detection of long chain

fatty acids (Cartoni et al., 2010). Results from this study showed that there was no significant difference between the hump fat salami taste and that of kidney fat salami with (P = 0.458>.05). The same was also seen between brisket fat salami and kidney fat salami taste with (P =.502>.05). There are speculations that fat may appear to increase the sweet, salty and sour tastes and to decrease the bitter taste (because quinine is fat-soluble) by concentrating the taste compounds into the aqueous phase (Metcalf and Vickers, 2001) but the effects fat has on taste are still poorly understood (Dransfield, 2008).

The texture of the brisket was found to be slightly harder and ranked highest compared to the hump and the kidney fat salami with a mean of 7.2±0.632. The hump fat salami was much harder than the brisket fat salami and the kidney fat salami which was very soft and watery. This may be explained by the fact that fatty acids have different melting points caused by differences in the number of double bonds and chain length. Therefore, high concentrations of saturated fatty acids cause fat to be hard at room temperature and affect mouth feel (Wassell and Young, 2007). This statement is also supported by the high percentage of 21.25% of the SFAs found in the hump fat compared to 20.1% SFAs of the brisket fat. The little SFAs composition of 13.52% of the kidney fat explains the softness of the kidney fat salami.

Hump fat salami ranked equally with brisket fat salami in consistency with (M=7.3±0.949) while kidney fat salami ranked lowest with (M= 6.8±1.476). This equal ranking of the hump and the brisket salami may be due to the fact that the amount of phosphate used in all the three samples was the same. In meat products, normally the ratio of sodium salt to phosphate will naturally vary with the nature of the meat product, the age of the animal from which it is produced and the fat content but will ordinarily fall between 2:1 and 8:1 (Hutton, 2002). A ratio of 4:1 will generally perform satisfactorily, with this ratio even meat products containing a high fat content and composed of a variety of meats can be converted into products of uniform fat distribution and of outstanding homogeneity (Hsu *et al.*, 2009). In this study, the ratio of sodium salt to phosphate was 2:1, so the observed difference of the kidney fat salami (M= 6.8±1.476) from the other salami was not clear.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6. 1. Conclusions

Fatty acids profiles showed that hump fat had the highest SFA with (21.25%), it had the second concentration of MUFAs (77.07%) and had lower levels of PUFAs (1.66%) among all samples. Kidney fat had the highest total amount of the MUFAs (83.82%) and the lowest SFAs (13.52%) while its PUFAs concentration (2.65%) was second after the brisket fat. Brisket fat had higher concentration of PUFAs (4.21%), lower concentration of MUFAs (75.69%) among all three samples and had the second concentrations of SFAs with 20.1%.

Although the hump fat salami was ranked the best as per appearance, flavor and taste, no significant differences were noted among products from other fat sources. Kidney fat salami was statistically different from hump fat salami in mealiness at ($P = .033 < .05$), the same as the kidney and brisket fat salami which were significantly different at ($P = .034 < .05$).

Hump fat salami is the best scored product among others (kidney and brisket fat salami) in terms of the studied sensory properties but it was found to have higher saturated fatty acids. Kidney fat is the best fat to be used in the meat industry in terms of fatty acid composition as it has a higher percentage of unsaturated fatty acids which is good for the health of consumers.

6. 2. Recommendations

The meat industry should take into consideration the sources of fats used in processing given their differences in FAs concentration and the effects they have on consumers' nutrition and health. In addition, enough and relevant information on ingredients used in processing should be provided on labels of meat products to guide consumers' choice.

Further studies are recommended to compare sensory properties of salami in which fats are collected from different cattle breeds and the one which fat from only one breed is used. It should be better also for further studies to process a product using a composite fat and compare it with the ones processed using individual fat sources.

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APPENDICES

APPENDIX I: Sensory evaluation form

Name: ……

Product: ……

Date: …….

Time: ……...

The sensory behavior of food products is the ultimate criterion for the acceptability of any product by the consumer. Unless the food products meet the desired standards of taste, flavor, texture, etc., the consumer will not accept the products. In other words, quality of food products to a consumer means the sensory behavior of products.

Test this sample and check appropriate box how much you like or dislike.

Use the appropriate scale to show your attitude by checking at the point that best describes your feelings about the sample. Remember you are the only one, who can tell what you like, an honest expression of your personal feeling will help me.

