OXIDATIVE STABILITY OF SELECTED VEGETABLE OILS AFTER DEEP FRYING IN DIFFERENT TYPES OF FOODS IN KENYA

Chebet Josphine

A Thesis Submitted to the Graduate School in Partial Fulfillment for the Requirements of the Award of the Master of Science Degree in Chemistry of Egerton University

EGERTON UNIVERSITY

APRIL, 2017

DECLARATION AND RECOMMENDATION

DECLARATION

This thesis is my original work and has not been submitted or presented for examination in any other institution of learning to the best of my knowledge.

Chebet Josphine
SM/11/2629/10
Signature …… Date ……...

RECOMMENDATION

This thesis has been submitted for examination with our approval as university supervisors.

Dr. Kinyanjui T.

Egerton University

Signature …… Date ……...

Prof. Cheplogoi P. K.
Egerton University

Signature …… Date ……...

DEDICATION

To my family, especially my husband for his encouragement, moral and financial support during my research.

COPYRIGHT

ACKNOWLEDGEMENT

I wish to express deep appreciation to my supervisors Dr. T. Kinyanjui and Prof. P.K. Cheplogoi who without them, this study would not have been a success. I appreciate them for their commitment, guidance and support throughout this study. I wish to thank the technical staff in the Department of Chemistry especially Mr. G.K Kibet for his support. I wish to thank my colleagues especially Ms C. Mudalungu for her encouragement. G.C analysis was done at the Tea Research Foundation of Kenya and I wish to appreciate the management especially Dr. J. Wanyoko for allowing me to use their facilities. Special thanks go to Mr. K Mwangi of Tea Research Foundation of Kenya for his assistance in the G.C analysis. I would like to appreciate the Rift Valley Institute of Science and Technology for allowing me to use their food science laboratory during frying.

ABSTRACT

Vegetable oils are triglycerides extracted from plants. Deep frying is one method which involves submerging the food in hot oil. When this oil is overused it undergoes a series of chemical reactions which may affect human health. This has made it important to study the oxidative stability of selected vegetable oils as relates to the different types of food fried. The main objective was to determine the relative thermal oxidative stability of the palm, corn, peanut, soybean, and sunflower oils after deep frying in different types of foods. In this study five types of foods were deep fried in five types of oils for 6 hrs. The oils were then divided into two equal portions. One portion was refrigerated at 4^0C and the other at room temperature for 5 days then used for frying for another 6 hrs. It was found that the storage conditions had significant effects on the oils. Fatty acid composition was done using gas chromatography where it was found that myristic, palmitic, linoleic and linolenic acid varied significantly ($p \leq 0.05$) per the type of oils. Soybean oil was found to have relatively higher linoleic (1025.59 ug/ml) and linolenic (43.90 ug/ml). Linoleic and linolenic acids were least concentrated in Palm 157.5 ug/ml and 0.7986 ug/ml. Peroxide values increased in all the oils after frying food. The iodine values of oils before and after frying food were compared and out of this, it was found that there was decrease in iodine value in all the oils after frying food. The highest decrease in Iodine value was observed in soybean after frying the five types of food and the values in g of I_2/100g of oil were as follows chicken 27.0, chips 22.0, fish 35.5, mandazi 38.1 and smokies 17.3. The least decrease was indicated by palm: chicken 5.1, chips 4.6, fish 7.6, mandazi 2.0 and smokies 6.6. The study concluded that in regard to oil suitability, the parameters that were of major interest were peroxide, *para*-anisidine, iodine, refractive index and density values. Soybean oil proved to be relatively unstable; while palm and peanut were more stable compared to the other oils. In terms of food, the oil in which chicken and fish were fried contained more degradation products.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF APPENDICES

ABBREVIATIONS AND ACRONYMS

ANOVA	Analysis of Variance
AOAC	Association of Official Analytical Chemists
AOCS	America Oil Chemists Society
A.V	Acid Value
ASTM	America Standard Test method
FAME	Fatty acid methyl esters
FFA	Free fatty acids
GC	Gas chromatography
HDL	High Density lipoprotein
I.S	Internal standard
I.V	Iodine Value
LDL	Low Density lipoprotein
MUFAS	Monounsaturated fatty acids
P-AV	P-anisidine value
PUFAS	Polyunsaturated fatty acids
PV	Peroxide value
R.I	Refractive index
SFA	Saturated fatty acid
SV	Saponification Value
TPC	Total polar compounds

CHAPTER ONE
INTRODUCTION

1.1 Background information

A vegetable oil is any oil that comes from plants. The term "vegetable oil" can be narrowly defined as referring only to substances that are liquid at room temperature (Saroj, 1999) or broadly defined without regard to a substance's state of matter at a given temperature (Robin, 1999). There are different types of vegetable oils; edible and non-edible. Edible vegetable oils are used in cooking and as supplements. These include coconut, groundnut, sesame, sunflower, soybean, safflower, peanut, olive, palm, rapeseed, canola and many others. The common non-edible oils include Jatropha and castor (Ulbert and Roubicek, 1993). Oxidative stability is a measure of an oil or fat's resistance to oxidation. It is an important parameter for the quality assessment of fats and oils (Laubli and Bruttel, 1986).

Over the years, vegetable oils have been used in various methods of cooking. They are used in deep-frying, baking and food processing. Deep frying involves submerging the food in hot, liquid fat at a high temperature of 150 - 190^0C (Yamsaengsung and Moreira, 2002). It is primarily a dehydration process, which means that water and water-soluble substances are extracted from the product being deep fried and transferred to the cooking fat (Choe and Min, 2007). In this method of cooking; water, oxygen and heat are the main factors, which determine the kinetics of oxidation and polymerization processes (Gertz *et al.,* 2001).

As deep fat frying is normally carried out at high temperatures (between 150^0C and 190^0C) and in the presence of air and moisture, these frying oils and fats will undergo physical and chemical deterioration which will affect their frying performance and the storage stability of the fried products (Fauziah *et al*., 2000). During deep-frying, the fat and oil decompose forming volatile, non-volatile, monomeric and polymeric, oxidised or non-oxidised compounds (Gertz and Kochhar, 2001). These products result from oxidation of unsaturated fatty acids, lipid hydrolysis, and transformation of linear fatty acids in cyclical compounds and fatty acid or lipid polymerization (Chang *et al*., 1978). The intensity of these reactions depends on duration, method of heat treatment, frying medium and type of product (Blumenthal., 1991; Stevenson *et al.,* 1984).

These compounds are of concern because they accumulate in the frying oil, promote further degradation, absorbed by the fried food, enter the diet and affect the public human health (Stevenson *et al.*, 1984; Sebedio *et al.*, 1990; Romero *et al.*, 1998; Che man *et al.*, 2003).

Their amounts and chemical structures depend on the nature of fat or oil used, the temperature, frying time, the food being fried, and on the accessibility of air (oxygen) (Gertz and Kochhar, 2001).

Foods commonly prepared by deep frying in Kenya include fish, chicken, French fries, mandazi and sausages. After deep frying, many people are tempted to keep the oil to be reused for long periods of time. This causes adverse effects on flavour, stability, colour and texture of fried product and may be harmful to human health (Sharoba and Ramadan, 2012). Many people are not aware of the adverse effects that oxidized lipids could cause on the human body. This study was done to comparatively establish the oxidative stabilities of five types of vegetable oil commonly used in Kenya after deep frying chicken, chips, smokies, mandazi and fishes. The vegetable oils that were compared are Palm, sunflower, corn, soybean and peanut oils.

1.2 Statement of the problem

Deep frying is the most commonly used method of cooking foods like fish, chicken, sausages, chips and many more. After deep frying lots of oil remain and many people are tempted to keep the oil for reuse so that they can cut down the costs. When this oil is overused it undergoes a series of oxidative chemical reactions forming toxic compounds. These compounds are absorbed by the fried foods, eventually they gets to the human body. This can cause adverse effects on the body. Currently, there have been many cases of heart diseases and one of the possible causes may be overusing of vegetable fats and oils.

1.3 Hypotheses

1. There is no significant difference in the fatty acid composition between vegetable oils commercially available in Kenya.
2. There is no significant difference in the oxidative stability of the various vegetable oils before and after frying food.
3. There is no significant difference on the effects of frying different types of food and storage conditions on oxidative stability of vegetable oils.

1.4 Main objective

To determine the relative oxidative stability of selected vegetable oils after deep frying in different types of food.

1.5 Specific objectives

1. To chemically characterize vegetable oils commercially available in Kenya
2. To compare the oxidative stability of various vegetable oils.
3. To determine the effect of frying different types of food and storage conditions on oxidative stability of the oils.

1.6 Justification

Deep frying is the most commonly used method of cooking in Kenya. After deep frying many people keep the oil for reuse. When cooking oil is kept for sometimes after use, it undergoes a number of chemical reactions including oxidative degradation which may result in the formation of toxic compounds and free radicals. These compounds may be absorbed from food causing adverse effects on human body. There are increased cases of coronary heart diseases and cancer which may be caused by cooking oil of questionable quality. The oxidative stability, the compounds formed by oils after deep frying food and the health effects they can pose need to be established because very many people are not aware of their dangers.

CHAPTER TWO
LITERATURE REVIEW

2.1 Introduction

This study aims at determining the oxidative stability of selected vegetable oils after deep frying various foods in Kenya. Currently, no research has been done on this and therefore it will help in pointing out the relative stability of five types of oils and suitable storage conditions for frying oils after use.

2.2 Vegetable oils

Vegetable oils are substances which are obtained from oil containing seeds, fruits, or nuts by different pressing methods, solvent extraction or a combination of these (Bennion, 1995). There are numerous vegetable oils derived from various sources such as soybean, cottonseed, peanuts, sunflower, palm, palm kernel, coconut seed, castor seed and rapeseed oil. Also, they are defined as triglycerides formed by the reaction of one glycerol molecule and three fatty acids to yield one molecule of triglyceride and three molecules of water (Patterson, 1989). Vegetable oil production has increased over the years with palm recording the highest growth compared to other oils as shown in figure 2.1.

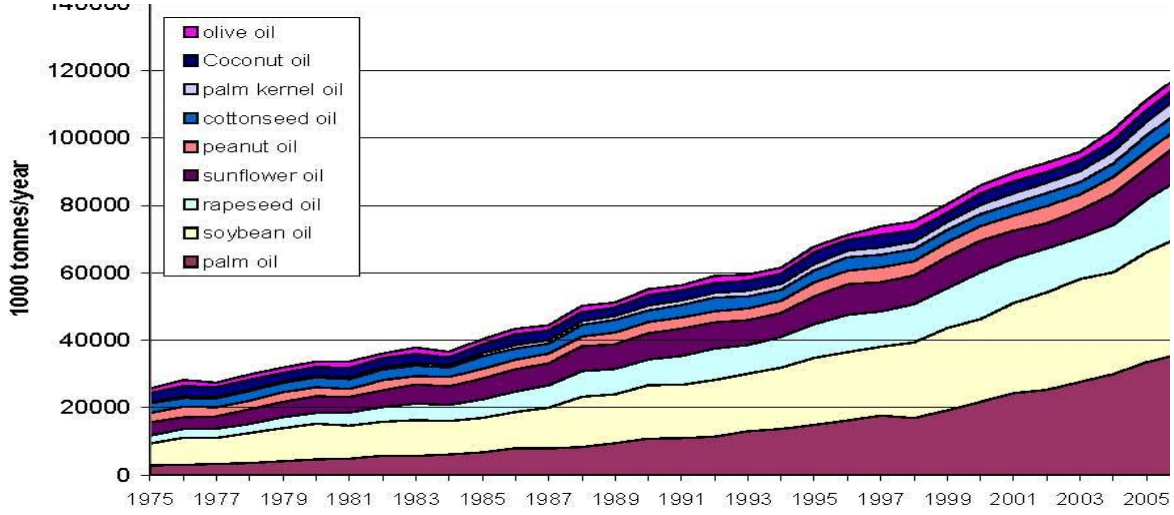


Figure 2.1: World's vegetables oil production, 1975- 2007(Source of data: www.fas.usda.gov/psdonline)

Many vegetable oils are used to make soaps, skin products, candles, perfumes and other personal care and cosmetic products. Some of these are particularly suitable as drying oils and are used in making paints and wood treatment.

2.3 Fatty acids

Vegetable oils differ in their composition of fatty acids. Fatty acids are classified into saturated and unsaturated fatty acids. Unsaturated fatty acids are either monounsaturated (one double bond) or polyunsaturated (two or more double bonds). The levels of five major fatty acids in some vegetable oils are shown in Table 1.

Table 2.1: Fatty acid composition (%) of some vegetable oils (Firestone and Reina, 1996)

Fatty acid	Palm oil	Palm olein	Olive oil	Rapeseed oil	sunflower oil	Corn oil
Palmitic (C16.0)	40-46	38-43	7.5-20	3.3-6.0	2.7-4.2	10-17
Stearic (C18.0)	4-7	3.7-4.8	0.5-5.0	1.0-2.5	3-5	1.6-3.3
Oleic (C18.1)	36-41	40-44	55-83	52-67	80-87	25-42
Linoleic(C18.2)	9-12	10-13	3.5-21	16-25	4-9	39-61
Linolenic(C18.3)	0.1-0.4	0.1-0.6	0-1.5	6.0-14	-	0.7-1.3

2.4 Chemical changes in frying oil

During deep-fat frying, the fat is continuously or repeatedly exposed to temperatures of between 150-180°C in the presence of the substrate, air and water. Under these conditions, a complex series of reactions takes place; namely hydrolysis, oxidation, polymerization, isomerization and cyclization (Razali and Badri, 2003). As a result, oils are degraded from thermal oxidation to form volatile and non-volatile decomposition products (Melton *et al*., 1994). Food fried in fat make significant contributions to the calories in the average diet. In the course of deep frying, food contacts oil at about 180^{o}C and is partially exposed to air for various periods of time. Thus frying, more than any other standard method food process or handling method, has the greatest potential for causing chemical changes in fat, and sizeable amounts of this fat are carried with the food (5-40% fat by weight is absorbed) (Nawar, 1996).

2.5 Quality of fried foods

Deep-fat frying is the cooking of food in pre-heated deep oil/fat at a high temperature of 150 to 180°C (Choe and Min, 2007). It is a popular method for food preparation, in which vegetable oils are used as a heat-exchange medium and contribute to the quality of fried

products (Shahidi and Wanasundara, 2002). The chemical changes in frying oil result in changes in the quality of fried food. Since the fatty acid composition of the frying oil is an important factor affecting fried food flavor and its stability, it should have low level of polyunsaturated fatty acid such as linoleic or linolenic acids and high level of oleic acid with moderate amounts of saturated fatty acid (Mehta and Swinburn, 2001; Kiatsrichart *et al*., 2003). Soyabean oil has a good nutritional profile due to high level of unsaturated fatty acid but less oxidative stability (Steenson and Min, 2000). The fat and oil usually used in the frying processes are made of fatty acids which are either saturated or unsaturated. Most naturally occurring fatty acids have a chain of an even number of carbon atoms, from 4 to 28 (Alireza *et al.*, 2010). Fried foods have desirable flavor, color, and crispy texture, which make deep-fat fried foods very popular to consumers (Boskou *et al*., 2006). However, the changes taking place in oil due to repeated frying are often deteriorative and fatty acids undergo chemical changes and make the food that is fried an unsuitable product in terms of nutritional value (Sebedio *et al*., 1996). The over-use of deep-frying oil causes adverse effects on flavour, stability, colour and texture of fried product and may be harmful to human health. It is, therefore, necessary to examine some of the major changes which occur in the oils during deep frying (Sharoba and Ramadan, 2012).

2.6 Oxidation at high temperature

The chemistry of lipid oxidation at the high temperatures of food processes like baking and frying is highly complex since both oxidative and thermal reactions are involved simultaneously. As the temperature increases, the solubility of oxygen decreases drastically, although all the oxidation reactions are accelerated (Frankel, 2005; Velasco *et al.*, 2008). Resistance to oxidation during prolonged exposure to high temperature is one of the main properties that industrial frying oil should possess (Kochhar, 2001).

Figure 2.2 shows the well-known scheme of the oxidation process which proceeds *via* a free radical mechanism of chain reactions, where RH represents the triacylglycerol molecule undergoing oxidation in one of its unsaturated fatty acyl groups. In the initiation stage, an alkyl radical (˙R) is formed by ,abstraction of a hydrogen radical from an allylic or bisallylic position of an unsaturated fatty acid. In the propagation step, the alkyl radical reacts with oxygen at rates controlled by diffusion to form peroxyl radicals that in turn react with new triacylglycerol molecules giving rise to hydro peroxides as the primary oxidation products

and new alkyl radicals that propagate the reaction chain. Finally, in the termination stage, radicals react between them to yield relatively stable non-radical species.

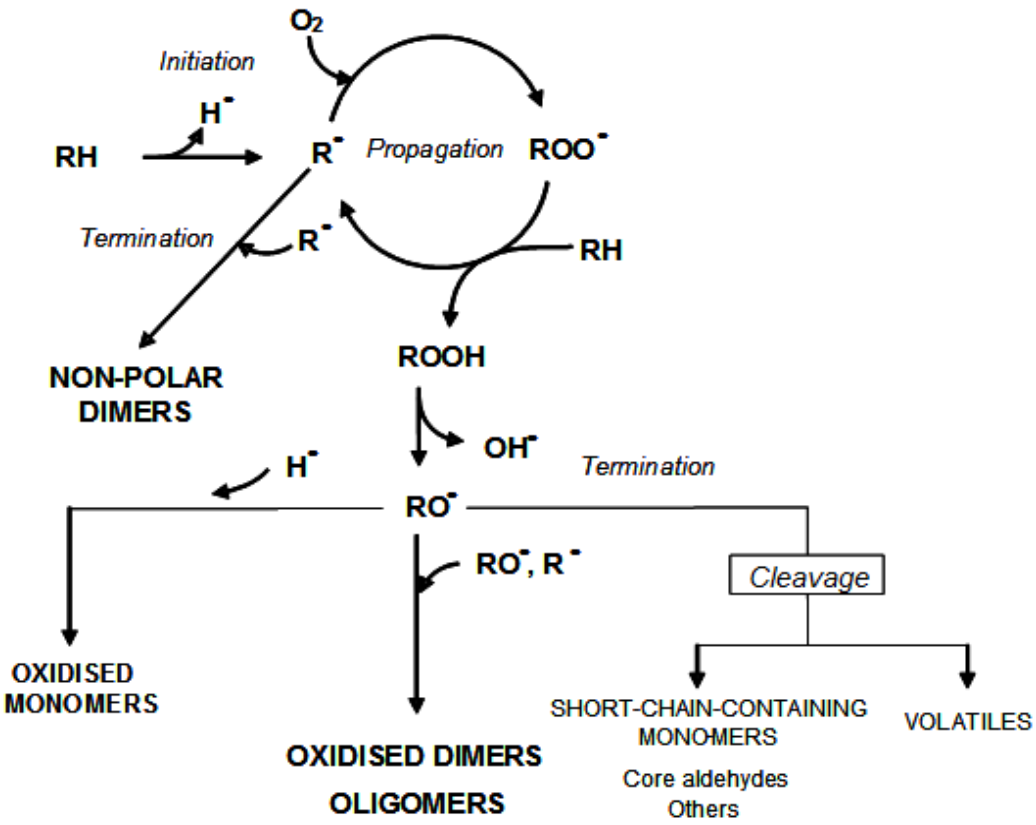


Figure 2.2: Simplified scheme of thermal oxidation (Dobarganes and Marquez-Ruiz, 2006).

At frying temperature, as the oxygen pressure is reduced, the initiation reaction becomes more important and the concentration of alkyl radicals ($R^{\bullet}$) increases with respect to alkyl peroxyl radicals ($ROO^{\bullet}$). As a result, polymeric compounds are mainly formed through reactions mainly involving alkyl ($R^{\bullet}$) and alkoxyl ($RO^{\bullet}$) radicals.

At low or moderate temperatures, formation of oxidation compounds during the induction period is slow; hydro peroxides (ROOH) are the major compounds formed and their concentration increases until advanced stages of oxidation. Polymerization compounds only become significant in the accelerated stage of oxidation after the end of the induction period (Dobarganes and Márquez-Ruiz, 2006). At high temperatures, formation of new compounds is very rapid, ROOH are practically absent above 150ºC, indicating that the rate of ROOH decomposition becomes higher than that of their formation, and polymeric compounds are formed from the very early stages of heating. Also, the formation of significant amounts of

non-polar triacylglycerol dimers (R-R), typical compounds formed in the absence of oxygen through interaction of alkyl radicals, is a clear indication of the low oxygen concentration (Dobarganes and Pérez-Camino, 1987).

2.7 Classes of compounds produced from the oil during frying.

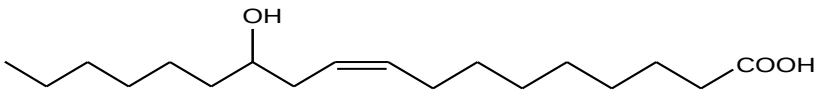
According to Nawar (1996), there are four classes of compounds that are produced from the oil during frying. These compounds are volatiles, non-polymeric polar compounds of moderate volarity, dimeric and polymeric acids and glycerides; and free fatty acids. They are responsible for a variety of physical and chemical changes in viscosity and free fatty acid content, development of a dark colour, decreases in iodine value and surface tension, changes in refractive index, and an increased tendency to foam.

2.7.1 Volatiles

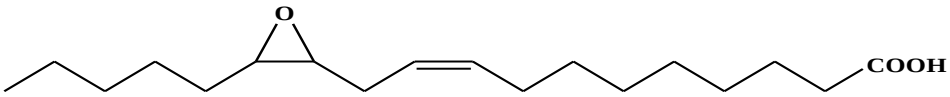
During deep-fat frying, a serial of complex reactions such as oxidation, hydrolysis, isomerization, and polymerization take place during the deep-fat frying course and influence quality attributes of the final product such as flavor, texture, shelf life and nutrient composition. The influence of these reactions results from a number of their products including volatile compounds, hydrolysis products, oxidized triacylglycerol monomers, cyclic compounds, *trans* configuration compounds, polymers, sterol derivatives, nitrogen- and sulphur-containing heterocyclic compounds and acrylamide which are present in both frying oil and the fried food (Zhang *et al.*, 2012).

2.7.2 Non polymeric polarity compounds of moderate volarity

These compounds are produced according to the various oxidative pathways involving the alkoxy radical. The examples include hydroxyl and epoxy acids.



12-Hydroxy-octadec-9-enoic(ricinoleic) acid



Vernolic acid

2.7.3 Dimeric and polymeric acids and glycerides

These compounds occur, as, expected, from thermal and oxidative combinations of free radicals. As frying oil deteriorates, non-volatile (reactive) compounds are also formed. These molecules remain in the frying oil and begin "polymerizing" or bonding together at high oil temperatures to form clusters that accumulate on the oil's surface. These particles are large enough to cause foaming and increase the possibility of hydrolysis (Heness, 2006).

Table 2.2: Summary of compounds produced during frying

Alteration	Causative agent	Compounds
Hydrolysis	Moisture	Fatty acids Diacylglycerols
Oxidation	Air	Oxidized monomerictriacylglycerols Oxidized dimeric and oligomerictriacylglycerol Volatile compounds (aldehydes, ketones, alcohols, hydrocarbons.)
Thermal alteration	Temperature	Cyclic monomeric triacylglycerols Isomeric monomeric triacylglycerols Nonpolar dimeric and oligomerictriacylglycerols

Source: (Dobarganes and Marquez -Riuz, 2006)

2.8 Animal oils versus vegetable oils.

There has been growing controversy about which oil is healthier for consumption. Animal oils are primarily made up of saturated fats (or saturated triglycerides*)* whereas vegetable oils are made up of unsaturated fats (polyunsaturated or monounsaturated). The growing amount of evidence today suggests that animal oils are unhealthy if taken in regularly. They are thought to cause various cardiovascular diseases and may lead to an increased risk of heart attacks or strokes and in extreme cases, even cancer. This is because regular consumption of saturated fats leads to higher levels of low-density lipoproteins, also known as bad cholesterol in the blood system. This increased level of LDLs can clog arteries and cause serious, adverse effects on the cardiovascular system (http://vegetableoils.org/vegetableoil/).

However, some vegetable oils can cause even more harm to your health than animal oils. These are the hydrogenated vegetable oils. They increase LDLs associated with cardiovascular and cerebrovascular diseases. They also decrease HDLs (Stender and Dyerberg, 2003). Oils with a high content of saturated fatty acids are more stable in the frying process but because of the negative health attributions associated with these, the interest in using monounsaturated oils in frying has increased (McDonald and Eskin, 2006).

According to joint Food and Agriculture Organization of the United Nations/World Health Organization (FAO/WHO) report 2008/ 2009, there is convincing evidence that replacing dietary saturated fats with polyunsaturated fats (PUFA) decreases risk of cardiovascular diseases. Therefore, PUFA rich foods such as vegetable oils, fatty fish, and marine omega-3 supplements are recommended. However, PUFA are easily oxidizable when heated and there is concern about possible negative health effects from intake of oxidized lipids. Little is known about the degree of lipid oxidation in such products (Halvorsen and Blomhoff, 2011).

A high consumption of omega-6-polyunsaturated fatty acids (PUFAs), which are found in most types of vegetable oil, may increase the likelihood that postmenopausal women will develop breast cancer (Sonestedt *et al.*, 2008). A similar effect was observed on prostate cancer in mice (Berquin *et al.*, 2007).

2.9 Dangers of overusing cooking oil

Although re-using cooking oil is a somewhat common practice, it can pose some serious health hazards (http://goaskalice.columbia.edu/reusing-cooking-oil-safe). This is because food absorbs varying amounts of oil during deep frying (potato chips have a final fat content

of 35%), resulting in the need for frequent or continuous addition of fresh oil. The food itself can release some endogenous lipids into the frying oil and consequently the oxidative stability of the new mixture may be different from that of the original frying oil. The presence of food causes the oil to darken at accelerated rate (Nawar, 1996). The most common danger when recycling cooking oil is that it becomes rancid or spoiled.

In addition to having off flavors and odors, rancid oil may contain possibly carcinogenic free radicals (http://goaskalice.columbia.edu/reusing-cooking-oil-safe).

2.10 Rancid oils

According to WHO (2009), overheating or over-using the frying oil leads to formation of rancid-tasting products of oxidation, polymerization, and other deleterious, unintended or even toxic compounds such as acrylamide (from starchy foods). Deep frying under vacuum helps to significantly reduce acrylamide formation (Granda *et al.*, 2004). However, this process is not widely used in the food industry due to the high investment cost involved.

Rancid oils may produce damaging chemicals and substances that may not make you immediately ill, but can cause harm over time. Chemicals such as peroxides and aldehydes can damage cells and contribute to atherosclerosis. Free radicals produced by rancid oils can also damage DNA in cells (Nummer, 2011).

2.11 Types of rancidity

There are two basic types of rancidity: hydrolytic rancidity, which occurs when water breaks larger compounds into smaller ones; and oxidative rancidity, in which the double bond of an unsaturated fatty acid reacts chemically with oxygen to result in two or more shorter molecules (Brown, 2011).

2.11.1 Hydrolytic rancidity

Fats become rancid through the addition of water because water hydrolyzes the bonds in the triglyceride, causing it to break down into smaller compounds. This hydrolytic rancidity has implications for deep-fat frying, because placing cold, wet food in heated frying oil introduces water and makes the oil prone to hydrolytic rancidity (Brown, 2011).

2.11.2 Oxidative rancidity

The heat treatment causes the oxidative rancidity resulting in an increase in the free fatty acids. This is why heated and unheated fats and oils should be monitored by means of

analysis studies (Orthoefer and Cooper, 1996; Tyagi and Vasishtha, 1996; Choe and Min, 2007).

2.12 Analysis of vegetable oils

Quality evaluation of frying fat may be carried out in different ways. Physical methods estimate oxidative degradation by monitoring changes in physical properties of frying fats, such as molecular weight, specific gravity, smoke point, refractive index, chromatic parameter, viscosity, surface tension, and dielectric constant (Perkins, 1992). Chemical methods include the iodine value, saponification value, free fatty acid content, peroxide value, or *p*-anisidine value, among others (Shahidi and Zhong, 2005).

2.12.1 Peroxide value

The peroxide value is a measure of the concentration of peroxides and hydro peroxides formed in the initial stages of lipid oxidation. Lipid oxidation involves the continuous formation of hydro peroxides as primary oxidation products that may break down to a variety of non-volatile and volatile products (Dobarganes and Velasco, 2002). The peroxide value (PV) is an indicator of the initial stages of oxidative change (Riuz et al., 2001). The free radical mechanism of lipid oxidation is usually described as three stages of initation, propagation, and the termination steps (Berezin and Denisov, 1996)

Initiation

$$RH \longrightarrow R\bullet + H\bullet \quad \text{..... (i)}$$

Initiation starts with the abstraction of a hydrogen atom adjacent to a double bond in a fatty acid (RH) molecule, and this may be catalyzed by light, heat, or metal ions to form a free radical.

Propagation

$$R\bullet + O_2 \longrightarrow ROO\bullet \quad \text{..... (ii)}$$

$$ROO\bullet + RH \longrightarrow ROOH + R\bullet \quad \text{..... (iii)}$$

The resultant free radical (R•) reacts with atmospheric oxygen to form an unstable peroxy free radical (ROO •), which may in turn abstract a hydrogen atom from another unsaturated fatty acid to form a hydro peroxide (ROOH) and a new alkyl free radical initiates further oxidation and contributes to the chain reaction.

Termination

$R\bullet + R\bullet \longrightarrow R - R$ ……..…… (iv)

$R\bullet + ROO\bullet \longrightarrow ROOR$ …….……(v)

$ROO\bullet + ROO\bullet \longrightarrow ROOR + O_2$ … …….……(vi)

In termination stage of the autoxidation, free radicals interact or react with each other and turn to normal state cause formation of non-radicals (R – R, ROOR).

The PV is one of the most common quality indicators of fats and oils during production and storage (Antolovich et al., 2002; Riuz et al., 2002). The lower the peroxide value, the better the quality of the fat or oil (Gunstone *et al.*, 2007). The peroxide value is determined by measuring the amount of iodine which is formed by the reaction of peroxides (formed in fat or oil) with iodide ion (AOCS, 2014)

$2\,I^- + H_2O + HOOH \longrightarrow HOH + 2OH^- + I_2$……..……...…… (vii)

The base produced in this reaction is taken up by the excess of acetic acid present. The iodine liberated is titrated with sodium thiosulphate (AOCS, 2014)

$2S_2O_3^{2-} + I_2 \longrightarrow S_4O_6^{2-} + 2\,I^-$… …….…….…….…… ….. (viii)

2.12.2 Iodine value

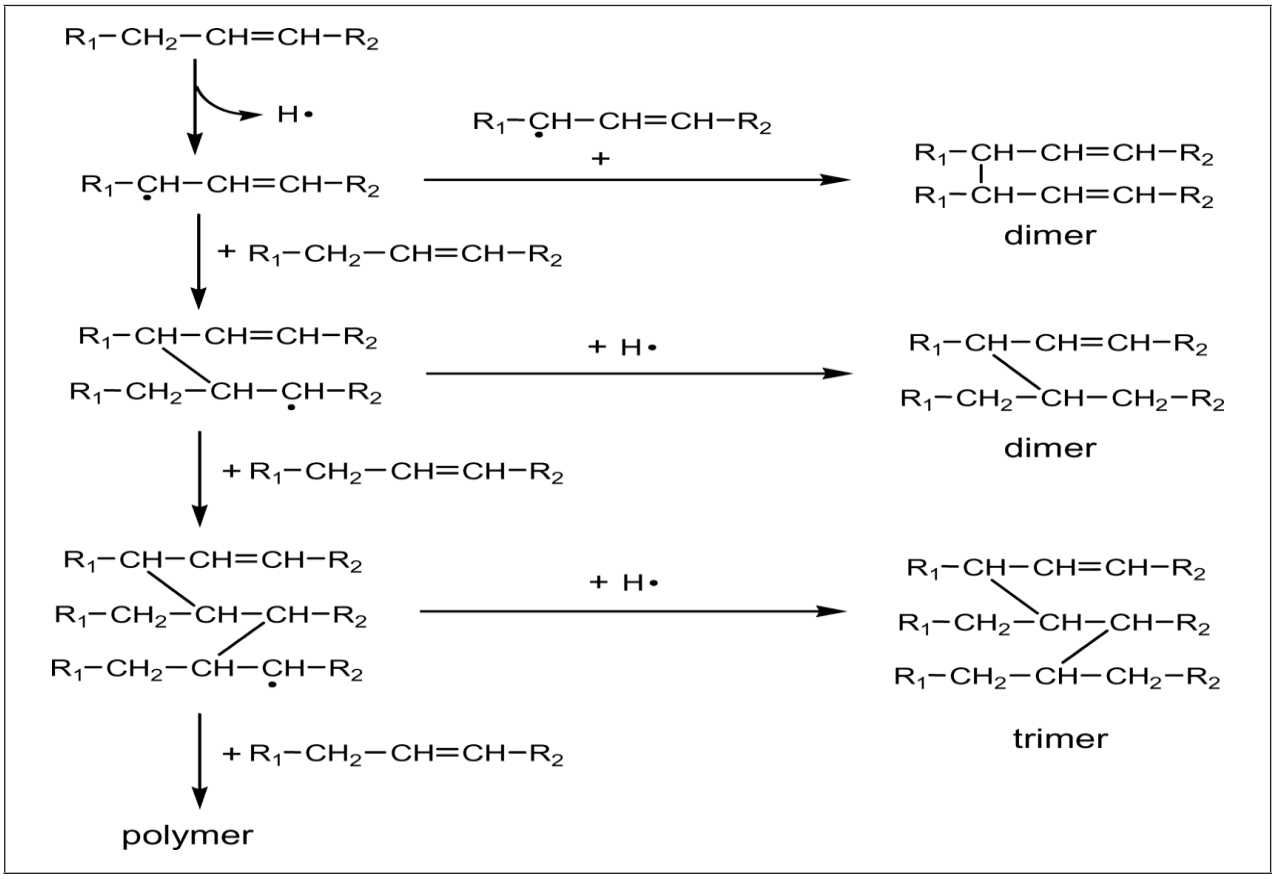
Iodine value is used to measure unsaturation or the average number of double bonds in fats and oils. It is defined as the number of grams of iodine that could be added to 100 g of oil, which is measured with the AOCS Method cd 1-25. A high, iodine value indicates high unsaturation. (AOCS, 1990).The chemical reaction associated with this method of analysis involves formation of the diiodo alkane (Firestone, 1984)

$R_1\text{- CH=CH-}R_2 + I_2 \longrightarrow R_1\text{-CHI-CHI-}R_2$……(ix)

2.12.3 Polymer content

The content of polymers components in oils increases during frying process. The determinations of polar compounds and polymers are the two most recommended analytical techniques for the quality control of used frying fats and oils (DGF, 2000; Marmesat *et al.*, 2007). During frying and heating, oxidation, polymerization, isomerization (in both frying and heating) and hydrolysis (only during frying) occur in the oil generating a multitude of

products (Belitz *et al*., 2004). Among these products, higher molecular weight products compared to triacylglycerols are generated originating from polymerization and oxidation reactions (Dobarganes and Márquez-Ruiz, 1996; Kalogianni *et al*., 2009, 2010). This increases the polymer content.

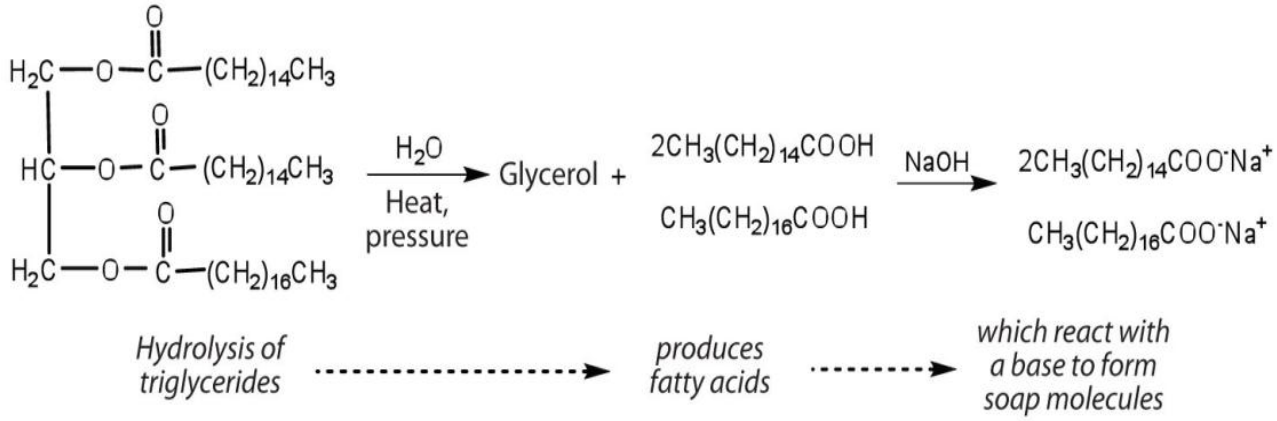


(Adapted from Choe and Min, 2007)

Figure 2.3: Polymer formation during heating

2.12.4 Saponification value

Saponification value is a measure of the alkali-reactive groups in fats and oils and is defined as the mg of KOH needed to saponify 1 g of oil. Shorter chain fatty acids give higher saponification values than do longer chain fatty acids (Shahidi, 2005). Hydrolysis of fats and oils in the presence of an alkali is called saponification. Most soap is prepared by the hydrolysis of triglycerides



(Source: Ball, 2012)

2.12.5 *P*-Anisidine value

Anisidine value measures the amount of unsaturated aldehydes in fats and oils. It is a method for measuring secondary decomposition products such as aldehydes and ketones (Mariod *et al.,* 2006). The *p*-AnV is a reliable indicator of oxidative rancidity in fats and oils and fatty foods (Merwe van der *et al.*, 2003). High anisidine levels usually indicate harsh or excessive processing (AOCS, 1990).

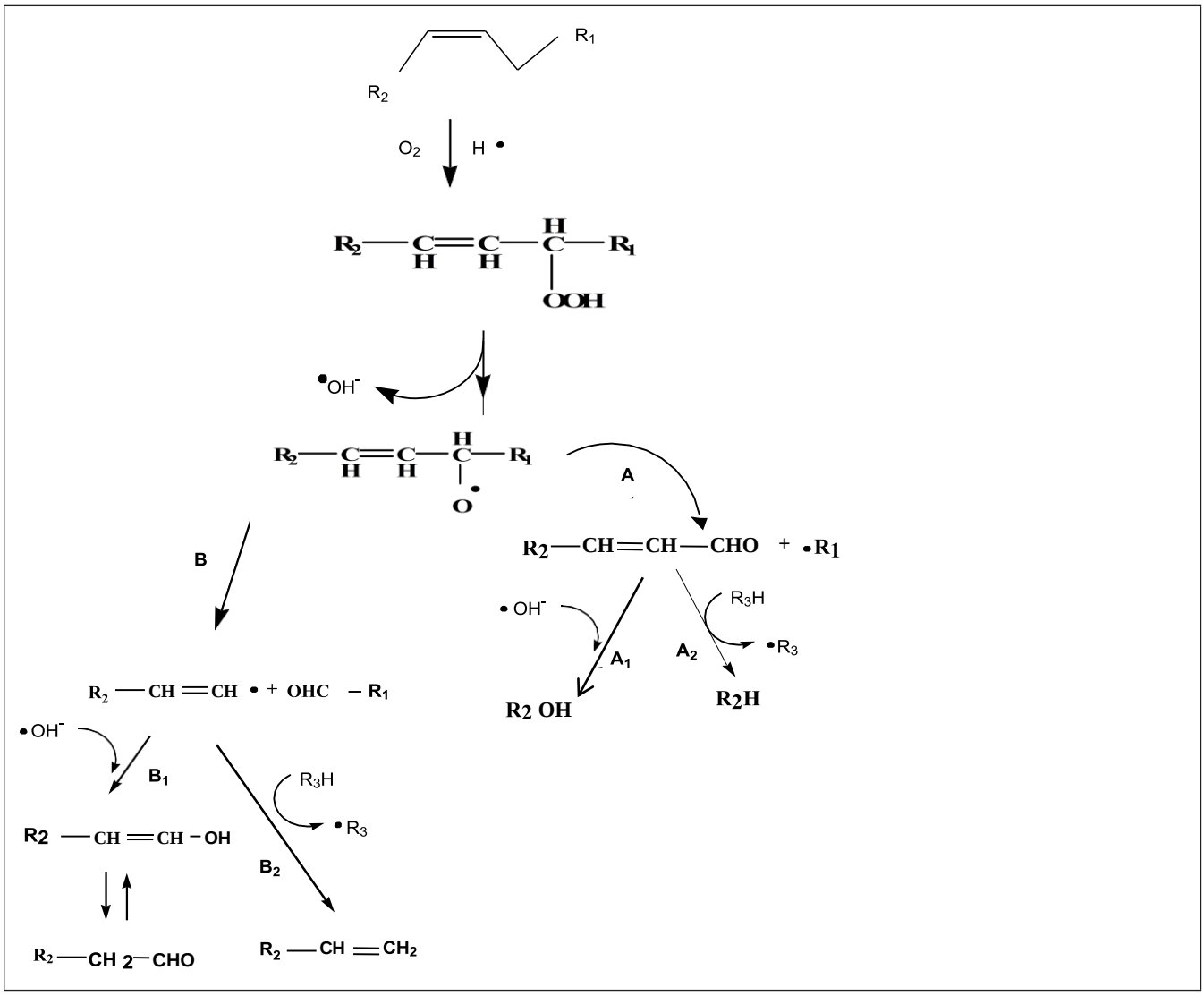


Figure 2.4: Mechanisms of hydro peroxide decomposition to form secondary oxidation products (Choe and Min, 2006).

2.12.6 Gas chromatography

Gas chromatography (GC) is a common type of chromatography used in analytical chemistry for separating and analyzing compounds that can be vaporized without decomposition. Typical uses of GC include testing the purity of a particular substance, or separating the different components of a mixture (the relative amounts of such components can also be determined). In some situations, GC may help in identifying a compound (Pavia *et al.*, 2006). In GC the components in a sample are identified by comparing the retention times of the peak in the sample chromatogram with those of the standard compounds. In

gas chromatography the stationary phase is contained in a column. The column generally is a coiled metallic or glass tube. An injector near the entrance to the column is used to add the analyte. The mobile phase gas usually is contained in a high pressure gas cylinder that is attached by metallic tubing to the injector and the column. A detector, placed at the exit from the column, responds to the separated components of the analyte. The detector is electrically attached to a recorder or other readout device (like a computer) that displays the detector response as a function of time. The plot of the detector response as a function of time is a chromatogram. Each separated component of the analyte appears as a peak on the chromatogram (https://www.britannica.com/science/stationary-phase-chromatography)

CHAPTER THREE
MATERIALS AND METHODS

3.1 Preparation of the sample

Chips: Fresh potatoes were peeled and sliced to a thickness of 2 mm using a mechanical slicer

Fish: Fresh fishes were washed with warm water and sliced into sizeable portions

Chicken: Chicken were placed in the sink and rinsed off with cold water to clean it. Each piece of chicken was then sprinkled with the seasoning.

Smokies: Smokies were cut into single pieces

Mandazi: Milk, oil and water were mixed up in one bowl (watery stuff). The flour, baking powder and sugar (dry stuff) was also mixed up in a different bowl. All the dry stuff was then poured into a large bowl. A hole was made in the middle and the watery stuff poured slowly. After putting half of the watery stuff, stirring was done. This continued until dough that could be rolled out was obtained. It was left to rest for about 20 minutes. The dough was placed on a flat surface and rolled into a large round shape then cut into pieces ready for frying

3.2 Frying process

Vegetable oils were bought from various markets in Kenya. The vegetable oils were then analyzed before use. About 5 litres of each of the five vegetable oils was heated in a domestic fryer at a temperature of 160-190^{0}C and allowed to equilibrate at this temperature for 10 minutes. About 1kg of each of the five types of food was intermittently fried in the heated oils for 20 minutes at intervals of 30 min for a period of 6 hours. After 6 hours, about 250 ml of the heated oil was drawn for analysis. The remaining was left to cool at different temperature conditions; 2 litres at room temperature and the remaining 2 litres refrigerated at about 4^{0}C. After 5 days, the procedure was repeated using the stored oils but the ratio of food to oil was considered. The oil samples were put in bottles and stored ready for analysis.