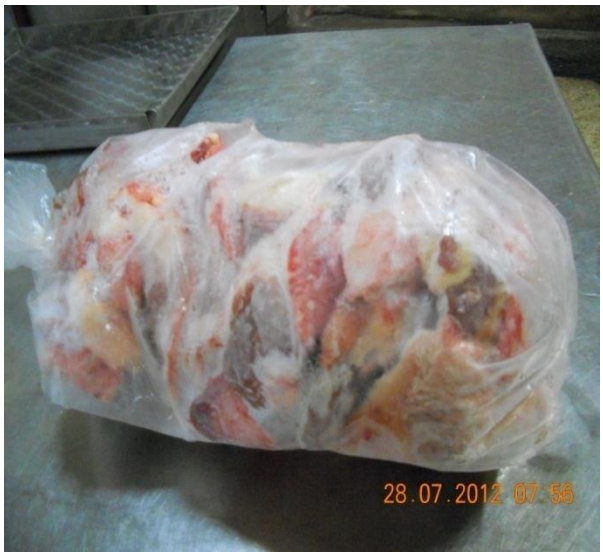
Like extremely	9
Like very much	8
Like moderately	7
Like slightly	6
Neither like nor dislike	5
Dislike slightly	4
Dislike moderately	3
Dislike very much	2
Dislike extremely	1

Attributes	**Sample 1**	**Sample 2**	**Sample 3**
Appearance			
Flavour			
Mealiness			
Taste			
Texture			
Consistency			

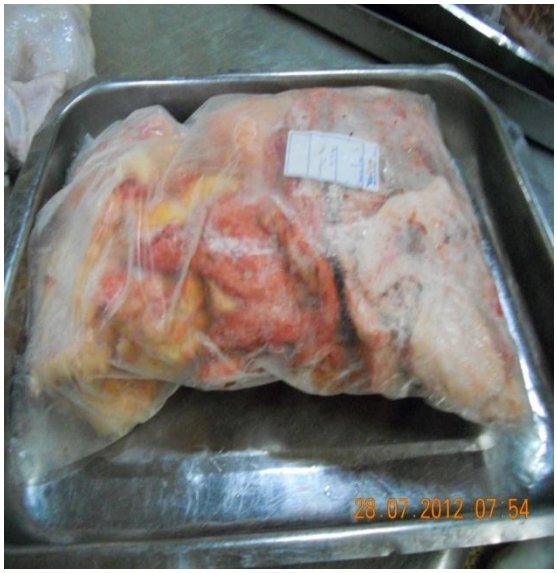
Comments:

Signature: ……

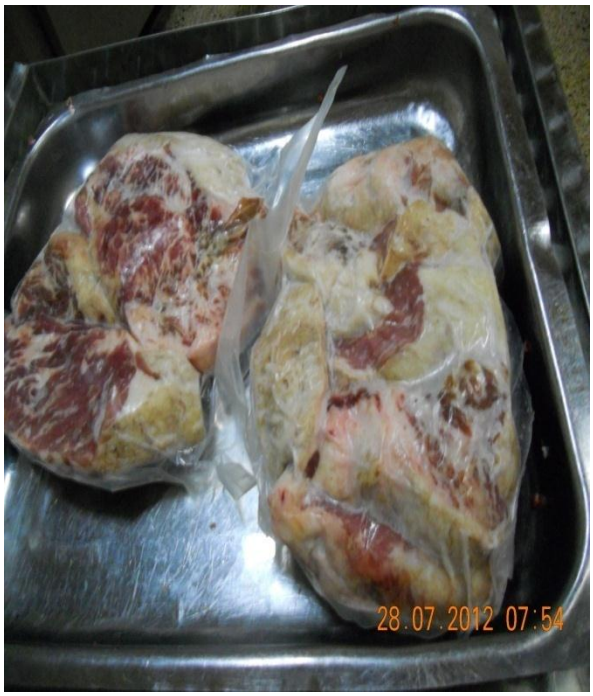
APPENDIX II: Salami processing photos



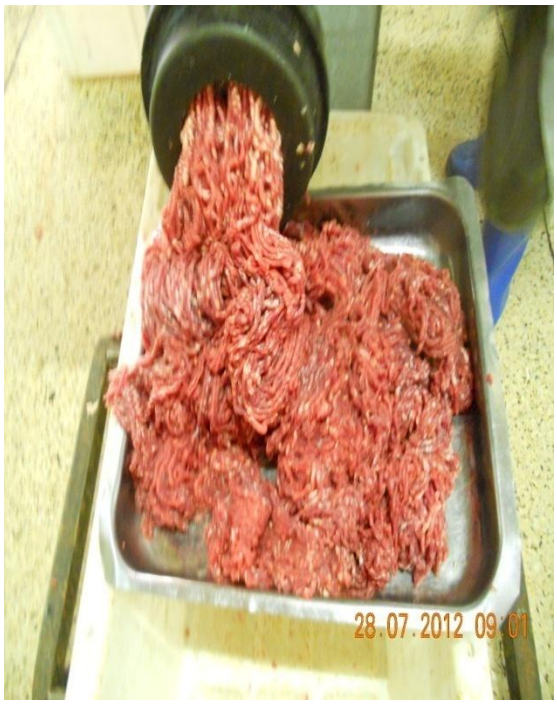
Collected Brisket fat



Collected Kidney fat



Collected fresh hump fat



Meat mincing

Chopping

Filling

Boiling

Slicing

Vacuum packed salami taken to the chiller

Vacuum packed fat sample taken to the lab.