3.3 Determination of peroxide value

About 5g of the sample of the oil was weighed and placed in a 250 mL conical flask. A sample of the mixture glacial acetic acid: chloroform ratio 3:1 was then added. The flask was swirled until the sample was completely dissolved. About 0.5 mL of the saturated potassium iodide solution was then added and allowed to stand in darkness for 1 minute with occasional shaking. About 30 mL of distilled water was added. The contents was stoppered and shaken

vigorously to liberate iodine from the chloroform layer. Sodium thiosulphate (0.1 M) was then added drop wise until the yellow colour of iodine disappear**.** About 0.5 mL of 1% starch solution was added and the titration continued while shaking vigorously until the blue colour disappeared.

$$\text{Peroxide value (PV)} = \frac{(a-b) \times M \times 1000}{\text{Weight of sample (g)}} \quad \ldots\ldots(x)$$

a=volume of $Na_2S_2O_3$ for the blank sample

b=volume of $Na_2S_2O_3$ for the sample

W= weight of sample (g)

M=0.01M, Concentration of Sodium thiosulphate (IFRA, 2011)

3.4 Determination of iodine value

About 5 g of oils was weighed into conical flask and 20 mL of carbon tetrachloride added to dissolve the oil. About 25 mL of Wijs reagent was added to the flask using a measuring cylinder in a fume chamber. It was then stoppered and the content of the flask vigorously swirled. The flask was then placed in the dark for 1 h. At the end of this period, 20 mL of 10% aqueous potassium iodide and 100 mL of water was added using a measuring cylinder. Excess iodine was then titrated with 0.1 M sodium thiosulphate solution. About 1% starch was used as an indicator. The same procedure was used for the blank test. The Iodine Value (I.V) is given by the expression;

$$\text{Iodine Value (IV)} = \frac{12.69C\,(V_1 - V_2)}{M} \quad \ldots\ldots(xi)$$

Where C = concentration of sodium thiosulphate

V_1 = volume of sodium thiosulphate used for blank

V_2 = volume of sodium thiosulphate used for determination

M = mass of sample

12.69= Constant. (AOCS, 1991)

3.5 Determination of the saponification value

An American Standard for Testing Material (ASTM) method was used for the determination of the Saponification Values of the vegetable oils. About 5 g of the oil was weighed into the conical flask. About 25 mL of 0.5 M ethanolic KOH was then added and the resulting

mixture refluxed for 1 hour. The resulting solution was subsequently titrated against 0.5 M HCl with phenolphthalein as indicator. The end point was obtained for each titration. The same procedure was used for the blank. The Saponification value (SV) was then calculated using the expression given below;

$$\text{Saponification value (SV)} = \frac{(B-S) \times N \times 56.1}{\text{Weight of sample}} \quad \text{.....(xii)}$$

Where;

B = volume of HCl required by blank

S = volume of HCl required by sample

N = Molarity of HCl

56.1 = Molar mass of KOH (ASTM, 2011)

3.6 Determination of refractive index

Abbey refractometer (Bellingham and Stanley RFM 330) was used in this determination. A drop of the sample was transferred into a glass slide of the refractometer. Water at 30^0C was then circulated round the glass slide to keep its temperature uniform. The refractive index of the oil was then read. This was repeated and the mean value noted and recorded as the refractive index.

3.7 Determination of density

Density was determined manually. A clean dry 50 mL graduated cylinder was weighed and approximately 30 mL of oil was placed on the weighed cylinder. The volume of the oil was then read and recorded. The graduated cylinder with its contents was then placed on the weighing scale. The density was then determined using the formula below. The procedure was repeated to find the density of each liquid.

Mass of graduated cylinder = A

Mass of cylinder and oil =B

Mass of oil = B-A

Volume of oil = V

$$\text{Density of oil} = \frac{B-A}{V} \quad \text{.....(xiii)}$$

3.8 Polymer content

Polymer content (PC) was determined according to the method described by Peled *et al.* (1975). Polymer content was measured to determine the portion of oil that remains insoluble in methanol at 25^0C. Subsequently, 250 mL methanol was added to a 500 mL conical flask

containing 1% sulphuric acid and 250 g of heated oil. The conical flask was then placed on a hot-plate magnetic stirrer; the methanol-oil mixture was boiled under a reflux condenser for 2 h and cooled to 25^0C. The methanolic miscella was decanted thoroughly and the methanol-insoluble fraction washed with 15 mL of methanol. The insoluble portion was then dissolved in 40 mL petroleum ether (60-80 ^{0}C) and then transferred to a pre-weighed flask. The solvent was evaporated initially by a rotary vacuum evaporator NEB 1001, Japan) and then by placing the flask in a vacuum oven for 2 h at 140 ^{0}C. The weight of polymers was then recorded.

3.9 Determination of *p*-Anisidine value.

Para-Anisidine was dissolved in glacial acetic acid to make up a 0.25 g/100 mL solution. *Iso*-octane was used as a solvent for the oil samples. The test was conducted in triplicates for all samples. About 0.6 g of the oil samples were accurately weighed in 25 mL volumetric flasks and then diluted to the mark with *iso*-octane. The absorbance (A_b) of the resulting solutions at 350 nm was measured using *iso*-octane as the reagent blank. Glass cuvettes were used for all absorbance measurements. About 5 mL of the solution was then pipetted into a test tube and 5 mL of *iso*-octane into another test tube. *Para*-Anisidine solution (1 mL) was added to both and the solutions mixed. The absorbance (A_s) of the sample solutions was then read with the *iso*-octane as blank after 10 minutes. The *p*-Anisidine values were then calculated using the following equation:

$$AV = \frac{25\,(1.2A_s - A_b)}{M}$$

A_s = Absorbance of test solution B at 350 nm

A_b = Absorbance of test solution A at 350 nm

M = Weight in g of the substance to be examined in test solution A (AOCS, 2011)

3.10 Gas chromatography analysis of fatty acids

3.10.1 Purification of the lipid extracts

About 100 mL of 0.88 per cent potassium chloride solution in water was added to the total lipid extracts and the mixture thoroughly shaken before being allowed to settle. The solvents partitioned into a lower layer and an upper layer. The lower layer contained the purified lipids and the upper phase contained the non-lipids contaminants. The purified lipid layer was filtered before the solvent was removed at 35^0C on a rotary evaporator. The weight of the lipid recovered was determined.

3.10.2 Preparation of fatty acid methyl esters (FAMES)

Based-catalyzed trans esterification was used in conjunction with acid catalyzed esterification and trans esterification. About 1.0 g of the lipid mixture and 10 mL of 0.5 N methanolic NaOH was placed in a 100 mL round bottomed flask fitted with a condenser. The mixture was refluxed for 10 minutes until the lipids dissolved. 12 mL of boron trifluoride-methanol complex (about 14% w/w BF_3) was added through the top of the condenser and refluxing done for a further two minutes. The solution was cooled, 5 mL hexane added and the mixture boiled once more for two minutes. A solution of saturated sodium chloride was then added, the organic layer separated and dried with anhydrous sodium sulphate. Under the conditions described no isomerization of double bonds in polyunsaturated fatty acid occurs.

3.10.3 Analysis of the FAMES by GLC

Two standard types of chromatographic stationary phases were used for this purpose. Both were polyester phases, namely polyethylene glycol adipate (PEGA) and diethylene glycol succinate (DEGS). These polyester column materials resolve fatty acids esters according to both chain length and the degree of unsaturation. Two stainless steel columns (2 mm by 3.2 mm Od) were used. One column was supplied already packed with 10% (w/w) PEGA on acid washed chromosorb w, A/W mesh size 80/100. The second column was packed in the laboratory with 15% DEGS on acid- washed chromosorb w, A/W mesh 80/100. The two columns were first conditioned by passing nitrogen gas through them at 180^0C for 5 hours. The columns were then fitted on a GOW-MAC gas chromatograph equipped with flame ionization detector (F.I.D), column temperature was isothermal throughout the column length at 180 ± 1^0C. Injector- detector temperature was $240 \pm 1\ ^0$C, nitrogen gas flow rate was 40 mL/ min. A sample of the FAMES in hexane solution was injected into the GC using a micro liter syringe. There was no need to control the amount of the solution actually injected as long as peaks of reliably measurable size were obtained.

Several major peaks were observed on the GC chart paper. Thus there were seven major fatty acids in the injected sample. Provisional identification of the fatty acids was done by direct comparison of the retention times of their methyl esters with those of some known standard methyl esters (Sigma Chemicals, U.S.A) on the same two columns under identical conditions. The identity of the FAMES was confirmed by carrying out their gas chromatograph analyses at the Tea Research Foundation of Kenya.

For gas chromatographs equipped with a flame ionization detector (FID) the areas under the peaks on the GC chart traces are linearly proportional to the amount of material eluting from the columns. For the purposes of quantitative fatty acid composition analysis it was assumed that all the sample together with the internal standard were injected and that the area response recorded for the internal standard represented its weight. The weights of the other peaks were then directly calculated from the weight of the internal standard. The peak areas of the component FAMES were measured manually by multiplying the height of the peak by the width at half length. The peak area of the sample components were then corrected by a multiplying factor and divided by the peak area of the internal standard to give the concentration of the particular component. This can be expressed in the form of an equation as follows,

$$\frac{\text{Peak area of component} \times \text{multiplying Factor} \times \text{weight of internal standard}}{\text{Peak area of internal standard}} = \text{Weight of the component}$$

The multiplying factor was obtained by performing GC of a standard solution of representative fatty acids prepared on an equal weight basis. The multiplying factors or response ratios in terms of the internal standard was then calculated by dividing the peak area of the internal standard by the peak area of each component. Weight of the internal standard is the amount (0.015 g) of internal standard added to the lipid extracts.

3.11 Statistical analysis

Data obtained was presented in form of tables. All statistical analysis was done using STATA 13. Results of analysis were displayed in form of tables, mean and standard deviations. ANOVA was used to determine the significant differences ($p \leq 0.05$) of the oil quality indicators.

CHAPTER FOUR
RESULTS AND DISCUSSION

4.1 Introduction.

The chapter presents the results of analysis with pertinent discussions as per the study objectives. Various physical and chemical methods were used to assess the oils and these are peroxide value, saponication value, iodine value, *Para*-anisidine, polymer content, refractive index, density and fatty acid composition. At the beginning of the chapter, various descriptive statistics are displayed including tables, chart as well as summary statistics such as means and standard deviations.

4.2 Fatty acid composition (ug/ml).

Fatty acid composition of oils before and after use were compared using a chart.

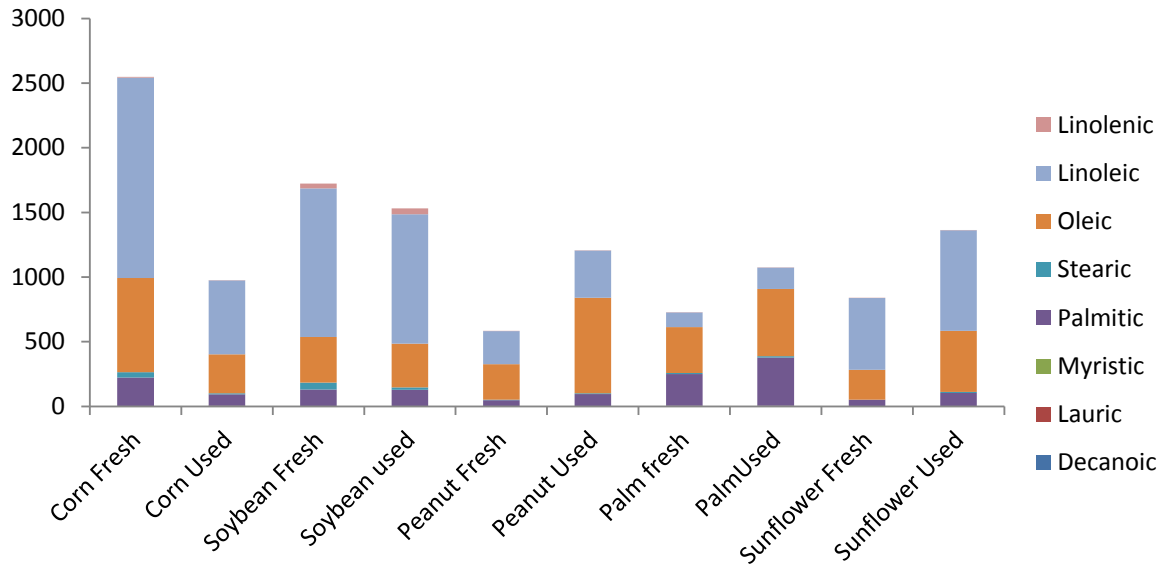


Figure 4.1: Fatty acid composition (ug/mL)

From figure 4.1, Corn oil recorded high linoleic (1550.00 ug/ml) but low linolenic (4.48 ug/ml) acids. Soybean on the other hand had high values of linoleic (1149.20 ug/ml) and linolenic (36.23 ug/ml) (appendix 1). Fresh oils of peanut and palm showed relatively lower linoleic and linolenic acid contents than other fresh samples. Palm oil had low linoleic and linolenic acids and high Palmitic acid contents. It is evident from the results that palm oil had low polyunsaturated fatty acids (PUFAS) and high monounsaturated fatty acid (MUFAS) while soybean and corn had high PUFAS and low MUFAS. According to Kochhar and Henry

(2009), Vegetable oils which are rich in PUFA are more prone and less stable to oxidation compared to those which are rich in MUFA, whereas oils that are rich in MUFA, such as palm oil and olive oil, can better withstand oxidation and form less degradation products on repeated heating **(**Kochhar and Henry, 2009). Palm oil also has an abundant content of vitamin E, which may play an important role in its ability to withstand thermal oxidative changes (Quiles *et al*., 1999). Vitamin E which effectively protects fatty acids in the oil from oxidation deteriorates after each frying episode (Adam *et al*., 2007). Therefore, repeated heating of frying oils destroys the vitamin E content and exposes the fatty acids to oxidation. The vitamin E content of palm oil mainly consists of tocotrienols, while the vitamin E in soybean oil mainly consists of tocopherols (Adam *et al*., 2007). Tocotrienols have better antioxidant capacity than tocopherols (Bardhan *et al*., 2011) and this may have contributed to the better resistance of palm oil to oxidative changes due to repeated heating.

From the results in this study (appendix 1), it is observed that soybean oil recorded a sharp decrease in linoleic and linolenic acids. It also indicated an increase in myristic, palmitic and stearic acid contents. This is due to oxidation. Comparatively, decanoic was highest in corn (1.11) followed by palm (0.01) while on average 0 content in other oil types. Similarly, lauric was highest in corn (3.17) and least in peanut (0), but with slightly above 0 content in other oil types on average. In regard to myristic, the highest concentration was in palm (4.08) with least concentration in corn (0.47). Likewise, palmitic had least concentration in peanut (83.56) but highest concentration in palm (353.30); stearic was most concentrated in soybean (24.18) and least in peanut (4.93). At the same time oleic was least concentrated in soybean but highest in peanut. Concerning Linoleic, it was most concentrated in soybean (1025.59) and least concentrated in palm (157.50). Similarly linolenic was least concentrated in palm (0.80) and highest in soybean (43.90) (Figure 4.1)

Most of the samples indicated higher linoleic and linolenic acids before frying than after frying (appendix 1). There was an increase in palmitic acid in the samples. According to Sharoba and Ramadhan (2012), the increase in SFA during repeated frying may not due to oxidative alteration but to interaction between the bath oil and the lipids of the fried food products. The oils in which mandazi was fried recorded a relatively higher decrease in linoleic acids than the oils used to fry other types of food. There was an increase in oleic acids in all the samples. Soybean oils had relatively high PUFAS and low palmitic acid contents. Palm oils had relatively lower PUFAS than the other oils. Palm oil in which chips

were fried had higher PUFAS than the one for fishes. It was also noted that palm oil samples had high palmitic acid contents. There was an increase in PUFAS in sunflower and peanut oils (Appendix 1).

The oil which was used to fry chicken for the second time after storage at room temperature for 5 days indicated relatively higher increase in myristic, palmitic, stearic and oleic acids (appendix 1). It also recorded a sharp decrease in PUFAS. This indicates oxidation in the said oil. It might be due to the differences in the composition of the fried product (Anwaar *et al*., 2012). Chicken produced more degradation products than other food in the frying oil. The type of food being fried influences the oil or fat fry-life (Freire *et al*., 2013). Oil, in which chips were fried on the other hand, had low linolenic (0.72) and linoleic (207.34) acids. It also had high palmitic (454.49) and myristic (5.52) acids (Appendix1). According to Sharoba and Ramadhan (2012), level of PUFA tended to decrease, whereas that of SFA increased during frying. It was reported that heat treatment of fats induces modifications of fatty acids with two or three double bonds (Orthoefer and Cooper, 1996). ANOVA was done to establish the fatty acids that varied per the type of oil. It was found that myristic, palmitic, linoleic and linolenic acid varied significantly ($p \leq 0.05$) per the type of oils (Appendix 16).

4.3 Oxidation stability of oils after frying various foods.

Seven quality parameters were used to measure oxidative stability of various oils used to fry different food types. Changes in certain physical and physicochemical properties of oils during frying have been blamed on the food that undergoes frying (Kaloggiani *et al*., 2010).

Table 4.1: Recommended physicochemical parameters by FAO/WHO (1993)

Type of oil	Peroxide value	Saponification value	Iodine value	Refractive index	Density
Palm	≤10	190-209	50-55	1.449-1.459	0.891-0.899
Corn	≤10	187-195	103-135	1.465-1.468	0.917-0.925
Peanut	≤10	190	84-105	-	0.909-0.921
Soybean	≤10	189-195	120-143	1.466-1.470	0.909-0.921
Sunflower	≤10	188-194	110-143	1.467-1.469	0.909-0.921

4.3.1 Peroxide value.

The peroxide value is a measure of the concentration of peroxides and hydro peroxides formed in the initial stages of lipid oxidation. High peroxide values indicate high oxidation; it shows that there is high amount of active oxygen bound by the oil. During frying, peroxides and hydro peroxides are formed immediately. However, they decompose spontaneously at temperatures of above 150^0C.

Table 4.2: Peroxide values of oils before and after frying food (meq/kg).

Type of Oil	Fresh oil	Chicken			Chips			Fish			Smokies				mandazis				
		6 hrs	2nd fry RT	2nd fry R	6hrs	2nd fry RT	2nd fry R	6hrs	2nd fry RT	2ndfry R	6hrs	SRT 5d	2nd fry RT	R 5d	6hrs	SRT 5d	2nd fry RT	R 5d	2nd fry R
Palm	5.0±0.08	6.6±0.08	6.5±0.16	6.4±0.16	6.0±0.16	5.4±0.32	4.4±0.08	5.2±0.48	3.8±0.06	8.4±0.16	4.8±0.08	5.4±0.04	5.6±0.21	5.6±0.49	6.2±0.08	5.0±0.04	4.8±0.08	5.4±0.49	6.2±0.04
Corn	2.8±0.04	3.6±0.07	3.4±0.49	4.4±0.16	3.4±0.45	3.2±0.03	3.4±0.49	3.6±0.07	3.4±0.05	3.2±0.06	3.6±0.08	3.8±0.11	3.8±0.16	3.6±0.04	3.4±0.49	3.6±0.04	3.8±0.03	3.6±0.28	4.2±0.31
Peanut	4.2±0.08	4.2±0.07	4.4±0.03	4.2±0.02	3.4±0.24	3.6±0.49	3.4±0.23	4.8±0.08	6.2±0.49	4.8±0.23	4.2±0.08	4.4±0.02	4.6±0.03	4.6±0.07	4.6±0.02	4.8±0.03	5.2±0.02	5.2±0.49	5.4±0.06
Soy-bean	3.8±0.04	4.2±0.08	4.8±0.18	4.4±0.21	2.8±0.26	5±0.28	3.2±0.31	3.6±0.02	3.4±0.07	6.2±0.16	4.4±0.08	4.6±0.04	5.2±0.11	4.6±0.16	4.6±0.07	5.4±0.49	5.2±0.28	4.8±0.02	4.8±0.03
Sunflo-wer	0.8±0.04	0.8±0.03	1±0.07	0.9±0.06	5.2±0.49	1.4±0.49	0.8±0.08	4.8±0.21	4.8±0.08	4.7±0.07	1.2±0.08	5.8±0.07	1.4±0.16	5.4±0.08	1.4±0.02	5.2±0.03	5.4±0.02	4.8±0.07	5.2±0.07

6 hrs: Used for frying for 6 hrs **2nd fry RT:** Used for frying after 5 days of storage at room temp (Total frying time: 12 hrs)

SRT 5 d: Stored at room temp for 5 days

R 5 d: Stored at 4^0C for 5 days

2nd fry R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs)

Table 4.3: Peroxide values after frying in different foods (meq/kg).

	Food type					
Type of oil	**Chicken**	**Chips**	**Fishes**	**Mandazi**	**Smokies**	**Mean**
Corn	3.80±0.50	3.33±0.10	3.40±0.20	3.72±0.10	3.72±0.30	3.59
Palm	6.50±0.10	5.27±0.80	5.80±2.30	5.52±0.40	5.28±0.70	5.67
Peanut	4.27±0.10	3.47±0.10	5.27±0.80	5.04±0.20	4.52±0.30	4.51
Soybean	4.47±0.30	3.67±1.10	4.40±1.60	4.96±0.30	4.72±0.30	4.44
Sunflower	0.90±0.10	2.47±2.68	4.77±0.00	4.40±2.30	3.84±1.70	3.28
Mean	3.99	3.64	4.73	4.73	4.42	4.30

Table 4. 4: Peroxides values of oils based on the storage conditions (meq/kg).

	Type of oil					
Storage	**Corn**	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	2.80±0.0471	5.00±0.08165	4.20±0.08165	3.80±0.0471	0.80±0.0471	3.32
6 hrs	3.52±0.1095	5.76±0.7403	4.24±0.5367	3.92±0.7294	2.68±0.1335	4.02
Refri 5days	3.60±0.0000	5.50±0.1000	4.90±0.3000	4.70±0.1000	5.10±0.3000	4.76
SRT 5days	3.70±0.1000	5.20±0.2000	4.60±0.2000	5.00±0.4000	5.50±0.3000	4.33
2nd fry ref	3.80±0.4560	6.08±1.3775	4.52±0.6764	4.68±0.9600	3.40±2.0947	4.49
2nd fry SRT	3.52±0.2400	5.22±0.8953	4.86±0.0.9024	4.72±0.6764	2.80±1.8931	4.22
Mean	3.62	5.55	4.62	4.60	3.86	4.32

Codex alimentarius commission of FAO/WHO recommends a P.V of ≤ 10meq/Kg (table 4.1). The results showed that peroxide values for fresh oils ranged from 0.8-5.0 meq/kg (table 4.3) which was below the limit indicated. According to Gunstone 2008, freshly refined vegetable oils should have a PV of less than 1.0meq/kg and considered to be rancid if it is above 10 meq/kg.

Average peroxide value was recorded after frying different foods under varied conditions such as first fry, second fry, second fry after storage at room temperature and second fry after storage at below 4^0C (table 4.2). The results showed that peroxide value (meq/kg) in Corn oil did not vary widely among the food type chicken 3.80; chips 3.33; fishes 3.40; mandazi 3.72; smokies 3.72, and having a mean of 3.59 (table 4.3). Peroxide value in corn oil was highest when used to fry Chicken and least when used to fry chips. Palm oil had relatively high values of peroxide value in meq/kg: chicken 6.5; chips 5.27; fishes 5.8; mandazi 5.52;

smokies 5.28 and a mean of 5.67 (table 4.2). For peanut, the highest peroxide value was recorded in food type fishes (5.27 meq/kg) followed by mandazi with 5.04 meq/kg with a mean of 4.51. For soybean oil, all the peroxide values were below 5.00 with the highest peroxide value recorded in mandazi (4.96 meq/kg) and least in chips (3.67 meq/kg). The mean was 4.4. The peroxide values in sunflower oil were relatively lower. The highest P.V was 4.77 meq/kg and least 0.9 meq/kg with respect to food type fishes and chicken respectively. Soybean and palm oils indicated high peroxides values (4.47 and 6.50 meq/kg respectively) with respect to food type chicken (table 4.3). Peanut and sunflower on the other hand showed high peroxide values when used to fry chips.

In terms of mean, the highest P.V was recorded in palm and the least in sunflower. The highest increase was indicated by sunflower with respect to fish (4.00meq/kg). Palm oil also recorded a relatively higher increase (1.50 meq/kg) with respect to chicken. This therefore means that oils are highly oxidized while frying fish and chicken. This could be due to the fish oil introduced to the frying oil while frying fish. Fish oil is very susceptible to autoxidation because of the high degree of polyunsaturated fatty acids (Medina *et al.,* 2009). This therefore means that the oils in which fish is fried should not be used for prolonged period of time. Overall, palm oil indicated high peroxide values across the five types of food leading to the highest mean of 5.67 meq/kg. Peroxide values for in-use vegetable ranged from 0.90-6.50 meq/kg (table 4.2). A good quality frying vegetable oil should have a PV of less than 2.00 meq/kg (Sulieman *et al*., 2006). The PVs of frying oils were below the rancidity limit of 10.00 meq/kg as suggested by FAO/WHO. This could be due to the rapid decomposition of peroxides and hydrogen peroxides immediately they are formed.

From the foregoing analysis, there was preliminary evidence that the quality of different oils as measured by the Peroxide value were affected differently based on the food that was being fried. To understand the nature of the relationship, a two-way ANOVA was used to assess whether the type of food affected the quality of oil. The main effects were significant ($p \leq 0.05$) as well as the interaction effect (Appendix 2).

Peroxide values increased in all the samples subjected to different frying time and storage conditions. The oils that were used for only 6 hours recorded small increase as compared to other treatments with a mean of 4.02 (table 4.4). Palm oil recorded highest mean of 5.55 while corn had the lowest mean of 3.62 (table 4.4). The oils that were refrigerated for five days before being used for frying (12 hr) recorded relatively higher P.Vs (Table 4.4).

Preliminary evidence shows that peroxide values of oils are affected by different storage conditions. It is clear that the peroxide value is affected by duration of storage as well as frying (table 4.2). From the results the PVs obtained could be lower than expected since peroxides and hydro peroxides decompose at high temperature. In some oils there was a decrease in the peroxide values when the oils were used for the second time. This could be due to formation of secondary oxidation products. According to Bester *et al* (2008), PV decreases as oxidation proceeds due to rapid decomposition of hydro peroxides and therefore may not indicate the actual extent of oil deterioration and is not recommended for measuring oil deterioration during the frying process (Farhoosh and Moosavi, 2009). This is because their total accumulation may be greatly underestimated. From the results it was noted that there was a significant relationship between the oils and storage conditions with the peroxide value. This is in agreement with the previous findings which stated that storage temperature had a significant effect on the peroxide value of all oils regardless of the oils matrix (Jamie *et al*., 2012).

Anova was done to ascertain whether the storage conditions affect the type of oil. From the results, the model showed significant difference as well as the main effect oil. The interaction effects were not significant (Appendix 9).

4.3.2 Saponification values.

Saponification value is an indication of the molecular weights of triglycerides in oil. It is inversely proportional to the average molecular weight or chain length of the fatty acids (Muhammad *et al*., 2011). Therefore, the shorter the average chain length (C4-C12) the higher is the SV (Tamzid *et al*., 2007). Increase in saponification value indicates oxidation and it shows that the oil could be used in the production of soap

Table 4.5: Saponification values of oils before and after frying food

Oil	Fresh Oil	Chips			Chicken			Fish			Smokies					Mandazis				
		6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R
A	185.13 ±0.09	189.5 ±0.81	193.6 ±1.22	211.8±1.88	203.4 ±1.91	218.8±3.10	210.4 ±3.27	206.18±3.5	213.3±2.69	200.6±2.94	189.5 ±3.5	151.5±1.8	231.5 ±1.88	203.4±3.02	202.0±2.94	202.0 ±3.10	224.4 ±4.50	231.4 ±3.5	172.5 ±3.43	242.6 ±3.51
B	173.9±0.09	173.9 ±1.47	200.6 ±2.04	199.2 ±1.39	199.2 ±1.88	196.4±2.12	196.4 ±2.37	200.6±2.61	189.4±2.69	204.8±3.10	217.4 ±3.02	242.6±3.18	232.8±2.78	189.7±2.69	232.8±2.61	171.1 ±1.88	182.3 ±2.12	161.3 ±1.22	173.9 ±1.79	213.1 ±1.91
C	164.1±47.14	185.1 ±2.04	204.8 ±1.39	197.8±1.88	210.4 ±4.50	218.8±3.51	197.8 ±2.45	210.4±2.61	210.4±2.94	188.0±1.04	255.3 ±6.37	165.5±1.39	210.4±1.88	199.2±2.04	225.8±2.20	155.7 ±1.63	211.8 ±1.22	238.4 ±1.46	164.1 ±1.91	200.6 ±2.04
D	173.9±4.62	196.4 ±2.22	204.8 ±2.20	204.8±2.20	195.0 ±3.43	203.4±3.10	192.2 ±1.22	204.8±2.20	204.8±2.29	190.3±1.91	216.9 ±5.23	224.4±1.91	231.4±1.39	217.3±3.02	224.4±2.45	126.0 ±3.10	230.0 ±3.43	204.8 ±1.80	193.6 ±2.12	224.4 ±2.61
E	168.3±8.16	185.13±2.61	190.8 ±2.86	189.4±1.39	196.4 ±1.39	190.8±1.63	187.9 ±1.39	196.4±1.88	204.8±2.37	187.9±2.69	231.4 ±8.12	165.5±1.47	234.2±1.39	220.2±2.04	232.8±2.45	161.3 ±2.45	171.1 ±3.02	199.2 ±2.94	164.1 ±2.12	144.5 ±2.12

6hrs: Used for frying for 6 hrs
SRT 5 d: Stored at 25^0C for 5 days
2nd fr RT: Used for frying after 5 days of storage at room temp (Total frying time: 12 hrs)
R 5d: Stored at 4^0C for 5 days
2nd fr R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs)

A: Palm
B: Corn
C: Peanut
D: Soybean
E: Sunflower

Table 4.6: Mean Saponification values of oils after deep frying in food

	Food type					
Type of oil	**Chicken**	**Chips**	**Fish**	**Mandazi**	**Smokies**	**Mean**
Corn	191.20±1.6	197.28±15.0	198.23±8.0	180.36±19.0	203.09±63.0	194.03
Palm	194.94±7.7	210.85±16.2	206.68±6.4	214.58±27.0	186.54±37.0	202.72
Peanut	195.88±10.6	208.98±9.6	202.89±13.0	194.10±34.0	211.21±33.0	202.61
Soybean	201.97±5.8	196.82±4.9	199.93±8.4	213.74±14.0	222.89±6.0	207.07
Sunflower	188.41±4.3	191.68±2.9	196.35±8.4	168.02±19.0	216.84±29.0	192.26
Mean	194.48	201.12	200.82	194.16	208.11	199.74

`

Table 4.7: Saponification values of oils based on storage conditions (mg KOH/g of oil)

Storage	Corn	Palm	Peanut	Soybean	Sunflower	Mean
Fresh oil	173.9±0.0942	185.1±0.9454	164.1±47.1475	173.9±4.6226	168.3±8.1649	173.06
6 hrs	192.4±19.5763	187.1±26.1057	203.4±36.7334	205.8±10.4261	194.1±25.2966	196.56
Refri 5d	181.4±8.3800	187.9±15.4375	181.6±17.5250	205.4±11.8875	192.2±28.0500	189.70
SRT 5d	212.5±30.1500	187.9±34.4625	188.6±23.1375	227.2±2.8000	168.3±2.8000	196.90
2nd fry ref	209.3±13.1121	213.5±15.2453	201.9±12.6709	207.2±14.9088	188.5±27.9517	204.08
2nd fry RT	196.1±22.9200	217.74±14.000	216.5±11.8200	210.5±10.5488	203.9±16.0547	172.94
Mean	198.34	198.82	198.40	211.22	189.4	198.68

Saponification values of fresh oils ranged from 164.10- 185.13 mg KOH/g (table 4.6) which were way below the range suggested of (187-209mg KOH/g) by FAO/WHO. The average S.Vs of frying oils ranged from 168.02-214.58 mg KOH/g. Sunflower recorded relatively lower values as follows: chicken = 188.41, chips = 191.68, fish = 196.35, mandazi = 168.02 and smokies = 216.84 (table 4.6). This led to the least mean recorded in sunflower (192.26). The saponification values were relatively higher in soybean oil and the values were as follows (in mg KOH/g): smokies = 222.89, mandazi 213.74, chicken = 201.97, fishes = 199.93 and chips 196.82 giving the highest mean of (207.07). High S.V indicated by soybean was expected because of high PUFAS observed in table 4.1 for this oil. In terms of the food, smokies recorded the highest mean of 208.11 and the least in mandazi with 194.16. The S.Vs was found to be significantly lower in oils before frying than after frying the food (table 4.5). Tyagi and Vasishtha, 1996 observed the same trend while frying potato chips at 180^0C. Increase in saponification values means oxidation. It indicates the presence of high percentage of fatty acids in the oil and therefore implies the possible tendency to soap formation (Omolara and Dosumu, 2009). From the results in table 4.6, the higher S.Vs observed in oils after frying food implied that oils are oxidized when used to fry food.

ANOVA was done to ascertain whether there was a significant difference ($p<0.05$) between the main effects oil and food as well as interaction between the two. The main and interaction effects were not significant (Appendix 3).From these results, the samples that were stored at room temperature for 5 days recorded a relatively higher S.V (217.54) (table 4.7). The saponification value was found to increase with storage time. This trend explains that with the long storage of these oils, fatty acids are likely to be formed which increase the SV. This also indicates that these long stored degraded oils can play a favorable role in producing soaps and toiletry products profitably (Bazlul *et al.,* 2010). The highest increase was observed in the oil stored at room temperature and for frying the second time 217.54 (table 4.5).

The results above showed that the saponification value of oils is affected by the storage conditions. To understand this, Anova was done to see whether there was significant difference between the main effects as well as interaction effects. Storage showed significant difference ($p<0.05$) while oil and interaction effects were not significant (Appendix 10).

4.3.3 Polymer content

This measures the polymers which build up in the oils during frying. Increase in polymer contents indicate oxidation due formation of larger molecular size compounds which are formed during frying. High temperatures of the frying operation produce high molecular cyclic fatty acid (FA) monomers, and TG dimers and oligomers (Henry and Chapman, 2002).

Table 4.8: Polymer contents of oils before and after frying food (g).

Type of Oil	Fresh Oil	Chicken			Chips			Fish			Smokies				Mandazis				
		6hrs	2nd fry RT	2nd fry R	6hrs	2nd fry RT	2nd fry R	6hrs	2nd fryRT	2nd fry R	6hrs	SRT 5d	2nd fry RT	R 5d	2nd fry R	6hrs	SRT 5d	2nd fry RT	R 5 d	2nd fry R
Palm	1.83± 0.01	1.88 ±0.08	1.89 ±0.07	2.05 ±0.07	1.76 ±0.49	1.85 ±0.06	1.77 ±0.28	1.84 ±0.31	2.00 ±0.02	1.85 ±0.24	1.89 ±0.24	1.97 ±0.06	2.03 ±0.28	1.92 ±0.03	1.97 ±0.07	1.85 ±0.03	1.89 ±0.03	2.55 ±0.02	1.86 ±0.07	1.95 ±0.06
Corn	1.75± 0.01	1.85 ±0.03	1.87 ±0.03	1.86 ±0.03	1.79 ±0.07	1.80 ±0.16	1.86 ±0.02	1.83 ±0.03	2.18 ±0.02	1.86 ±0.02	1.86 ±0.16	1.79 ±0.16	1.92 ±0.06	1.89 ±0.04	1.92 ±0.06	1.79± 0.11	1.92 ±0.16	1.71 ±0.06	1.83 ±0.04	1.67 ±0.11
Peanut	1.79± 0.00	1.83 ±0.07	1.89 ±0.07	1.86 ±0.02	1.81 ±0.03	1.94 ±0.02	1.82 ±0.06	1.84 ±0.07	1.77 ±0.03	1.84 ±0.02	1.79 ±0.16	1.81 ±0.03	1.83 ±0.06	1.79 ±0.03	1.79 ±0.23	1.81± 0.07	1.83± 0.16	1.80 ±0.02	1.79 ±0.31	1.96 ±0.28
Soybean	1.82± 0.00	1.96 ±0.03	2.00 ±0.04	1.85 ±0.16	1.90 ±0.02	1.89 ±0.24	1.92 ±0.23	1.78 ±0.04	1.81 ±0.11	1.24 ±0.07	1.84 ±0.11	1.86 ±0.06	2.03 ±0.49	1.85 ±0.08	1.89 ±0.07	1.94± 0.16	1.73 ±0.02	2.03 ±0.02	1.96 ±0.02	1.97 ±0.16
Sunflower	1.77± 0.00	1.83 ±0.06	1.85 ±0.28	1.84 ±0.07	1.90 ±0.16	2.03 ±0.02	1.95 ±0.16	1.89 ±0.28	2.03 ±0.28	1.90 ±0.02	1.95 ±0.24	1.77 ±0.06	1.97 ±0.11	1.85 ±0.04	1.92 ±0.02	1.78± 0.02	1.84± 0.23	1.92± 0.07	1.84 ±0.16	1.85 ±0.02

6hrs: Used for frying for 6 hrs.
SRT 5 days: Stored at 25^0C for 5 days.
2nd fry RT: Used for frying after 5 days of storage at room temp (Total frying time: 12 hrs).
R5 d: Stored at 4^0C for 5 days.
2nd fry R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs).

Table 4.9: Polymer content of oils after deep frying food (g).

	Food type					
Type of oil	**Chicken**	**Chips**	**Fish**	**Mandazi**	**Smokies**	**Mean**
Corn	1.86±0.0306	1.82±0.4110	1.96±0.1940	1.78±0.0989	1.88±0.0541	1.8587
Palm	1.94±0.0954	1.79±0.0403	1.90±0.0896	2.02±0.2988	1.96±0.0537	1.9212
Peanut	1.86±0.0300	1.86±0.0723	1.82±0.0404	1.85±0.0665	1.80±0.0179	1.8367
Soybean	1.94±0.0777	1.90±0.0153	1.61±0.3208	1.93±0.1146	1.89±0.0783	1.8540
Sunflower	1.84±0.0100	1.96±0.0656	1.94±0.0781	1.85±0.0498	1.89±0.1	1.8956
Mean	1.8873	1.8700	1.8440	1.8840	1.8840	1.8732

Table 4.10: Polymer content based on storage conditions (g).

	Type of oil					
Storage	**Corn**	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	1.75±0.0047	1.83±0.0124	1.79±0.00816	1.82±0.00125	1.77±0.0047	1.79
6 hrs	1.82±0.3198	1.84±0.0513	1.82±0.0195	1.88±0.0740	1.87±0.0660	1.85
Refri5days	1.86±0.03	1.89±0.0300	1.84±0.0150	1.73±0.2350	1.85±0.005	1.84
SRT 5days	1.90±0.1586	2.00±0.3357	1.82±0.0100	1.80±0.0650	1.80±0.0350	1.89
2nd fry ref	1.83±0.0852	1.92±0.1873	1.87±0.0531	1.77±0.2699	1.90±0.0400	1.89
2nd fryord	1.82±0.1485	2.29±0.3677	1.86±0.05713	1.95±0.0877	1.96±0.0687	1.98
Mean	1.85	1.98	1.84	1.83	1.86	1.87

Polymer contents of fresh oils ranged from 1.75-1.83g (table 4.8).Peanut contained lower amounts of polymer contents than the other oils when used to fry food. Its mean was 1.84 (table 4.9). Palm oil recorded relatively higher values of P.Cs with a mean of 1.9212 (table 4.9).In terms of food; the highest level was recorded in food type chicken (1.89) and the least in fish with a mean of 1.84 (table 4.9). Mandazi and smokies recorded the same mean of 1.88 (table 4.9). This indicates higher polymer formation while frying chicken than other types of food. This could be due to fats introduced to the oils by chicken. Polymer content was found to be lower in oils before than after frying food (table 4.8) which is in agreement with previous studies showing that amount of polymers increase with number of frying and frying time (Tompkins and Perkins, 2000). Choe and Min (2007) also reported that formation of

polymers is aggravated with increased frying temperature and total number of frying. These polymers cause higher oil absorption of foods (Tompkins and Perkins, 2000).

From Anova analysis, the model was significant ($p<0.05$). As can be appreciated, there was no significant difference in the main effects oil and food; while interaction effects showed significant difference ($p<0.05$) (appendix 4). There was an increase in polymer content when the oils were used for six hours. Polymer content increased further when the oils were used for the second time with the refrigerated oils showing lower values than the ones stored at room temperature (table 4.8).

From the results, peanut oil is less polymerized (table 4.9). According to Bastida and Sanchez-muniz (2007), the oil rich in linoleic acid is more easily polymerized during deep-fat frying than the oil rich in oleic acid. This is consistent with the findings in this study showing higher oleic acid than linoleic acid in all peanut oils. From the results it is noted that there is a correlation between polymer content and the density. The highest polymer content (2.55) was noted in palm oil that was stored at room temperature for five days then used to fry mandazi for another 6 hours to make a total of 12 hours (table 4.10). This particular sample was noted to have almost the highest density. This can be concluded that the increase in density is due to the polymers formed. Changes in palm oil density are more significant. This is expected, due to the higher polymer generation when frying with palm oil (Kalogianni *et al*., 2009; 2010). The higher the amount of polymers, the higher the density. Paul and Mittal (1996) linked polymers to the increase of oils density. During frying and heating, oxidation, polymerization, isomerization (in both frying and heating) and hydrolysis (only during frying) occur in the oil generating a multitude of products (Belitz *et al*., 2004). Among these products, higher molecular weight products compared to triacylglycerols are generated originating from polymerization and oxidation reactions (Dobarganes and Márquez-Ruiz, 1996; Kalogianni *et al*., 2009; 2010). From Anova results, there was significant difference ($p\leq0.05$) in the main effects and the model (Appendix 11).

4.3.4 Iodine value

Iodine value is used to measure unsaturation or the average number of double bonds in fats and oils. Decrease in iodine value shows decrease in the number of double bonds and it indicates oxidation of the oil.