APPENDIX III: Laboratory results

Fatty Acid profile calculations (Hump fat)

FAMEs	fatty acid	replicate 1	replicate 2	mean (%)
butiric acid	C4	0	0	0,00
caproic acid	C6	0	0	0,00
caprylic acid	C8	0	0	0,00
capric acid	C10	0	0	0,00
undecanoic acid	C11	0	0	0,00
lauric acid	C12	0	0	0,00
tridecanoic acid	C13	0	0	0,00
myristic acid	C14	1,7342	1,7342	1,78
myristoleic acid	C14:1	0	0	0,00
pentadecanoic acid	C15	0	0	0,00
cis-10-pentadecenoic aicd	C15:1	0	0	0,00
palmitic acid	C16	9,0884	9,0884	9,35
palmitoleic acid	C16:1	0	0	0,00
heptadecanoic acid	C17	0	0	0,00
cis-10-heptadecanoic acid	C17:1	0	0	0,00
steraic acid	C18	3,8408	3,8408	3,95
oleic acid	C18:1w9cis	32,0866	32,0866	33,02
elaidic acid	C18:1w9trans	0	0	0,00
linoleic acid	C18:2cis	0,4683	0,4683	0,48
linolelaidic acid	C18:2trans	0	0	0,00
gamma-linolenic acid	C18:3w6	0	0	0,00
alfa-linolenic acid (ALA)	C18:3w3	1,1488	1,1488	1,18
arachidic acid	C20	1,8874	1,8874	1,94
cis-11-eicosenoic acid	C20:1	24,4288	24,4288	25,14
cis-11,14-eicosadienoic acid	C20:2	0	0	0,00
cis-8,11,14-eicosatrienoic ac	C20:3w6	0	0	0,00
heneicosanoic acid	C21	0	0	0,00
cis-11,14,17-eicosatrienoic	C20:3w3	0	0	0,00
arachidonic acid	C20:4w6	0	0	0,00
eicosapentaenic acid (EPA)	C20:5w3	0	0	0,00
behenic acid	C22	3,1849	3,1849	3,28
erucic acid	C22:1w9	18,3779	18,3779	18,91
cis-13,16-docosadienoic acid	C22:2	0	0	0,00
tricosanoic acid	C23	0	0	0,00
lignoceric acid	C24	0,9205	0,9205	0,95
docosahexaenic acid (DHA)	C22:6w3	0	0	0,00
nervonic acid	C24:1	0	0	0,00
	sum	**97,1666**	**97,1666**	**100,00**

Fatty Acid profile calculations (Kidney fat)

Fatty acids	fatty acid	replicate 1	replicate 2	mean (%)
butiric acid	C4	0	0	0,00
caproic acid	C6	0	0	0,00
caprylic acid	C8	0	0	0,00
capric acid	C10	0	0	0,00
undecanoic acid	C11	0	0	0,00
lauric acid	C12	0	0	0,00
tridecanoic acid	C13	0	0	0,00
myristic acid	C14	0,4389	0,4389	0,45
myristoleic acid	C14:1	0	0	0,00
pentadecanoic acid	C15	0	0	0,00
cis-10-pentadecenoic aicd	C15:1	0	0	0,00
palmitic acid	C16	4,4004	4,4004	4,49
palmitoleic acid	C16:1	0	0	0,00
heptadecanoic acid	C17	0	0	0,00
cis-10-heptadecanoic acid	C17:1	0	0	0,00
steraic acid	C18	1,5003	1,5003	1,53
oleic acid	C18:1w9cis	30,4963	30,4963	31,12
elaidic acid	C18:1w9trans	0	0	0,00
linoleic acid	C18:2cis	0,6059	0,6059	0,62
linolelaidic acid	C18:2trans	0	0	0,00
gamma-linolenic acid	C18:3w6	0	0	0,00
alfa-linolenic acid (ALA)	C18:3w3	1,9902	1,9902	2,03
arachidic acid	C20	0,4754	0,4754	0,49
cis-11-eicosenoic acid	C20:1	18,4651	18,4651	18,84
cis-11,14-eicosadienoic acid	C20:2	0	0	0,00
cis-8,11,14-eicosatrienoic ac	C20:3w6	0	0	0,00
heneicosanoic acid	C21	0	0	0,00
cis-11,14,17-eicosatrienoic	C20:3w3	0	0	0,00
arachidonic acid	C20:4w6	0	0	0,00
eicosapentaenic acid (EPA)	C20:5w3	0	0	0,00
behenic acid	C22	5,618	5,618	5,73
erucic acid	C22:1w9	33,1772	33,1772	33,86
cis-13,16-docosadienoic acid	C22:2	0	0	0,00
tricosanoic acid	C23	0	0	0,00
lignoceric acid	C24	0,8176	0,8176	0,83
docosahexaenic acid (DHA)	C22:6w3	0	0	0,00
nervonic acid	C24:1	0	0	0,00
	sum	**97,9853**	**97,9853**	**100,00**