Table 4.11: Iodine values of oils before and after frying food

Type of Oil	Fresh oil	Chicken			Chips			Fish			Smokies				Mandazis				
		6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr yR	6hrs	2nd fr RT	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5 d	2nd fr R
Palm	50.8 ±0.72	48.2 ±2.45	43.1 ±2.94	45.7 ±2.69	48.2± 3.18	45.1 ±3.36	45.1 ±2.29	45.7 ±2.20	40.6 ±2.37	43.1 4±1.22	48.2 ±1.63	43.1 ±0.82	40.6 ±1.63	45.7 ±1.22	43.1 ±1.39	50.8 ±1.47	50.8 ±1.80	48.2 ±1.22	48.2 ±0.82	45.7± 1.78
Corn	132.0 ±0.40	124.4±1.50	109.1±1.80	121.8±2.04	124.5 ±3.51	114.2±4.41	116.7±4.65	116.7±4.16	98.9 ±3.92	104.1±3.43	122.9 ±3.27	116.7±2.94	114.2±2.78	121.8±2.69	116.7±1.22	124.4 ±2.61	116.7 ±3.18	86.3 ±3.27	122.9 ±3.26	121.8 ±3.02
Peanut	104.1 ±3.32	99.0 ±2.69	96.4 ±3.18	99.0 ±2.44	101.5 ±1.80	93.9 ±2.20	99.0 ±2.37	98.9 ±2.45	86.3 ±2.61	96.4 ±2.78	101.5 ±1.88	96.4 ±1.91	86.3 ±1.87	98.3 ±2.04	96.4 ±1.39	96.4 ±2.45	88.8 ±1.22	83.8 ±1.63	93.9 ±1.88	88.8 ±1.91
Soy-bean	129.4 ±1.00	121.8±3.02	98.9 ±1.22	86.3 ±1.63	116.7 ±2.94	104.1±1.88	101.5±2.04	111.7±2.29	78.7 ±1.39	91.4 ±2.20	126.9 ±1.91	114.2±1.39	99.0 ±2.86	116.7±1.47	104.1±1.88	96.4± 2.12	91.4 ±1.91	86.3 ±2.94	93.9 ±2.04	88.8 ±1.47
Sunflower	126.9 ±1.67	124.4±1.91	119.3±2.12	121.8±3.18	124.4 ±2.78	119.3±2.86	121.8±2.45	121.8±1.47	116.7±2.86	119.2±1.91	124.4 ±3.02	121.8±1.63	119.3 ±1.22	124.4±1.80	121.8±1.46	121.8 ±2.20	116.7 ±1.91	114.2 ±1.88	119.2 ±2.69	116.1 ±2.86

6hrs: Used for frying for 6 hrs
SRT 5 days: Stored at 25^0C for 5 days
2nd fr RT: Used for frying after 5 days of storage at room temp (Total frying time: 12 hrs)
R 5 d: Stored at 4^0C for 5 days
2ndfrR: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs

Table 4.12: Mean iodine values of oils after deep frying food (g of iodine/100g of oil)

Type of oil	Food type					
	Chicken	**Chips**	**Fish**	**Mandazi**	**Smokies**	**Mean**
Corn	118.44±8.1	118.49±5.4	106.60±9.2	114.43±15.9	118.47±3.7	115.29
Palm	45.68±2.5	46.17±1.8	43.15±2.5	48.73±2.1	44.16±2.9	45.58
Peanut	98.14±1.5	98.14±3.8	93.91±8.9	90.35±4.9	95.80±5.7	95.27
Soybean	102.37±18.0	107.44±8.2	93.90±16.6	91.37±4.0	112.18±10.8	101.45
Sunflower	121.82±2.5	121.82±2.5	119.29±2.5	117.76±2.9	122.33±2.1	120.60
Mean	97.29	98.41	91.37	92.53	98.59	95.64

Table 4.12: Mean iodine values of oils based on storage conditions (g of iodine/100g of oil)

	Type of oil					
Storage	**Corn**	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	131.98±0.40	50.76±0.72	104.06±3.32	129.44±1.00	126.90±1.67	108.63
6 hrs	122.58±3.32	48.22±1.79	99.49±2.12	114.72±11.69	123.35±1.39	101.67
Refri 5days	122.36±0.54	46.95±1.27	96.10±2.19	105.33±11.42	121.82±2.54	98.51
SRT 5days	116.71±0.03	46.95±3.09	92.63±3.09	102.79±11.42	119.29±2.54	92.87
2nd fry ref	116.24±6.50	44.56±1.17	95.94±5.39	94.4136±7.07	120.30±2.03	94.29
2nd fry ord	100.56±10.70	43.5±2.89	89.33±4.92	93.40±9.42	117.77±2.03	88.91
Mean	118.40	46.01	96.26	102.92	120.81	97.48

Table 3 shows the recommended standards as follows (g of iodine/100g of oil): Palm 50-55, corn 103-135, soybean 120-143 and sunflower 110-143. After frying different types of food, palm oil was found to have relatively lower iodine value of 45.58 (table 4.12). The highest iodine value observed was in sunflower (120.60), corn (115.29) and soybean (101.45) oils (table 4.12). In terms of food, smokies and chips recorded relatively higher amounts of Iodine (98.59 and 98.41) respectively. The least value recorded was in food type fishes (91.37). The iodine values of oils before and after frying food were compared and out of this, it was found that there was decrease in iodine value in all the oils after frying food. This was in agreement with Reblova *et al*. (1999) who reported a decreasing trend in iodine value of the oil during deep-fat frying. The decrease in iodine value with time of frying could be attributed to the changes in fatty acids taking place with duration of frying (Tynek *et al*., 2001). The highest

decrease in iodine value was observed in soybean after frying the five types of food and the values were as follows chicken 27.00, chips 22.00, Fish 35.50, mandazi 38.10 and smokies 17.30 (Appendix 17). The least decrease was indicated by palm: chicken 5.10, chips 4.60, fish 7.60, mandazi 2.00 and smokies 6.60 (appendix 17). These values were measured in g of iodine/ 100g of oil. A decrease in iodine value is an indicator of lipid oxidation (Naz *et al*., 2004) and is consistent with the decrease in double bonds as oil becomes oxidized (Alireza *et al*., 2010). Soybean oil has high amounts of PUFAS (linoleic and linolenic) compared to palm oil which is rich in MUFAS and saturated fatty acids (table 4.1). Vegetable oils which are rich in PUFA are more prone and less stable to oxidation compared to those which are rich in MUFA (Kochhar and Henry, 2009). This explains why soybean was highly oxidized with respect to all types of food fried. There were significant differences in the main effects as well as interaction effects (Appendix 5).

Iodine values of fresh oils ranged from 50.76-129.90 (table 4.11). All the oils recorded a decrease in iodine values when subjected to different frying and storage conditions. The highest decrease was observed in the oils used for frying after storage for 5 days at room temperature. The oil samples used frying for 6 hours recorded a small decrease (table 4.11). According to Sharoba and Ramadhan 2012, the iodine value of the oils decreases versus frying time due to consumption of double bonds by oxidation and polymerization. Palm and peanut had relatively lower contents of iodine values as compared to soybean, corn and sunflower. The greater the degree of unsaturation (or high IV), the more rapid the oil tends to be oxidized, particularly during deep-fat frying (Alireza *et al*., 2010). From Anova results, the model showed significant difference $(p<0.05)$ as well as the main and interaction effects (Appendix 12).

4.3.5 *Para*-anisidine value.

Anisidine value (AV) determination is an empirical test for assessing advanced oxidative rancidity of oils and fats. It estimates the secondary oxidation products of unsaturated fatty acids, principally conjugated dienals and 2-alkenals. Aldehydes are largely considered responsible for the off-flavors in fats and oils due to their low sensory threshold values (Labrinea *et al*., 2001). During frying anisidine value increases indicating oxidation.

Table 4.13: Para- anisidine values of oils before and after deep frying food

Type of Oil	Fresh Oil	Chicken			Chips			Fish			Smokies				Mandazis				
		6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	SRT 5d	2nd fr RT	R5d	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R
Palm	18.0± 0.01	23.3 ±0.09	19.8± 0.10	29.0 ±0.11	34.3 ±0.08	20.5 ±0.07	47.5 ±0.11	17.9 ±0.49	33.5 ±0.06	37.6 ±0.12	34.0 ±0.07	27.8 ±0.11	39.1 ±0.04	28.2 ±0.16	39.7 ±0.12	24.4 ±0.13	26.0 ±0.13	41.5 ±0.10	24.4 ±0.11	35.9± 0.23
Corn	10.5± 0.03	18.5 ±0.13	58.5± 0.14	49.9 ±0.11	51.9 ±0.14	37.8 ±0.02	86.5 ±0.03	53.8 ±0.16	32.0 ±0.13	76.9 ±0.12	22.8 ±0.07	30.9 ±0.03	39.7 ±0.26	32.0 ±0.02	38.4 ±0.03	25.6 ±0.02	19.1 ±0.02	46.2 ±0.05	19.1 ±0.07	54.3 ±0.02
Peanut	5.2 ±0.02	17.9 ±0.03	19.8 ±0.06	23.3 ±0.04	45.9 ±0.11	29.0 ±0.07	33.7 ±0.10	13.5 ±0.11	45.1 ±0.09	55.9 ±0.11	41.1 ±0.07	43.9 ±0.16	34.4 ±0.28	32.9 ±0.07	38.5 ±0.03	30.0± 0.02	31.9 ±0.07	43.0 ±0.03	29.8± 0.12	46.6± 0.15
Soybean	7.7 ±0.02	27.9 ±0.16	37.7 ±0.03	75.0 ±0.02	27.9 ±0.16	61.5 ±0.07	60.1 ±0.02	46.5 ±0.04	26.9 ±0.06	36.9 ±0.16	ND	ND	ND	ND	ND	32.5 ±0.17	32.9± 0.02	49.5 ±0.14	33.7± 0.11	59.7± 0.13
Sunflower	12.3 ±0.22	35.3 ±0.10	55.1 ±0.23	59.7 ±0.49	30.9 ±0.28	60.1 ±0.15	54.4 ±0.17	18.6 ±0.06	60.5 ±0.07	53.4 ±0.16	23.3 ±0.16	35.0 ±0.11	42.5 ±0.10	27.9 ±0.10	28.7 ±0.28	46.4 ±0.07	59.4 ±0.16	60.5 ±0.02	67.1 ±0.03	75.9 ±0.02

2nd fr RT: Used for frying for 6 hrs after 5 days of storage at room temp. **6hrs:** used for frying for 6 hrs
2nd fr R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs**).** **R 5d:** Stored at 4^0C for 6 hrs
SRT 5d: Stored at 25^0C for 5 days

Table 4.14: Para-anisidine values of oil based on food fried

Type of oil	Food type					
	Chicken	**Chips**	**Fish**	**Mandazi**	**Smokies**	**Mean**
Corn	42.31±21.1	58.71±25.1	57.59±23.1	32.83±16.3	32.76±6.7	44.84
Palm	24.04±4.6	34.11±13.5	29.66±10.4	30.47±7.8	33.78±5.7	30.41
Peanut	20.33±2.7	36.22±8.7	41.50±19.8	36.28±7.9	38.15±4.6	35.83
Soybean	47.17±25.4	49.84±15.9	36.77±13.8	41.67±12.3	ND	43.86
Sunflower	50.03±10.6	48.47±12.6	39.55±12.8	61.86±10.9	31.51±7.4	51.87
Mean	37.45	49.46	45.27	40.62	24.05	41.36

Table 4.15: Para-anisidine values of oils based on storage conditions

Storage	Type of oil					
	Corn	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	10.52±0.03266	18.02±0.0125	5.18±0.02445	7.68±0.0163	12.27±0.2204	10.73
6 hrs	34.42±15.1483	26.78±7.1859	29.67±14.1052	26.96±8.7826	30.90±10.8206	29.46
Refri 5 day	25.55±6.4500	26.30±1.9000	31.35±1.5500	29.80±0.0000	47.5±19.6000	32.10
SRT 5days	25.00±5.9000	26.90±0.9000	37.90±6.0000	32.00±0.0000	47.20±12.2000	33.80
2nd fry ref	61.20±17.7860	37.94±5.9782	39.60±11.1032	57.93±13.6151	54.42±15.1778	50.22
2nd fry ord	42.90±9.1500	30.88±9.1403	34.24±9.2576	43.90±12.9282	60.32±13.2480	42.45
Mean	37.47	30.30	25.77	33.80	44.58	32.99

Para- anisidine values for fresh oils ranged from 5.18-18.03 (table 4.14). According to Gupta 2005, a desirable *p*-AV for fresh frying oil is less than 4.0, with an upper limit of 6.00. He further stated that if fresh frying oil had a *p*-Av of above 6.00, it would be highly oxidized. From the results, only peanut oil had a *p*-AV of below 6.00. This implies that the other types of oil are susceptible to oxidation. It shows high oxidation levels above the recommended levels. *p*- AV is the most accurate and reliable method for oxidative state of the oils (Van der Merwe *et al*., 2003). This means all the oils did not meet the acceptable oxidation status. *Para*-Anisidine values were relatively higher in sunflower oil (51.87) and least in palm (30.41) (table 4.15). Palm oil has an abundant content of vitamin E, which may play an important role in its ability to withstand thermal oxidative changes (Quiles *et al*., 1999). In terms of food, relatively higher and lower values were recorded in chips (49.46) and smokies

(27.24) (table 4.15) respectively. The high *p*-AV in oils in which chips were fried could be due to the high water content of potatoes. During frying this water is removed and mixes with the frying oils producing secondary products. A rapid increase in *p*-Av was noted for all the oils after frying food. The highest increase was noted in soybean oil with respect to all types of food: chicken 40.00, chips 52.20, fish 29.10 and Mandazi 34.00 (appendix 17). This is in agreement with previous researchers who found out that as the heating continues p-AV of heated oils increases due to decomposition of oil hydro peroxides (Abdulkarim *et al.*, 2007). Contact with air (oxygen) during frying, and unsaturation level in fatty acids are the factors that affect the p-AV level (Khan *et al*., 2011).

ANOVA showed that the main effects were significant ($p<0.05$) as well as the interaction between the type of food and the oil. This showed that the quality of oil was affected by the food fried (Appendix 6).From this study, it was found that *p*-AV values increased with frying time which is in agreement with the findings of Tarmizi *et al.* (2013) who concluded that the amount of *p*-anisidine gradually increased with time of frying. It is a fact indicating that in the first hours of use, the oxidation reaction is already occurring and primary products (peroxides, hydro peroxides, conjugated dienes, hydroxides and ketones) are formed and degraded, thus producing secondary products (epoxides, compounds volatile and non-volatile) obtained by cleavage and rearrangement of peroxides (Gomes *et al*., 2003).

Para-anisidine values increased in all the samples when subjected to frying and different storage conditions. The least increase was noted in the oil that was used for frying for 6 hrs (30.16) and the highest (46.64) was observed in the oils that were refrigerated at 4^0C for five days (table 4.16). When this oil was refrigerated for 5 days, relatively higher increase was observed (table 4.15). In terms of fresh oil, peanut oil had the lowest value. Soybean oil also had a relatively lower value. Fresh palm oil had the highest *p*-AV value of 18.02 (table 4.14); however it recorded the least increase as compared to other oils. *Para* anisidine values of the oils proved to be affected by the storage conditions and frying time. Anova showed that there was significant difference ($p<0.05$) in the main effect; storage. The interaction effects were however not significant (Appendix 13).

4.3.6 Refractive index

The refractive index is the degree of the deflection of a beam of light that occurs when it passes from one transparent medium to the other. It increases with the length of chains and with the number of carbon atoms present (Barkatullah *et al*., 2012. Refractive index increases with oxidation. This could be due to the polymers formed and increase in saturation.

Table 4.16: Refractive indices of oils based on storage conditions

Type of Oil	Fresh Oil	Chicken			Chips			Fish			Smokies				Mandazis				
		6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	2nd fr RT	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R	6hrs	SRT 5d	2nd fr RT	R 5d	2nd fr R
Palm	1.49 ±0.00	1.49 ±0.02	1.49 ±0.08	1.49 ±0.28	1.49 ±0.31	1.49 ±0.02	1.49 ±0.03	1.49 ±0.07	1.49 ±0.16	1.4± 0.02	1.49 ±0.02	1.49± 0.07	1.49 ±0.07	1.49 ±0.49	1.49 ±0.31	1.49 ±0.28	1.49 ±0.02	1.49 ±0.03	1.49 ±0.06	1.49± 0.02
Corn	1.50± 0.00	1.50 ±0.50	1.50 ±0.02	1.50 ±0.23	1.50 ±0.07	1.50 ±0.16	1.50 ±0.02	1.50 ±0.03	1.50 ±0.02	1.50 ±0.16	1.50 ±0.11	1.50 ±0.23	1.50 ±0.07	1.50 ±0.16	1.50 ±0.12	1.50 ±0.11	1.50 ±0.13	1.50 ±0.14	1.50 ±0.15	1.50 ±0.16
Peanut	1.49± 0.00	1.49 ±0.28	1.50 ±0.03	1.49 ±0.23	1.49 ±0.03	1.49 ±0.31	1.49 ±0.02	1.49 ±0.09	1.49 ±0.10	1.49 ±0.11	1.49± 0.09	1.49± 0.11	1.50 ±0.28	1.49 ±0.07	1.50 ±0.23	1.49± 0.06	1.50 ±0.23	1.50 ±0.07	1.50 ±0.16	1.49± 0.02
Soybean	1.50 ±0.00	1.50 ±0.15	1.50 ±0.16	1.50 ±0.17	1.50 ±0.03	1.50 ±0.07	1.50 ±0.09	1.50 ±0.07	1.50 ±0.06	1.50 ±0.02	ND	ND	ND	ND	ND	1.50 ±0.03	1.50 ±0.02	1.50 ±0.03	1.50± 0.07	1.50 ±0.16
Sunflower	1.49± 0.00	1.50 ±0.31	1.50 ±0.11	1.50 ±0.12	1.49 ±0.13	1.50 ±0.09	1.50 ±0.16	1.50 ±0.11	1.50 ±0.06	1.49 ±0.16	1.50 ±0.11	1.50 ±0.09	1.50 ±0.13	1.50 ±0.12	1.50 ±0.14	1.49 ±0.15	1.49± 0.17	1.49 ±0.16	1.50± 0.12	1.50 ±0.10

6 hrs: used for frying for 6 hrs.
R 5d: Stored at 4^0C for 6 hrs.
SRT 5d: Stored at 25^0C for 5 days
2nd fr RT: Used for frying for 6 hrs after 5 days of storage at room temp.
2nd fr R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs**).**

Table 4.17: Refractive Index values of oils after deep frying food

Type of oil	Food type					
	Chicken	**Chips**	**Fish**	**Mandazi**	**Smokies**	**Mean**
Corn	1.4973±0.0010	1.4975±0.0011	1.4963±0.0004	1.4967±0.0014	1.4981±0.0013	1.4972
Palm	1.4884±0.0002	1.4891±0.0008	1.4880±0.0002	1.4887±0.0013	1.4894±0.0014	1.4887
Peanut	1.4947±0.0011	1.4925±0.0009	1.4935±0.0010	1.4943±0.0013	1.4942±0.0014	1.4938
Soybean	1.4972±0.0004	1.4978±0.0004	1.4979±0.0009	1.4987±0.0009	ND	1.1983
Sunflower	1.4993±0.0010	1.4993±0.0010	1.4986±0.0024	1.4983±0.004	1.4985±0.0012	1.4988
Mean	1.4954	1.4953	1.4949	1.4953	1.4950	1.4354

Table 4.18: Refractive Indices of oils based on storage conditions

	Type of oil					
Storage	**Corn**	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	1.4959±0.0001	1.4881±0.0001	1.4927±0.0001	1.4968±0.0001	1.4971±0.0001	1.49
6 hrs	1.4964±0.0010	1.4881±0.0004	1.4931±0.0008	1.4979±0.0008	1.4986±0.0011	1.49
Refri 5days	1.4973±0.0011	1.4882±0.0004	1.4932±0.0007	1.4978±0.0008	1.4988±0.017	1.49
SRT 5days	1.4969±0.0012	1.4889±0.0011	1.4941±0.0014	1.4979±0.0010	1.4988±0.0013	1.49
2nd fry ref	1.4991±0.0010	1.4904±0.0006	1.4956±0.0005	1.4990±0.0000	1.4987±0.0008	1.5
2nd fry ord	1.4981±0.0002	1.4902±0.0001	1.4954±0.0001	1.4991±0.0010	1.4988±0.0008	1.5
Mean	1.5	1.49	1.49	1.49	1.5	1.49

The refractive index of oils depends on their molecular weight, fatty acid chain length, degree of unsaturation, and degree of conjugation. Values of refractive index for different oils generally vary between 1.449 and 1.470 (table 4.2). Refractive indices of fresh oils were as follows: palm 1.4881, corn 1.4959, peanut 1.492, soybean 1.4968 and sunflower 1.4971 (table 4.17). The refractive indices of fresh oils were already above the specified range. All the oils recorded higher refractive index after frying food. Palm oil had relatively lower mean value (1.4887) and higher mean value in sunflower (1.4988) after frying food (table 4.18). In terms of food, fish oil showed relatively lower values with a mean of 1.4949 and higher values were observed in chicken (mean 1.4954). This means that the unsaturation of oils decreased after frying chicken due to oxidation. This could be due to chicken fried products.

According to Aniołowska and Kita (2015), RI is related to the fat autoxidation reaction, and its value increases after the formation of peroxides. From appendix 7, significant differences were observed in the main effects as well as interaction effects. This shows that refractive index of the oils is affected by the type of food fried. From table 4.19, it was noted that refractive indices increased. A similar trend was observed by Ali *et al*., (2013), they noted that refractive index increases with frying time. RI increases with an increase in polymerization, molecular cohesiveness among the components of increased chain length, saturation of carbon-carbon double bonds, moisture in food and opaqueness and turbidity (Kress Rogers *et al*., 1990). From Anova results; there was significant difference in the main effect oil. It is evident from the statistical results that no significant effect ($p \leq 0.05$) was observed on refractive index of oils based on the storage conditions (Appendix 14).

4.3.7 Density

Density is the ratio of mass to volume of the oil. Density increases with the increase in oxidation of vegetable oils. This could be due to higher molecular weight compounds formed during frying. Very small food residues could also increase the density of the oils.

Table 4.19: Density of oils before and after frying food (g/cm3)

Type of Oil	Fresh Oil	Chicken			Chips			Fish			Smokies				Mandazis				
		6hrs	2nd fry RT	2nd fry R	6hrs	2nd fry RT	2nd fry R	6hrs	2nd fry R	2nd fry R	6hrs	SRT5 days	2nd fry RT	R5d	2nd fry R	6hrs	SRT5d	2nd fryRT	R 5d	2nd fry R
Palm	0.893 ±0.01	0.936 ±0.006	0.982 ±0.006	0.960 ±0.001	0.908 ±0.004	0.974 ±0.001	0.951 ±0.002	0.925 ±0.001	0.936 ±0.006	0.943 ±0.002	0.901 ±0.001	0.909 ±0.006	0.914 ±0.006	0.918± 0.001	0.894 ±0.004	0.912± 0.004	0.901± 0.002	0.964± 0.001	0.939± 0.002	0.913± 0.002
Corn	0.896 ±0.01	0.883 ±0.002	0.958 ±0.003	0.942 ±0.004	0.877 ±0.005	0.960 ±0.005	0.878 ±0.006	0.913 ±0.001	0.95± 0.006	0.879 ±0.003	0.893 ±0.002	0.973 ±0.002	0.924 ±0.003	0.931± 0.003	0.913 ±0.002	0.890± 0.004	0.870± 0.006	0.882± 0.004	0.919± 0.003	0.923± 0.002
Peanut	0.929 ±0.01	0.958 ±0.005	0.974 ±0.002	0.966 ±0.006	0.936 ±0.001	0.927 ±0.002	0.936 ±0.003	0.879 ±0.003	0.921 ±0.002	0.936 ±0.006	0.945 ±0.002	0.950 ±0.002	0.953 ±0.003	0.950± 0.003	0.940 ±0.004	0.939± 0.004	0.939± 0.005	0.952± 0.005	0.945± 0.001	0.968± 0.002
Soybean	0.917 ±0.01	0.943 ±0.004	0.94± 0.001	0.948 ±0.001	0.943 ±0.002	0.945 ±0.003	0.948 ±0.004	0.930 ±0.004	0.951 ±0.003	0.948 ±0.006	ND	ND	ND	ND	0.948 ±0.003	0.948± 0.002	0.946± 0.003	0.949± 0.004	0.950± 0.005	0.946± 0.006
Sunflower	0.909 ±0.01	0.937 ±0.002	0.945 ±0.002	0.927 ±0.002	0.918 ±0034	0.939 ±0.004	0.946 ±0.003	0.966 ±0.006	0.937 ±0.004	0.951 ±0.001	0.912± 0.006	0.951 ±0.002	0.921 ±0.001	0.952± 0.003	0.925 ±0.003	0.932± 0.004	0.959± 0.005	0.964± 0.004	0.945± 0.006	0.953± 0.001

2nd fry RT: Used after 5 days of storage at room temp. **6hrs:** Used for frying for 6 hrs

2nd fry R: Used for frying after 5 days of storage at 4^0C (Total frying time: 12 hrs) **SRT 5 days:** Stored at room temp for 5 days

R 5d: Stored at 4^0C for 5 days

Table 4.20: Density values of oils after deep frying food (g/cm3)

Type of oil	Food type					
	Chicken	**Chips**	**Fishes**	**Mandazi**	**Smokies**	**Mean**
Corn	0.9277±0.01	0.9050±0.11	0.9140±0.01	0.8968±0.01	0.9268±0.01	0.9141
Palm	0.9593±0.01	0.9443±0.01	0.9347±0.01	0.9258±0.01	0.9072±0.01	0.9343
Peanut	0.9660±0.01	0.9330±0.01	0.9120±0.01	0.9486±0.01	0.9476±0.01	0.9414
Soybean	0.9463±0.01	0.9453±0.01	0.9430±0.01	0.9478±0.01	0.9618±0.01	0.9488
Sunflower	0.9363±0.01	0.9343±0.01	0.9513±0.01	0.9506±0.01	0.9322±0.01	0.9409
Mean	0.9471	0.9324	0.931	0.9339	0.7807	0.9050

Table 4.21: Density values of oils based on storage conditions

	Type of oil					
Storage	**Corn**	**Palm**	**Peanut**	**Soybean**	**Sunflower**	**Mean**
Fresh oil	0.9±0.01	0.89±0.01	0.93±0.01	0.92±0.01	0.91±0.01	0.91
6 hrs	0.89±0.012	0.92±0.012	0.93±0.027	0.75±0.007	0.93±0.019	0.88
Refri 5days	0.93±0.000	0.93±0.004	0.95±0.002	0.76±0.000	0.95±0.003	0.9
SRT 5days	0.92±0.052	0.91±0.004	0.94±0.005	0.76±0.000	0.96±0.004	0.91
2nd fry ref	0.91±0.025	0.93±0.024	0.95±0.015	0.95±0.000	0.88±0.118	0.93
2nd fry ord	0.94±0.029	0.95±0.025	0.95±0.019	0.95±0.002	0.94±0.013	0.84
Mean	0.91	0.93	0.94	0.83	0.93	0.9

The density of liquid oils is in the range of 0.8910–0.9250 as recommended by FAO/WHO (table 4.1). The densities of palm; soybean and sunflower oils were within the range specified. However peanut oil showed a relatively higher density before use. The densities of fresh oils (table 4.20) were compared with the density values of oils after deep frying. From table 4.21, it is noted that the highest and lowest means were with respect to chicken (0.9471) and smokies (0.7807) respectively. This could be due to chicken fried products. Chicken has a lot of fats and also introduces a lot of water to the frying oil leading to high saturation. From previous studies, it is noted that density decreases with an increase in unsaturation and increases with high saturation and polymerization (Kim *et al.*, 2010). Palm and sunflower oils used for frying fish also indicated relatively higher increase in densities (table 4.21). Fish has natural oil which blends with the frying oil. This increases the density of oils. After deep frying food, soybean oil indicated high density values across the five types of food resulting

in the highest mean of 0.9488 compared to other oils (table 4.21). This means that soybean oil becomes highly saturated when used to fry food.

There were significant differences ($p<0.05$) in the main effects as well as the interaction effects. This showed that the type of food fried affected the quality of the oil (Appendix 8)
The samples had close density values. Generally all the oils recorded a notable increase in density after frying. The smallest increase was noted in the oils that were used for frying for 6 hrs while the highest noted in the oils that were stored at room temperature then used for frying for 6hrs (table 4.22). There was increase in density in oils as frying time increases, Paul and Mittal (1996) linked polymers to the increase of oils density. During frying and heating, oxidation, polymerization, isomerization (in both frying and heating) and hydrolysis (only during frying) occur in the oil generating a multitude of products (Belitz *et al*., 2004). Among these products, higher molecular weight products compared to triacylglycerols are generated originating from polymerization and oxidation reactions (Dobarganes and Márquez-Ruiz, 1996; Kalogianni *et al*., 2009; 2010). No significant difference was observed in the model, the main effects as well as the interaction effects (Appendix 15).

4.4 Relationship between oil types and the quality indicators.

For measure of relative oil stability, a univariate ANOVA test was run to find out if there were significant difference between the oils based on the oil quality indicators. Where significant differences were proven, additional regression analysis was carried out so as to ascertain the relative concentration of each measure in each oil type. This was to help with ranking the different types of oils from the one with the least concentration to the one with the highest concentration.

Table 4.22: ANOVA Tests to Assess differences in the oil quality parameters

Variable	Source	Partial SS	df	MS	F	Prob> F	R-squared
Peroxide value	Model	68.2324	4	17.0581	12.84	0	0.3509
	Residual	126.221	95	1.32865			
	Total	194.454	99	1.96418			
Saponification values	Model	3742.69	4	935.672	1.46	0.2203	0.0579
	Residual	60856.3	95	640.592			
	Total	64598.9	99	652.515			
Polymer content	Model	0.11557	4	0.02889	1.94	0.1104	0.0754
	Residual	1.41623	95	0.01491			
	Total	1.5318	99	0.01547			
Iodine value	Model	71532.5	4	17883.1	242.32	0	0.9107
	Residual	7011	95	73.8			
	Total	78543.5	99	793.369			
Para-anisidine	Model	4157.95	4	1039.49	3.32	0.0136	0.1227
	Residual	29719.1	95	1039.49			
	Total	33877.1	99	312.833			
Refractive index	Model	0.00133	4	0.00033	243.04	0	0.911
	Residual	0.00013	95	1.37E-06			
	Total	0.00147	99	1.5E-05			
Density	Model	0.01617	4	0.00404	7.74	0	0.2458
	Residual	0.04963	95	0.00052			
	Total	0.0658	99	0.00066			

From the results obtained in table 4.23, Peroxide value was found to be significantly affected by the type of oil [F (4, 95) = 12.84, p < 0.001), the model explained 35.09 of the total variation in Peroxide value. Likewise Iodine value [F (4, 95) = 242.32, p < 0.001) had a significant relationship with oil type; it was found that the type of oil determined the Iodine value showing 91.07% of the total variation. Concerning *para*-anisidine, significance differences between oils was found [F (4, 95) = 3.32, p <0.05). Another significant relationship was found in regard to Refractive index [F (4, 95) = 243.04, p < 0.001), explaining 91.1% of the total variation. Finally, Density [F (4, 95) = 7.74, p < 0.001) also showed significant variation depending on the oil type, which accounted for 24.58% of the total variation. This means that the indicators that are of major interest are peroxide value,

iodine value, *para* anisidine, refractive Index and density since they all showed significant relationship with the type of oil.

4.5 Ranking of oils based on the oil quality parameters

Since, the ANOVA tests showed that certain oil parameters were determined by the type of oil, an additional procedure aimed at finding out which oils are significantly different from one another was done.

Table 4.23: Regression coefficients depicting oil differences

		Coef.	Std. Err.	T	P>|t|	[95% Conf. Interval]	
Peroxide value	_cons	3.32	0.25774	12.88	0	2.80831	3.83169
	Palm	2.335	0.36451	6.41	0	1.61136	3.05864
	Corn	0.26	0.36451	0.71	0.477	-0.4636	0.98364
	Peanut	1.23	0.36451	3.37	0.001	0.50636	1.95364
	Soybean	1.17	0.36451	3.21	0.002	0.44636	1.89364
	Sunflower	(dropped)					
Iodine value	_cons	120.809	1.92094	62.89	0	116.995	124.622
	Palm	-74.798	2.71662	-27.53	0	-80.192	-69.405
	Corn	-4.457	2.71662	-1.64	0.104	-9.8502	0.93616
	Peanut	-25.541	2.71662	-9.4	0	-30.935	-20.148
	Soybean	-17.893	2.71662	-6.59	0	-23.286	-12.5
	Sunflower	(dropped)					
Para-Anisidine	_cons	0.51675	0.2138	2.42	0.018	0.0923	0.94119
	Palm	-1.0117	0.30236	-3.35	0.001	-1.612	-0.4115
	Corn	-0.4398	0.30236	-1.45	0.149	-1.04	0.16049
	Peanut	-0.772	0.30236	-2.55	0.012	-1.3722	-0.1717
	Soybean	-0.3603	0.30236	-1.19	0.236	-0.9606	0.23996
	Sunflower	(dropped)					
Refractive index	_cons	1.49855	0.00026	5719.09	0	1.49803	1.49907
	Palm	-0.0098	0.00037	-26.31	0	-0.0105	-0.009
	Corn	-0.0014	0.00037	-3.78	0	-0.0021	-0.0007
	Peanut	-0.0047	0.00037	-12.68	0	-0.0054	-0.004
	Soybean	-0.0004	0.00037	-0.94	0.347	-0.0011	0.00039
	Sunflower	(dropped)					
Density	_cons	0.93945	0.00511	183.82	0	0.9293	0.9496
	Palm	-0.0108	0.00723	-1.49	0.138	-0.0251	0.00355
	Corn	-0.0268	0.00723	-3.7	0	-0.0411	-0.0124
	Peanut	0.0027	0.00723	0.37	0.71	-0.0116	0.01705
	Soybean	0.00975	0.00723	1.35	0.181	-0.0046	0.0241
	Sunflower	(dropped)					

Consequently, from table 4.24 it is clear that in terms of peroxide value, there was significant difference between sunflower and Palm ($\beta = 2.335$, $p < 0.001$), Peanut ($\beta = 1.23$, $p = 0.001$), and soybean ($\beta = 1.17$, $p < 0.01$), but not with Corn ($\beta = 0.26$, $p < 0.477$). Therefore it can be concluded that palm had the highest level of peroxide value, followed by peanut and then soybean respectively while the levels were least in sunflower as well as in corn which were not significantly different from one another. In regard to Iodine value, differences were found with palm ($\beta = -74.798$, $p < 0.001$), peanut ($\beta = -25.541$, $p < 0.001$), and Soybean ($\beta = -17.893$, $p < 0.001$), no significant difference with Corn ($\beta = -4.457$, $p = 0.104$).

Thus in ranking the oils in terms of iodine value, least contents were found in palm, peanut, and soybean, respectively in ascending order. However, no significant difference was found between sunflower and corn. At the same time, in regard to para-anisidine, results showed that sunflower differed significantly from palm ($\beta = -1.0117$, $p = 0.001$), did not differ significantly from Corn ($\beta = -0.4398$, $p = 0.149$), but differed significantly from peanut ($\beta = -0.772$, $p < 0.05$), and yet did not differ significantly from soybean ($\beta = -0.3603$, $p = 0.236$).

The results in this section showed that the oxidative stability of vegetable oils is affected by food fried and storage conditions.

CHAPTER FIVE
CONCLUSIONS AND RECOMMENDATIONS

5.1 Introduction

This chapter summarizes the key findings, draws conclusions from those findings, provides recommendations based on those findings, and finally suggests areas in need of research efforts.

5.2 Conclusions.

The study concluded that lipids were largely evenly distributed among the oils studied except Soybean which had significantly higher linoleic and and linolenic but least oleic contents. Palm oil on the other hand had low levels of linoleic and linolenic fatty acids. Corn also had significantly higher levels of linolenic.

From the findings in this study, it was concluded that the best oil for deep frying chicken is palm followed by sunflower. For chips, the best oils were palm and sunflower. Sunflower oil proved to be the best for mandazi. Peanut and palm also recorded less oxidative changes with mandazi. Palm and sunflower were observed to be more stable than others while frying fishes. In smokies, three oils showed stability. These were palm, sunflower and peanut. In all the treatments done, soybean oil proved to be less resistant to oxidation. This is because of the high amounts of PUFAS in soybean oil noted in this study. Overall, the best oil for deep frying is palm since it is more resistant to oxidation.

In terms of storage conditions, it was observed that storage conditions had significant effects on the oil. This finding demonstrates that there is need to refrigerate oils after use. This is because oils refrigerated were more stable than the ones stored at room temperature. However, there was a disparity in *p*-AV and P.V. The oil samples that were refrigerated had relatively higher *p*-anisidine and peroxide values than the ones stored at room temperature. This could be due to the difference in rate of formation of peroxides during induction period. In the refrigerated oils, formation of peroxides and hydro peroxides is slow during the induction period. These compounds increase in concentration with time. On the other hand, peroxides and hydro peroxides in the oils stored at room temperature are formed rapidly and then decompose with time. By the time analysis is done, the refrigerated oils indicate higher P.V and *p*-AV than the ones stored at room temperature. The study concludes that oils in

which chicken and chips were fried contained more and less degradation products respectively. In regard to oxidative stability of oils after deep frying in food different types of food, the oil quality parameters that were of major interest are peroxide value, iodine value, *para* anisidine, refractive index and density since they all showed significant relationship with the type of oil.

5.3 Recommendations.

Based on the data from this study, the following recommendations were made:

- Research needs to be done on the oxidative changes that take place when different types of food are fried using the same type of oil.
- Further research on oxidative stability of vegetable and animal fats after deep frying food needs to be done.
- There is need for further research to establish the rancidity levels of each type of oil when used to fry particular food.
- There is need to do further research to assess the oxidative stability of the frying oils used in restaurants in Kenya.
- Further research needs to be done on the food fried to assess the compounds absorbed.

REFERENCES

Abdulkarim S.M., Long K., Lai O.M., Muhammad S.K.S. and Ghazali H.M., (2007). Frying quality and stability of high-oleic *Moringa oleifera* seed oil in comparison with other

Adam, S. K., Sulaiman, N. A., Top A.G.M., Jaarin, K. (2007). Heating reduces vitamin E content in palm and soy oils. *Malaysia J Biochemistry and Molecular Biology* **15**(**2**):76–79.

Ahmad Tarmizi, A. H., Niranjan, K., and Gordon, M. (2013). Physicochemical changes occuring in oil when atmospheric frying is combined with post-frying vacuum application. *Food Chemistry journal.* **136**: 902-908.

Ali M.A., Najmaldien A.H.A., Latip R.A., Othman N.H., Majid F.A.A. and Salleh L.M., (2013). Effect of heating at frying temperature on the quality characteristics of regular and high-oleic acid sunflower oils. *Acta Science. Pol., Technology. Aliment*.**12**:159-167.

Alireza S., Tan, C. P., Hamed M. and Che Man, Y.B. (2010). Effect of Vegetable-Based Oil Blends on Physicochemical Properties of Oils during Deep-Fat Frying. *American Journal of Food Technology,* **5***:* 310-323.

Alireza, S., Tan, C. P., Hamed, M. and Che Man, Y. B. (2010).Effect of frying process on fatty acid composition and iodine value of selected vegetable oils and their blends. *International Food Research Journal.* **17***:* 295-302.

Al-Kahtani, H. A. (1991). Survey of quality of used frying oils from restaurants. *Journal of the American Oil Chemists Society* **68**: 857–862.

Altar, Ted. (2006)"More Than You Wanted To Know About Fats/Oils". *Sundance Natural Foods.* Retrieved 2006-08-31.

Aniołowska, M and Kita, A. (2015). The effect of type of oil and degree of degradation on glycidylgsters content during the frying of french fries. *Journal of America Chemists society* **92**:1621-1631

Antolovich, M., Prenzler, P.D., Patsalides, E., McDonald, S. and Robards, K. (2002).Methods for testing antioxidant activity Analyst **127***:* 183-198.

Anwaar, A., Nisar, A. M., Muhammad, A. R., Saeed A., Asif A., Hajra, A.. and Syed S.S (2012) . Changes in Vegetable Oil Used for Commercial Frying: *Journal of chemical society* Pak **34**(5):107

AOAC, (1984). Official methods of analysis, 14th Edn. Association of Official Analytical Chemists,Washington D.C. USA. Pp. 120-127.

AOCS Official Method Cd 18-90, (2011). Determination of para anisidine value

AOCS, (1990). Official Methods and Recommended Practices of the American Oil Chemists' Society.4thed., AOCS Press, Champaign, Illinopp.50-54.

AOCS, (1991). Iodine Value of Fats and Oils, Wijs Method. Official Method Cd 1–25.

AOCS, (2014). Lubrizol standard test procedure AATM-51601

ASTM, D5558-95 (2011). Standard Test Method for Determination of the Saponification Value of Fats and Oils, ASTM International, West Conshohocken.

Ball D.W. (2012). Introduction to Chemistry: General, Organic, and Biological Chapter 17

Bardhan, J., Chakraborty, R., Raychaudhuri, U., (2011). The 21st century form of vitamin E - Tocotrienol. *Current Pharmaceutical Design*. **17**(**21**):2196–2205.

Barkatullah, M. I., Abdur, R. and Inyat-Ur-R. (2012). Physicochemical Characterization of Essential and Fixed Oils of *Skimmialaureola* and *Zanthoxylumarmatum Middle-East J. Med. Pl. Res.*,**1(3)**: 51-58

Bastida, S. and Sanchez-Muniz F.J. (2001). Thermal oxidation of olive oil, sunflower oil and a mix of both oils during forty discontinuous domestic fryings of different foods. *Food Science Technology.***7**15–21.

Bazlul S., Anees A.,MohamadH.I., Sufia H., Mohd R., and Mohd, Omar A. K. (2010) Physico-chemical properties of blends of palm olein with other vegetable oils pp 423-429

Belitz, H. D., Grosch, W.and Schieberle, P., (2004). Food Chemistry, 3rd ed. Springer Verlag, Berlin. pp. 218–219.

Bennion, M.(1995). Introductory Foods.10th Edition. Prentice-Hall Inc., Upper Saddle River, New Jersey, USA. Pp. 165-166.

Berezin, I. V., Denisov, E. T. (1996). *The Oxidation of Cyclohexane*, Pergamon Press, New York.

Berquin, I. M., Younong, M., Ruping, W., Jiansheng, W., Donna P., MarkCline J., Mike J. T., Thornburg. T., George K., Adrienne. S., Iris, J. E., Ralph, D. A., Zhang, H., Hong, W., Jing, X. K. and Yong, Q. C. (2007). Modulation of prostate cancer genetic risk by omega-3 and omega-6 fatty acids".*The Journal of Clinical Investigation.***117**: 1866–1875.

Besˇter, E., Butinar, B., Bucar-Miklavcic, M. and Golob, T. (2008). Chemical changes in extra virgin olive oils from Slovenian Istra after thermal treatment. *Food Chemistry* **108**:446–454. .

Blumenthal M.M, (1991).A new look at the chemistry and physics of deep-fat frying. *Food Technology*.**45**: 68–71.

Boskou, G., Salta, F., Chiou, A., Troullidou, E., and Andrikopoulos N.K. (2006). Content of trans, trans-2, 4-decadienal in deep-fried and pan-fried potatoes. *European Journal of Lipid Science and Technology.* **108**:109-115.

Brown, A. (2011). Understanding Food: Principles and Preparation 4th edition pp.471-473.

Che Man, Y., Ammawath, W., Rahman, R. and Yusof, S. (2003). Quality characteristics of refined, bleached and deodorized palm olein and banana chips after deep-fat frying. *Journal of Science and Food Agriculture*.**83**: 395-401.

Choe, E. and Min, D. B.(2006) . Mechanisms and Factors for Edible Oil Oxidation

Choe, E. and Min, D. B. (2007).Chemistry of deep-fat frying oils. *Journal of Food Science*.**72**: 77–86.

Codex Alimentarius Commission, Joint FAO/WHO Food Standards Programme (1993), Report of the fourteenth session of the codex committee on fats and oils, London, United Kingdom, Rome, Italy.

comprehensive reviews in Food science and Food safety **5**:169-186

Dobarganes, M. C.and Marquez-Ruiz, G. (2006). Formation and analysis of oxidized monomeric, dimeric and higher oligomeric triglycerides. In: Erickson, M.D. (Ed.) *Deep Frying; Chemistry, Nutrition and Practical Applications.* AOCS Press, Urbana, IL, pp. 87-110.

Dobarganes, M.C and Velasco, J., (2002). Analysis of lipid hydroperoxides. *European Journal of Lipid Science and Technology*.**104**: 420-428.

Dobarganes, M.C. and Pérez-Camino, M.C. (1987).Non-polar dimer formation during thermoxidation of edible fats.*Fat Science and Technology*.**89**: 216-220.

Dobarganes, M.C., Márquez-Ruiz, G.,(1996). Dimeric and higher oligomerictriglycerides. In: Perkins, E.G., Erickson, M.D. (Eds.), Deep frying. Chemistry, Nutrition and Practical Applications AOCS Press, Illinois, pp. 89–111.

Farhoosh, R.and Moosavi S. M. R (2009).Evaluating the Performance of Peroxide and Conjugated Diene Values in Monitoring Quality of Used Frying Oils *Journal of Agriculture Science Technology.***11**: 173-179.

FAS-USDA – Foreign Agricultural Service – United States Department of Agriculture 2008.

Fauziah, A., Razali, I. and Nor-Aini, S. (2000). Frying performance of palm olein and high oleic sunflower oil during batch frying of potato crisps. *Palm Oil Developments* **3**:1-7.

Firestone, D. (May–Jun 1994). "Determination of the iodine value of oils and fats: summary of collaborative study". *J AOAC* Int. **77** (3): 674–6. PMID 8012219

Firestone, D. and Reina, R.J. (1996). Authenticity of vegetable oils. In: Ashurst, P.R, and Dennis, M. J. (Eds.) *Food Authentication*. Blackie Academic and Professional, London, pp 198-258.

formation during deep-fat frying of French fries in vegetable oils. Palm Oil Developments No. 38.

Frankel, E.N. (2005). Lipid Oxidation 2nd ed. Dundee (UK): The Oily PressPp.470-471.

German Society for Fat Research, DGF (2000). Proceedings of the 3rd International Symposium of Deep-Fat Frying - Final Recommendations. *European Journal of Lipid ScienceTechnology.***102:** 594-595.

Gertz, C and Kochhar, S.P. (2001) A new method to determine oxidative stability of vegetable fats and oils at simulated frying temperature. *Oil City Library* **1**:82-88.

Gomes, T., Caponio, F. and Felcuratolo, D. (2003). Non-conventional parameters for quality evaluation of refined oils with special reference to commercial class olive oil. *Food chemistry*, **83**: 403-408.

Granda, C., Moreira, R. G. and Tichy, S. E. (2004). Reduction of Acrylamide Formation in Potato Chips by Low-temperature Vacuum Frying. *Journal of Food Science.* **69**: 405–411.

Gunstone, F.D., Harwood, J. L.and Dijkastra, A. J. (2007). *The lipid handbook,* 3rd edition, New York (Chapman and Hall), ISBN 978-0-89-349688-5.