Fatty Acid profile calculations (Brisket fat)

Fatty acid	fatty acid	replicate 1	replicate 2	mean (%)
butiric acid	C4	0	0	0,00
caproic acid	C6	0	0	0,00
caprylic acid	C8	0	0	0,00
capric acid	C10	0	0	0,00
undecanoic acid	C11	0	0	0,00
lauric acid	C12	0	0	0,00
tridecanoic acid	C13	0	0	0,00
myristic acid	C14	3,37	3,37	3,69
myristoleic acid	C14:1	0	0	0,00
pentadecanoic acid	C15	0	0	0,00
cis-10-pentadecenoic aicd	C15:1	0	0	0,00
palmitic acid	C16	3,24	3,24	3,54
palmitoleic acid	C16:1	0	0	0,00
heptadecanoic acid	C17	0	0	0,00
cis-10-heptadecanoic acid	C17:1	0	0	0,00
steraic acid	C18	4,95	4,95	5,41
oleic acid	C18:1w9cis	21,79	21,79	23,84
elaidic acid	C18:1w9trans	0	0	0,00
linoleic acid	C18:2cis	1,95	1,95	2,13
linolelaidic acid	C18:2trans	0	0	0,00
gamma-linolenic acid	C18:3w6	0	0	0,00
alfa-linolenic acid (ALA)	C18:3w3	1,9	1,9	2,08
arachidic acid	C20	0,98	0,98	1,07
cis-11-eicosenoic acid	C20:1	31,34	31,34	34,28
cis-11,14-eicosadienoic acid	C20:2	0	0	0,00
cis-8,11,14-eicosatrienoic ac	C20:3w6	0	0	0,00
heneicosanoic acid	C21	0	0	0,00
cis-11,14,17-eicosatrienoic	C20:3w3	0	0	0,00
arachidonic acid	C20:4w6	0	0	0,00
eicosapentaenic acid (EPA)	C20:5w3	0	0	0,00
behenic acid	C22	3,43	3,43	3,75
erucic acid	C22:1w9	16,06	16,06	17,57
cis-13,16-docosadienoic acid	C22:2	0	0	0,00
tricosanoic acid	C23	0	0	0,00
lignoceric acid	C24	2,41	2,41	2,64
docosahexaenic acid (DHA)	C22:6w3	0	0	0,00
nervonic acid	C24:1	0	0	0,00
	sum	**91,42**	**91,42**	**100,00**

APPENDIX IV: significant difference among SFAs, MUFAs and PUFAs of the different fat sources

Table 6: Statistical comparison of the saturated fatty acids in the hump, kidney and brisket fat

	(I) Samples	(J) Samples	Std. Error	Sig.
Tukey HSD	Hump fat	Kidney fat	1.62762	.036
		Brisket fat	1.62762	.777
	Kidney fat	Hump fat	1.62762	.036
		Brisket fat	1.62762	.054
	Brisket fat	Hump fat	1.62762	.777
		Kidney fat	1.62762	.054

Dependent Variable: PUFA. The mean difference is significant at the .05 level

Table 7: Statistical comparison of the monounsaturated fatty acids in the hump, kidney and brisket fat

	(I) Samples	(J) Samples	Std. Error	Sig.
Tukey HSD	Hump fat	Kidney fat	.81142	.007
		Brisket fat	.81142	.339
	Kidney fat	Hump fat	.81142	.007
		Brisket fat	.81142	.004
	Brisket fat	Hump fat	.81142	.339
		Kidney fat	.81142	.004

Dependent Variable: PUFA. The mean difference is significant at the .05 level

Table 8: Statistical comparison of the polyunsaturated fatty acids in the hump, kidney and brisket fat

	(I) Samples	(J) Samples	Std. Error	Sig.
Tukey HSD	Hump fat	Kidney fat	.12000	.008
		Brisket fat	.12000	.000
	Kidney fat	Hump fat	.12000	.008
		Brisket fat	.12000	.002
	Brisket fat	Hump fat	.12000	.000
		Kidney fat	.12000	.002

Dependent Variable: PUFA The mean difference is significant at the .05 level