Gunstone, T.D. (2008) Oils and fats in the food industry (1sted) Wiley Blackwell chapter 8

Gupta, M. K. (2005). Frying oils. In F. Shahidi (Ed.), *Bailey's industrial oil and fat products*.**4**:1-31.

Halvorsen. B. L. and Blomhoff, R. (2011). Determination of lipid oxidation products in vegetable oils and marine omega-3 supplements; Food and Nutrition Research.**55:** 5792-5793.

Heness, P.(2006). Maximising cooking oil life. *Asian food journal*.**2**: 6-9.

Henry, C.J.K and Chapman C. (2002).The nutrition handbook for food processors. United Kingdom: Woodland Publishing Limited.

http//:"Vegetable Oil – Everything You Need To Know About Vegetable Oils". http://vegetableoils.org/vegetableoil/ Accessed on October 21st, 2012 at 8.00 pm.

http//:www.allcookingtips.com/2007/10/12/types-randicity/-United States accessed on October 20th, 2012 at 9.00 am.

http://www.goaskalice.columbia.edu/reusing-cooking-oil-safe accessed on September 14th 2012 at 14.00 pm. http://www.fas.usda.gov/psdonline/psd Home.aspx.accessed on December 16th, 2015 at 12.00pm

http://www.kkgm.gov.tr/TGK/ Teblig˘/2001-29.html Turkish Codex Standards (accessed Jan. 2010)

https://www.britannica.com/science/stationary-phase-chromatographyaccesed on 14th, 2016 at 9.00 a m.

IFRA Analytical Method (2011). Determination of the Peroxide Value

Jamie A., Rodney J. M. and Kerrie G. (2012). The Effect of storage conditions on extra virgin olive oil quality. *Publication no*. 12/024

Kalogianni, E.P., Kaastogiannidou, C., Karapantsios, T.D. (2009). Effect of presence and absence of potatoes under repeated frying conditions on the composition of palm oil. *Journal of the American Oil Chemists' Society* **86** (**6**):561–571.

Kalogianni, E.P., Karastogiannidou, C. and Karapantsios, T.D. (2010). Effect of potato presence on the degradation of extra virgin olive oil during frying. *International Journal of Food Science and Technology* .**45**:765–775.

Khan M.I., Asha, M.R., Bhat K.K. and Khatoon S., (2011). Studies on chemical and sensory parameters of coconut oil and its olein blends with sesame oil and palm olein during wheat flour-based product frying. *Journal of Food Science Technology* **48**:175-182.

Kiatsrichart, S., Brewer, M.S., Cadwallder, K. R. and Artz, W.E. (2003).Pan-frying stability of nusun oil, a mid-oleic sunflower oil, *Journal of the America Oil Chemists' Society,* **80**:479-483.

Kim, J., Kim, D.N., Lee, S.H., Yoo S.H. and Lee, S. (2010). Correlation of fatty acid composition of vegetable oils with rheological behaviour and oil uptake. *Food Chemistry* **118**:398-402.

Kochhar, S.P. (2001). The composition of frying oils. In: Rossell, J.B. (Ed.) *Frying – Improving Quality.* Woodhead Publishing. Cambridge, England. pp 87-114.

Kochhar, S.P. and Henry, C.J. (2009).Oxidative stability and shelf life evaluation of selected culinary oils. *International Journal of Food Science and Nutrition* **60**:289–296.

Kress Rogers E., Gillat P.N. and Rosel J.B. (1990). Development and evaluation of a novel sensor for the in situ assess-ment of frying oil quality. *Food Control* **1**:163-178

Labrinea, E.P.; Thomaidis, N.S. and Georgiou, C. A., (2001). Direct olive oil anisidine value determination by flow injection. *Analytica Chimica Acta*, **448**: 201-206.

Läubli, M. W. and Bruttel, P. A. (1986). Determination of the oxidative stability of fats and oils: comparison between the active oxygen method (AOCS Cd 12-57) and the Rancimat method. *Journal of American Oil Chemists' Society.***63**: 792-795.

Mariod, A., Matthaus, B., Eichner, K. and Hussein, I. H. (2006). Frying quality and oxidative stability of two unconventional oils. *Journal of the American Oil Chemists' Society* **83:** 529-538.

Marmesat, S., Rodrigues, E., Velasco, J. and Dobarganes, C. (2007). Quality of used frying fats and oils: comparison of rapid tests based on chemical and physical oil properties. *International Journal of Food Science and Technology.***42**: 601-608.

McDonald, B.E. and Eskin, M.N.A. (2006). Role of fat in the diet. In: Erickson, M.D. (Ed.) *Deep Frying; Chemistry, Nutrition, and Practical Applications*. AOCS Press, Urbana, IL, pp 167-171.

Medina, I., Lois, S., Alcántara D., Lucas R. and Morales J.C. (2009). Effect of lipophilization of hydroxytyrosol on its antioxidant activity in fish oils and fish oil-in water emulsions. *Journal of Agriculture and Food Chemistry*, **57(20):**9773–9779. [PubMed]

Mehta, U. and Swinburn, B. (2001).A review of factors affecting fat absorption in hot chips. *Critical review food science nutrition*, **4:** 133-154.

Melton, S. J., Danielle, S. and Trigiano, M. K.(1994). Review of stability measurements for frying oils and fried food flavor. *Journal of the American Oil Chemists' Society,* **71:** 1301-1308.

Merwe, van der G. H., Plessis, du L. M. and Taylor, J. R. N. (2003). Changesin chemical quality indices during long-term storage of palm-olein oil under heated storage and transport type conditions.*Journal of Science and Food Agriculture*.**84**: 52–58.

Muhammad, N., E. Bamishaiye, E., Bamishaiye, O., Usman L., Salawu, M.,Nafiu, M. and Oloyede, O. (2011). Physicochemical properties and fatty acid composition of cyperusesculentus (Tiger Nut) Tuber Oil. *Bioresearch Bulletin* **5**: 51-54.

Nawar, W.W. (1996). Lipids. In: Fennema, O. R. (Ed.) *Food Chemistry*. (3rd edition), Marcel Dekker, Inc., New York, USA, Pp. 225-319.

Naz S., Sheikh, H., and Saddiqi, S.A. (2004). Oxidative stability of olive, corn and soybean oil under different conditions. *Food Chem* **88**:253-259.

Nummer, B.A., Schaffner, D.W., Fraser, A.M. and Andress, E.L. (2011). Current food safety issues of home-prepared vegetables and herbs stored in oil. Food Protection Trends **31**: 336-342.

Omolara, O.O and Dosumu, O.O (2009) "Preliminary Studies on the effect of processing methods on the quality of three commonly consumed marine fishes in Nigeria," *Biokemistri,* **21**: 1-7.

Orthoefer, F. T. and Cooper, D. S. (1996). Evaluation of used frying oil, in: Deep frying chemistry, Nutrition and practical Applications, Ed by Perkin EG and Erickson MD. AOCS Press, Champaign, IL, pp. 285-310.

Patterson, H.B.W. (1989). Handling and storage of oil seeds, oils, fats and meal.Elsevier science publisher Ltd, UK, Pp. 301-311.

Paul, S. and Mittal, S.G. (1996). Frying based on physical properties. *Journal of Food Process Engineering*.**19** (**2**): 201–221.

Pavia, D. L., Lampman,M. G., Kritz, S. G. and Engel R.G. (2006). Introduction to Organic Laboratory Techniques (4th Ed.).Thomson Brooks/Cole. pp. 797–817.

Peled, M.T., Gutfinger T. and Letch, A. (1975). Effect of water and BHT on stability of cottonseed oil during frying. *Journal of science, food and agriculture, **26***: 1655-1656

Perkins, E. G. (1967). Formation of non-volatile decomposition products in heated fats and oils. *Food Technology* **21**: 125-130

Perkins, E. G. (1992). In A. J. St. Angelo, ed., Lipid Oxidation in Food, *American Chemical Society*, Washington, D.C. Pp. 310–321.

Quiles, J.L., Ramírez-Tortosa., M.C., Ibáñez, S., Alfonso González, J., Duthie, G.G. and Huertas, J.R. (1999). Vitamin E supplementation increases the stability and the in vivo antioxidant capacity of refined olive oil. *Free Radical Research* **31**: 129–135.

Razali, I. and Badri, M. (2003). Oil absorption, polymer and polar compounds

Reblova Z, Kudrnova J, Trojakova L. and Pokorny J. (1999). Effect of rosemary extracts on the stabilization of frying oil during deep fat frying. *Journal of Food Lipids.***6**:13–23.

Robin, D. (1999). *The International Cocoa Trade*. Woodhead Publishing.Pp.169-170.

Romero, A., Cuesta, C. and Sanchez-Muniz, F. J. (1998). Effect of oil replenishment during deep-fat frying of frozen foods in sunflower oil and high-oleic acid sunflower oil. *Journal of the American Oil Chemists' Society*. **75**: 161-167.

Ruiz, A. M., Cañada, J. A. and Lendl, B. (2001). "A rapid method for peroxide value determination in edible oils based on flow analysis with Fourier transform infrared spectroscopic detection. *Analyst.***126**: 242-246.

Saroj, P. (1999). *The Pearson Guide to the B.Sc.* Pearson Education India.Pp. 109.

Sebedio, J. L., Bonpunt, A., Grandgirard, A. and Prevost, J. (1990). Deep-fat frying of frozen pre-fried French fries: influence of the amount of linolenic acid in the frying medium. *Journal of Science and Food Agriculture.* **38**: 1862-1867.

Sebedio, J. L., Dobarganes M C., Marquez, G., Wester, I. and Christie W. (1996) Industrial production of crisps and pre- fried French fries using sunflower oils. *Grasasy Aceites.***47**: 1-2.

Shahidi F (2005). Quality assurance of oils: Bailey's Industrial Oil and Fat Products, 6 Volume Set, 6th Edition Hoboken: John Wiley & Sons, Inc.; Pp. 565-575.

Shahidi F. and Zhong Y. (2005). Lipid measurements: Bailey's Industrial Oil and Fat Products, 6 Volume Set, 6th Edition Hoboken: John Wiley & Sons, Inc.; Pp. 357-386.

Shahidi, F. and Wanasundara, U. N. (2002). In Akoh, C. C. and Min, D. B. eds., Food Lipids: Chemistry, Nutrition and Biotechnology, Marcel Dekker, Inc., New York, Pp. 465–487.

Sharoba, A. and Mohamed, F. R..(2012). Impact of frying on fatty acid profile and rheological behavior of some vegetable oils. *Journal of Food Process Technology*.**3**: 7-8.

Sonestedt, E., Ericson, U., Gullberg, B., Olsson, H. and Wirfält, E. (2008). "Do both heterocyclic amines and *omega*- polyunsaturated fatty acids contribute to the incidence of breast cancer in postmenopausal women of the Malmö diet and cancer cohort?" *The International Journal of Cancer*.**123**: 1637–1643.

Steenson, D. and Min, D. B. (2000). Effects of *B*-carotene and lycopene thermal degradation products on the oxidative stability of soybean oil. *Journal of the American Oil Chemists' Society,* **77**: 1153-1160.

Stender, S. and Dyerberg, J. (2003). *The influence of trans fatty acids on health. A report from Danish Nutrition council.* Fourth edition.Pp 86

Stevenson, S. G., Vaisey-Genser, M., and Eskin, N. A. M. (1984). Quality control in the use of deep frying oils. *Journal of the American Oil Chemists' Society,* **61**: 1102–1108.

Sulieman, A.E.R.M, Makhzangy A.E Ramadhan , M.F (2006). Antiridial performance and physiocochemical characteristics of vegetable oil upon frying of French fries A preliminary comparative, *journal of food lipid* **12**:259-276

Tamzid H. M., Alam M.T. and Islam M.A., (2007). Physico-chemical and nutritional studies of *Terminaliabelericaroxb.* Seed oil and seed kernel. *Journal of Biosciences,* **15***:*117-126.

Tompkins, C. and E.G.Perkins E. G. (2000).Frying performance of low-linolenic acid soybean oil.*Journal of American Oil Chemist's Society,* **77:**223-229

Tyagi, V. K. and Vasishtha, A. K. (1996). Changes in the characteristics and composition of oils during deep-fat frying.*Journal of the American Oil Chemists' Society*. **73**: 499–506

Tynek M, Hazuka Z, Pawlowicz R. and Dudek M. (2001). Changes in the frying medium during deep frying of food rich in proteins and carbohydrates.*J Food Lipids***8**:251–261.

Ulbert, F. and Roubicek, D. (1993). Evaluation of a static headspace gas chromatographic method for the determination of lipid peroxides. *Journal of food chemistry*.**46**: 37-38.

Velasco, J., Marmesat, S. and Dobarganes, C. (2008). Chemistry of Frying. In: "*Deep Fat Frying of Foods*" (Edited by Sahin, S. and Sumnu, G. Taylor and Francis, U. S. A). Pp. 33-56.

Warner, K. (2002). Chemistry of frying oils. In Akoh, C. C. and Min, D. B. (Eds.), *Food lipids Chemistry*, *Nutrition*, and *Biotechnology*, 2nd *ed*., New York: Marcel Dekker, Inc. Pp. 205–221.

WHO/FAO (World Health Organization/Food and Agriculture Organization) *(*2009*).*Fats and fatty acids in human nutrition.Proceedings of the Joint FAO/WHO Expert Consultation. Geneva, Switzerland. *Annals of Nutrition and Metabolism.***55***:* 5–300.

Yamsaengsung, R. and Moreira, R.G. (2002). Modeling the transport phenomena and structural changes during deep fat frying, Part II: model solution & validation. *Journal of Food Engineering.***53**: 11-25.

Zhang Q., Saleh, A.S.M., Chen, J. and Shen Q. (2012). Chemical alterations taken place during deep-fat frying based on certain reaction products: a review. *Chemistry and Physics Lipids* **165**:662–681.

APPENDICES

Appendix 1: Fatty acid composition of oils (ug/ml)

Sample	Decanoic (C10:0)	Lauric (C12:0)	Myristic (C14:0)	Palmitic (C16:0)	Stearic (C18:0)	Oleic (C18:1)	Linoleic (C18:2)	Linolenic (C18:3)	Total unsaturation
1	0.00	0.00	2.58	221.33	40.20	728.26	1550.26	4.48	2283.00
2	3.16	0.00	0.00	32.18	5.17	95.98	192.48	0.64	289.10
3	0.00	0.00	0.00	17.94	2.35	46.09	78.92	0.96	125.97
4	3.49	17.90	0.00	235.55	42.13	800.28	1499.83	4.66	2304.77
5	0.00	1.12	0.10	62.63	0.00	223.45	436.99	1.32	661.76
6	0.00	0.00	0.14	91.08	0.00	337.68	648.53	2.06	988.27
7	0.00	0.00	0.46	129.78	53.74	353.22	1149.20	36.23	1538.65
8	0.00	0.00	0.63	185.08	1.00	454.25	1469.17	72.10	1995.52
9	0.00	0.00	0.55	140.88	56.73	361.26	1047.60	45.07	1453.93
10	0.00	0.10	0.33	74.45	0.00	215.49	596.89	20.66	833.04
11	0.00	0.00	0.40	105.61	33.62	266.99	829.58	38.78	1135.35
12	0.00	0.00	0.51	134.09	0.00	393.57	1061.07	50.58	1505.22

13	0.00	0.00	0.07	48.03	3.99	275.42	255.09	0.72	531.23
14	0.00	0.00	0.26	91.14	0.00	773.22	375.57	2.33	1151.12
15	0.00	0.00	0.69	114.02	8.31	977.37	436.53	2.12	1416.02
16	0.00	0.00	0.48	81.05	7.43	465.15	285.23	3.39	753.77
17	0.04	0.76	3.20	246.60	8.50	354.04	113.02	0.53	467.59
18	0.00	0.75	3.61	312.04	0.00	463.21	135.60	0.81	599.62
19	0.00	0.59	1.48	236.96	1.45	346.21	108.36	1.19	455.76
20	0.00	0.71	3.72	314.02	0.00	440.83	123.49	0.91	565.23
21	0.00	0.77	3.59	318.29	0.00	457.69	165.62	0.61	623.92
22	0.00	1.38	6.92	532.19	54.63	716.35	241.88	0.78	959.01
23	0.00	1.34	6.06	512.99	0.00	701.37	214.53	0.76	916.66
24	0.00	0.00	0.49	51.09	0.00	231.63	553.99	0.68	786.30
25	0.00	0.00	0.78	104.75	0.00	563.63	1445.34	1.07	2010.04
26	0.00	0.00	0.92	116.60	27.49	486.08	ND	ND	486.08
27	0.00	0.09	0.84	87.11	0.00	366.68	888.97	1.76	1257.41

1.	Corn fresh	14	Peanut fish 6hr
2.	Corn mandazi (6hr)	15	Peanut fish (12hr)-5days RT
3.	Corn mandazi (12hr)-5 days refri.	16	Peanut fish (12hr)-5days Corked
4.	Corn mandazi (12hr)-5days RT	17	Palm fresh
5.	Corn mandazi 5days RT	18	Palm fish (6hr)
6.	Corn mandazi 5 days refri	19	Palm fish (12hr) 5days RT
7.	Soybean fresh	20	Palm fish (12hr) 5days Corked
8.	Soybean mandazi (6hr)	21	Palm chips (6hr)
9.	Soybean mandazi(12hr) 5days refri	22	Palm chips (12hr) 5days RT
		23	Palm chips (12hr) 5days refri
10.	Soybean mandazi(12hr) 5days RT	24	Sunflower fresh
11.	Soybean mandazi 5days RT	25	Sunflower chicken (6hr)
12	Soybean mandazi 5days refri	26	Sunflower chicken (12hr) 5days RT
13	Peanut fresh	27	Sunflower chicken (12hr) 5days refri

Appendix 2: ANOVA: Peroxide values based on type of food fried

	Number of obs = 95			R-squared = 0.5950	
	Root MSE = 1.00629			Adj R-squared = 0.4561	
Source	Partial SS	df	MS	F	Prob> F
Model	104.129	24	4.33871	4.28	0
Oil	62.5026	4	15.6256	15.43	0
Food	15.3464	4	3.8366	3.79	0.0076
oil#food	31.4517	16	1.96573	1.94	0.0304
Residual	70.884	70	1.01263		
Total	175.013	94	1.86184		

Appendix 3: ANOVA: S.V based on type of food fried

	Number of obs = 95			**R-squared = 0.2739**	
	Root MSE = 25.1321			Adj R-squared = 0.0250	
Source	Partial SS	df	MS	F	Prob> F
Model	16680.9	24	695.037	1.1	0.3665
Oil	2847.31	4	711.826	1.13	0.3509
Food	2983.45	4	745.863	1.18	0.3268
oil#food	9856.89	16	616.056	0.98	0.4923
Residual	44213.7	70	631.624		
Total	60894.6	94	647.814		

Appendix 4: ANOVA: P.C after deep frying food

	Number of obs = 95			**R-squared = 0.3811**	
	Root MSE = .114934			Adj R-squared = 0.1689	
Source	Partial SS	df	MS	F	Prob> F
Model	0.56945	24	0.02373	1.8	0.0307
Oil	0.08429	4	0.02107	1.6	0.1852
Food	0.02231	4	0.00558	0.42	0.7921
oil#food	0.43117	16	0.02695	2.04	0.0218
Residual	0.92469	70	0.01321		
Total	1.49414	94	0.0159		

Appendix 5: ANOVA: I.V of oils based on food fried

	Number of obs = 95			**R-squared = 0.9474**	
	Root MSE = 7.41042			**Adj R-squared = 0.9293**	
Source	Partial SS	df	MS	F	Prob> F
Model	69214.4	24	2883.93	52.52	0
Oil	63380.8	4	15845.2	288.54	0
Food	889.407	4	222.352	4.05	0.0052
oil#food	1136.02	16	71.0012	1.29	0.2265
Residual	3844	70	54.9143		
Total	73058.4	94	777.217		

Appendix 6: ANOVA: p-AVof oils based on food fried

	Number of obs = 95			**R-squared = 0.5368**	
	Root MSE = 15.6932			**Adj R-squared = 0.3779**	
Source	Partial SS	df	MS	F	Prob> F
Model	19975.7	24	832.321	3.38	0
Oil	5036.18	4	1259.04	5.11	0.0011
Food	5785.2	4	1446.3	5.87	0.0004
oil#food	9372.12	16	585.757	2.38	0.0068
Residual	17239.4	70	246.277		
Total	37215.1	94	395.905		

Appendix 7: ANOVA: R.I of oils based on food fried

	Number of obs = 95			**R-squared = 1.0000**	
	Root MSE = .00108			**Adj R-squared = 1.0000**	
Source	Partial SS	Df	MS	F	Prob> F
Model	10.5909	24	0.44129	3.80E+05	0
Oil	1.25544	4	0.31386	2.70E+05	0
Food	1.64896	4	0.41224	3.50E+05	0
oil#food	6.61861	16	0.41366	3.50E+05	0
Residual	8.2E-05	70	1.17E-06		
Total	10.591	94	0.11267		

Appendix 8: ANOVA: Density of oils based on food fried

	Number of obs = 95			**R-squared = 0.7803**	
	Root MSE = .103312			**Adj R-squared = 0.7050**	
Source	Partial SS	df	MS	F	Prob> F
Model	2.65407	24	0.11059	10.36	0
Oil	0.28194	4	0.07049	6.6	0.0001
food	0.44579	4	0.11145	10.44	0
oil#food	1.67719	16	0.10482	9.82	0
Residual	0.74714	70	0.01067		
Total	3.40121	94	0.03618		

Appendix 9: ANOVA for peroxide values based on storage conditions.

	Number of obs = 100			**R-squared = 0.4833**	
	Root MSE = 1.19805			**Adj R-squared = 0.2693**	
Source	Partial SS	df	MS	F	Prob> F
Model	93.9819	29	3.24076	2.26	0.0029
Storage	9.4363	5	1.88726	1.31	0.2678
Oil	54.0685	4	13.5171	9.42	0
storage#oil	16.3132	20	0.81566	0.57	0.9216
Residual	100.472	70	1.43531		
Total	194.454	99	1.96418		

Appendix 10: ANOVA for S.Vs based on storage conditions.

	Number of obs = 100			**R-squared = 0.3157**	
	Root MSE = 25.1294			**Adj R-squared = 0.0322**	
Source	Partial SS	Df	MS	F	Prob> F
Model	20394.8	29	703.27	1.11	0.3488
Storage	11246.1	5	2249.21	3.56	0.0063
Oil	2484.74	4	621.186	0.98	0.4222
storage#oil	5406.07	20	270.304	0.43	0.982
Residual	44204.1	70	631.487		
Total	64598.9	99	652.515		

Appendix 11: ANOVA for polymer content based on storage conditions.

	Number of obs = 100			**R-squared = 0.4074**	
	Root MSE = .113875			**Adj R-squared = 0.1619**	
Source	Partial SS	df	MS	F	Prob> F
Model	0.62407	29	0.02152	1.66	0.044
Storage	0.2044	5	0.04088	3.15	0.0127
Oil	0.14995	4	0.03749	2.89	0.0283
storage#oil	0.3041	20	0.0152	1.17	0.3036
Residual	0.90773	70	0.01297		
Total	1.5318	99	0.01547		

Appendix 12: ANOVA for Iodine values based on storage conditions.

	Number of obs = 100			**R-squared = 0.9582**	
	Root MSE = 6.8444			**Adj R-squared = 0.9410**	
Source	Partial SS	Df	MS	F	Prob> F
Model	75264.3	29	2595.32	55.4	0
Storage	2534.49	5	506.898	10.82	0
Oil	50062.1	4	12515.5	267.16	0
storage#oil	1197.3	20	59.8651	1.28	0.2233
Residual	3279.21	70	46.8458		
Total	78543.5	99	793.369		

Appendix 13: ANOVA for p-Av of oils based on storage conditions

	Number of obs = 100			**R-squared = 0.3594**	
	Root MSE = 19.3783			**Adj R-squared = 0.0940**	
Source	Partial SS	df	MS	F	Prob> F
Model	14746.7	29	508.508	1.35	0.1521
Storage	7242.97	5	1448.59	3.86	0.0038
Oil	2442.82	4	610.704	1.63	0.1773
storage#oil	2940.89	20	147.045	0.39	0.9893
Residual	26286.3	70	375.519		
Total	41033	99	414.475		

Appendix 14: ANOVA for Refractive indices based on storage conditions.

	Number of obs = 100			**R-squared = 0.2814**	
	Root MSE = .330188			**Adj R-squared = -0.0164**	
Source	Partial SS	Df	MS	F	Prob> F
Model	2.98799	29	0.10303	0.95	0.5541
Storage	0.15257	5	0.03051	0.28	0.9227
Oil	1.73997	4	0.43499	3.99	0.0057
storage#oil	0.63013	20	0.03151	0.29	0.9985
Residual	7.63169	70	0.10902		
Total	10.6197	99	0.10727		

Appendix 15: ANOVA for density based on storage conditions

	Number of obs = 100			**R-squared = 0.2278**	
	Root MSE = .193751			**Adj R-squared = -0.0921**	
Source	Partial SS	df	MS	F	Prob> F
Model	0.77526	29	0.02673	0.71	0.844
Storage	0.04874	5	0.00975	0.26	0.9335
Oil	0.29104	4	0.07276	1.94	0.1137
storage#oil	0.22391	20	0.0112	0.3	0.9982
Residual	2.62775	70	0.03754		
Total	3.403	99	0.03437		

Appendix 16: Variation of fatty acids based on the type of oil

Variable	**Source**	**Partial SS**	**df**	**MS**	**F**	**Prob> F**	**R- squared**
Decanoic	Model	5.71301	4	1.42825	2.12	0.112	0.2786
	Residual	14.7967	22	0.67258			
	Total	20.5097	26	0.78883			
Lauric	Model	41.8649	4	10.4662	0.88	0.4926	0.1378
	Residual	262.001	22	11.9091			
	Total	303.865	26	11.6871			
Myristic	Model	66.4678	4	16.617	14.14	0	0.72
	Residual	25.8509	22	1.17504			
	Total	92.3187	26	3.55072			
Palmitic	Model	322687	4	80671.7	12.35	0	0.6919
	Residual	143717	22	80671.7			
	Total	466404	26	17938.6			
Stearic	Model	1283.15	4	320.787	0.79	0.5412	0.1263
	Residual	8879.42	22	403.61			
	Total	10162.6	26	390.868			
Oleic	Model	246425	4	61606.2	1.29	0.3037	0.1902
	Residual	1049211	22	47691.4			
	Total	1295636	26	49832.2			
Linoleic	Model	2933425	4	733356	4.64	0.0072	0.4577
	Residual	3474936	22	157952			
	Total	6408361	26	246475			
Linolenic	Model	8393.58	4	2098.39	31.03	0	0.8495
	Residual	1487.52	22	67.6143			
	Total	9881.09	26	380.042			

Appendix 17: Average values of various parameters of oils after frying food

Type of oil	chicken				Chips				Fishes				Mandazi				Smokies			
	P.V	paV	I.V	RI	P.V	p-Av	I.V	RI	PV	PaV	IV	RI	PV	Pav	IV	RI	PV	pav	IV	RI
Corn	1.0	31.8	13.5	14	0.5	48.2	13.5	16	0.6	47.1	25.4	4	0.9	22.3	17.5	8	0.9	22.2	13.5	22
Palm	1.5	6.0	5.1	3	0.3	16.1	4.6	15	0.8	11.6	7.61	-1	0.5	12.4	2.0	6	0.3	15.8	6.6	13
Peanut	0.1	15.6	5.9	20	-0.7	31.0	5.9	2	1.1	43.0	10.1	8	0.8	31.1	13.7	16	0.3	33.0	8.3	15
Soybean	0.7	40.0	27.0	4	-0.1	52.2	22.0	10	0.6	29.1	35.5	11	1.2	34.0	38.1	19	0.9	ND	17.3	ND
Sunflower	0.1	41.1	5.1	22	1.7	46.2	5.1	22	4.0	42.0	7.6	15	0.6	49.6	9.1	12	0.1	19.2	4.6	14

RI values = RI × 10^{-4}

Appendix 18: The standards

STANDARDS

Component	Pacomp	PAi.s	wt comp	wt i.s	RF
Decanoic Acid	785547	739316	0.015	0.015	0.941
Lauric Acid	103294	125579	0.015	0.015	1.216
Myristic Acid	178510	187653	0.02	0.02	1.051
Pulmitic Acid	182200	195415	0.03	0.02	1.609
Stearic Acid	142019	238253	0.02	0.02	1.678
Oleic Acid	545872	193727	0.09	0.02	1.597
Linoleic Acid	199068	148064	0.06	0.02	2.231
Linolenic Acid	476598	200387	0.06	0.02	1.261

SAMPLES NO.1

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	16274	0.02	0.941	10	0.00
Lauric Acid	0	16274	0.02	1.216	10	0.00
Myristic Acid	1000	16274	0.02	1.051	10	2.58
Pulmitic Acid	55966	16274	0.02	1.609	10	221.33
Stearic Acid	9748	16274	0.02	1.678	10	40.20
Oleic Acid	185531	16274	0.02	1.597	10	728.26
Linoleic Acid	282708	16274	0.02	2.231	10	1550.26
Linolenic Acid	1444	16274	0.02	1.261	10	4.48

SAMPLES NO.2

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	19785	117912	0.02	0.941	5	3.16
Lauric Acid	0	117912	0.02	1.216	5	0.00
Myristic Acid	0	117912	0.02	1.051	5	0.00
Pulmitic Acid	117912	117912	0.02	1.609	5	32.18
Stearic Acid	18151	117912	0.02	1.678	5	5.17
Oleic Acid	354334	117912	0.02	1.597	5	95.98
Linoleic Acid	508655	117912	0.02	2.231	5	192.48
Linolenic Acid	2977	117912	0.02	1.261	5	0.64

SAMPLES NO.3

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	26403	0.02	0.941	5	0.00
Lauric Acid	0	26403	0.02	1.216	5	0.00
Myristic Acid	0	26403	0.02	1.051	5	0.00
Pulmitic Acid	14723	26403	0.02	1.609	5	17.94
Stearic Acid	1850	26403	0.02	1.678	5	2.35
Oleic Acid	38096	26403	0.02	1.597	5	46.09
Linoleic Acid	46700	26403	0.02	2.231	5	78.92
Linolenic Acid	1000	26403	0.02	1.261	5	0.96

SAMPLES NO.4

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	2578	27825	0.02	0.941	10	3.49
Lauric Acid	10239	27825	0.02	1.216	10	17.90
Myristic Acid	0	27825	0.02	1.051	10	0.00

Pulmitic Acid	101837	27825	0.02	1.609	10	235.55
Stearic Acid	17467	27825	0.02	1.678	10	42.13
Oleic Acid	348590	27825	0.02	1.597	10	800.28
Linoleic Acid	467646	27825	0.02	2.231	10	1499.83
Linolenic Acid	2572	27825	0.02	1.261	10	4.66

SAMPLES NO.5

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	81727	0.02	0.941	5	0.00
Lauric Acid	3780	81727	0.02	1.216	5	1.12
Myristic Acid	386	81727	0.02	1.051	5	0.10
Pulmitic Acid	159051	81727	0.02	1.609	5	62.63
Stearic Acid	0	81727	0.02	1.678	5	0.00
Oleic Acid	571744	81727	0.02	1.597	5	223.45
Linoleic Acid	800397	81727	0.02	2.231	5	436.99
Linolenic Acid	4275	81727	0.02	1.261	5	1.32

SAMPLES NO.6

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	48613	0.02	0.941	5	0.00
Lauric Acid	0	48613	0.02	1.216	5	0.00
Myristic Acid	328	48613	0.02	1.051	5	0.14
Pulmitic Acid	137596	48613	0.02	1.609	5	91.08
Stearic Acid	0	48613	0.02	1.678	5	0.00
Oleic Acid	513949	48613	0.02	1.597	5	337.68
Linoleic Acid	706565	48613	0.02	2.231	5	648.53
Linolenic Acid	3968	48613	0.02	1.261	5	2.06

SAMPLES NO.7

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	26403	0.02	0.941	5	0.00
Lauric Acid	0	26403	0.02	1.216	5	0.00
Myristic Acid	575	26403	0.02	1.051	5	0.46
Pulmitic Acid	106484	26403	0.02	1.609	5	129.78
Stearic Acid	42280	26403	0.02	1.678	5	53.74
Oleic Acid	291985	26403	0.02	1.597	5	353.22
Linoleic Acid	680016	26403	0.02	2.231	5	1149.20
Linolenic Acid	37929	26403	0.02	1.261	5	36.23

SAMPLES NO.8

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	35891	0.02	0.941	5	0.00
Lauric Acid	0	35891	0.02	1.216	5	0.00
Myristic Acid	1079	35891	0.02	1.051	5	0.63
Pulmitic Acid	206423	35891	0.02	1.609	5	185.08
Stearic Acid	1072	35891	0.02	1.678	5	1.00
Oleic Acid	510438	35891	0.02	1.597	5	454.25
Linoleic Acid	1181758	35891	0.02	2.231	5	1469.17
Linolenic Acid	102606	35891	0.02	1.261	5	72.10

SAMPLES NO.9

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	27345	0.02	0.941	5	0.00
Lauric Acid	0	27345	0.02	1.216	5	0.00
Myristic Acid	716	27345	0.02	1.051	5	0.55
Pulmitic Acid	119713	27345	0.02	1.609	5	140.88
Stearic Acid	46226	27345	0.02	1.678	5	56.73
Oleic Acid	309286	27345	0.02	1.597	5	361.26
Linoleic Acid	642014	27345	0.02	2.231	5	1047.60
Linolenic Acid	48866	27345	0.02	1.261	5	45.07

SAMPLES NO.10

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	80914	0.02	0.941	5	0.00
Lauric Acid	343	80914	0.02	1.216	5	0.10
Myristic Acid	1257	80914	0.02	1.051	5	0.33
Pulmitic Acid	187204	80914	0.02	1.609	5	74.45
Stearic Acid	0	80914	0.02	1.678	5	0.00
Oleic Acid	545911	80914	0.02	1.597	5	215.49
Linoleic Acid	1082397	80914	0.02	2.231	5	596.89
Linolenic Acid	66278	80914	0.02	1.261	5	20.66

SAMPLES NO.11

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	40225	0.02	0.941	5	0.00
Lauric Acid	0	40225	0.02	1.216	5	0.00
Myristic Acid	766	40225	0.02	1.051	5	0.40
Pulmitic Acid	132016	40225	0.02	1.609	5	105.61
Stearic Acid	40302	40225	0.02	1.678	5	33.62
Oleic Acid	336250	40225	0.02	1.597	5	266.99
Linoleic Acid	747864	40225	0.02	2.231	5	829.58
Linolenic Acid	61845	40225	0.02	1.261	5	38.78

SAMPLES NO.12

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	36926	0.02	0.941	5	0.00
Lauric Acid	0	36926	0.02	1.216	5	0.00
Myristic Acid	895	36926	0.02	1.051	5	0.51
Pulmitic Acid	153867	36926	0.02	1.609	5	134.09
Stearic Acid	0	36926	0.02	1.678	5	0.00
Oleic Acid	455006	36926	0.02	1.597	5	393.57
Linoleic Acid	878103	36926	0.02	2.231	5	1061.07
Linolenic Acid	74064	36926	0.02	1.261	5	50.58

SAMPLES NO.13

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	69870	0.02	0.941	5	0.00
Lauric Acid	0	69870	0.02	1.216	5	0.00

Myristic Acid	223	69870	0.02	1.051	5	0.07
Pulmitic Acid	104288	69870	0.02	1.609	5	48.03
Stearic Acid	8297	69870	0.02	1.678	5	3.99
Oleic Acid	602500	69870	0.02	1.597	5	275.42
Linoleic Acid	400852	69870	0.02	2.231	5	255.99
Linolenic Acid	1999	69870	0.02	1.261	5	0.72

SAMPLES NO.14

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	34754	0.02	0.941	5	0.00
Lauric Acid	0	34754	0.02	1.216	5	0.00
Myristic Acid	423	34754	0.02	1.051	5	0.26
Pulmitic Acid	98430	34754	0.02	1.609	5	91.14
Stearic Acid	0	34754	0.02	1.678	5	0.00
Oleic Acid	841346	34754	0.02	1.597	5	773.22
Linoleic Acid	292524	34754	0.02	2.231	5	375.57
Linolenic Acid	3216	34754	0.02	1.261	5	2.33

SAMPLES NO.15

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	48734	0.02	0.941	5	0.00
Lauric Acid	0	48734	0.02	1.216	5	0.00
Myristic Acid	1611	48734	0.02	1.051	5	0.69
Pulmitic Acid	172677	48734	0.02	1.609	5	114.02
Stearic Acid	12074	48734	0.02	1.678	5	8.31
Oleic Acid	1491263	48734	0.02	1.597	5	977.37
Linoleic Acid	476776	48734	0.02	2.231	5	436.53
Linolenic Acid	4097	48734	0.02	1.261	5	2.12

SAMPLES NO.16

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	61126	0.02	0.941	5	0.00
Lauric Acid	0	61126	0.02	1.216	5	0.00
Myristic Acid	1406	61126	0.02	1.051	5	0.48
Pulmitic Acid	153948	61126	0.02	1.609	5	81.05
Stearic Acid	13541	61126	0.02	1.678	5	7.43
Oleic Acid	890190	61126	0.02	1.597	5	465.15
Linoleic Acid	390747	61126	0.02	2.231	5	285.23
Linolenic Acid	8214	61126	0.02	1.261	5	3.39

SAMPLES NO.17

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	186	86539	0.02	0.941	5	0.04
Lauric Acid	2709	86539	0.02	1.216	5	0.76
Myristic Acid	13158	86539	0.02	1.051	5	3.20
Pulmitic Acid	663162	86539	0.02	1.609	5	246.60
Stearic Acid	21929	86539	0.02	1.678	5	8.50
Oleic Acid	959256	86539	0.02	1.597	5	354.04
Linoleic Acid	219208	86539	0.02	2.231	5	113.02

SAMPLES NO.18

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	73786	0.02	0.941	5	0.00
Lauric Acid	2283	73786	0.02	1.216	5	0.75
Myristic Acid	12677	73786	0.02	1.051	5	3.61
Pulmitic Acid	715478	73786	0.02	1.609	5	312.04
Stearic Acid	0	73786	0.02	1.678	5	0.00
Oleic Acid	1070093	73786	0.02	1.597	5	463.21
Linoleic Acid	224240	73786	0.02	2.231	5	135.60
Linolenic Acid	2356	73786	0.02	1.261	5	0.81

SAMPLES NO.19

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	94091	0.02	0.941	5	0.00
Lauric Acid	2284	94091	0.02	1.216	5	0.59
Myristic Acid	6622	94091	0.02	1.051	5	1.48
Pulmitic Acid	692854	94091	0.02	1.609	5	236.96
Stearic Acid	4061	94091	0.02	1.678	5	1.45
Oleic Acid	1019894	94091	0.02	1.597	5	346.21
Linoleic Acid	228492	94091	0.02	2.231	5	108.36
Linolenic Acid	4440	94091	0.02	1.261	5	1.19

SAMPLES NO.20

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	66013	0.02	0.941	5	0.00
Lauric Acid	1939	66013	0.02	1.216	5	0.71
Myristic Acid	11694	66013	0.02	1.051	5	3.72
Pulmitic Acid	644177	66013	0.02	1.609	5	314.02
Stearic Acid	0	66013	0.02	1.678	5	0.00
Oleic Acid	911109	66013	0.02	1.597	5	440.83
Linoleic Acid	182690	66013	0.02	2.231	5	123.49
Linolenic Acid	2381	66013	0.02	1.261	5	0.91

SAMPLES NO.21

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	56726	0.02	0.941	5	0.00
Lauric Acid	1797	56726	0.02	1.216	5	0.77
Myristic Acid	9679	56726	0.02	1.051	5	3.59
Pulmitic Acid	561068	56726	0.02	1.609	5	318.29
Stearic Acid	0	56726	0.02	1.678	5	0.00
Oleic Acid	812857	56726	0.02	1.597	5	457.69
Linoleic Acid	210549	56726	0.02	2.231	5	166.62
Linolenic Acid	1379	56726	0.02	1.261	5	0.61

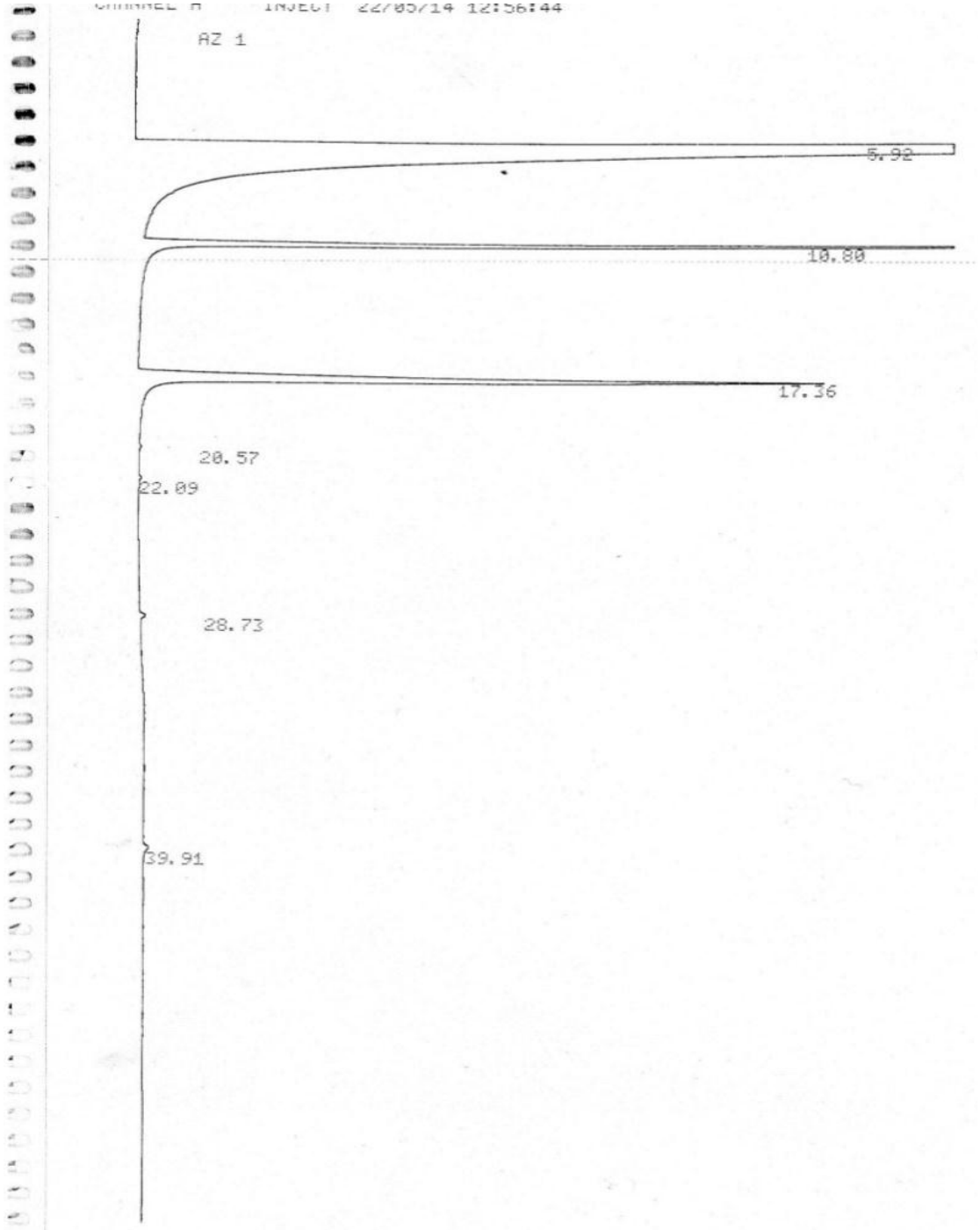
SAMPLES NO.22

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	14622	0.02	0.941	5	0.00

SAMPLES NO.27

Component	Pacomp	PAi.s	wt i.s	RF	DF	wt comp(ug/ml)
Decanoic Acid	0	39257	0.02	0.941	5	0.00
Lauric Acid	148	39257	0.02	1.216	5	0.09
Myristic Acid	1576	39257	0.02	1.051	5	0.84
Pulmitic Acid	106264	39257	0.02	1.609	5	87.11
Stearic Acid	0	39257	0.02	1.678	5	0.00
Oleic Acid	450678	39257	0.02	1.597	5	366.68
Linoleic Acid	782123	39257	0.02	2.231	5	888.97
Linolenic Acid	2747	39257	0.02	1.261	5	1.76

Appendix 19: Myristic acid + I.S



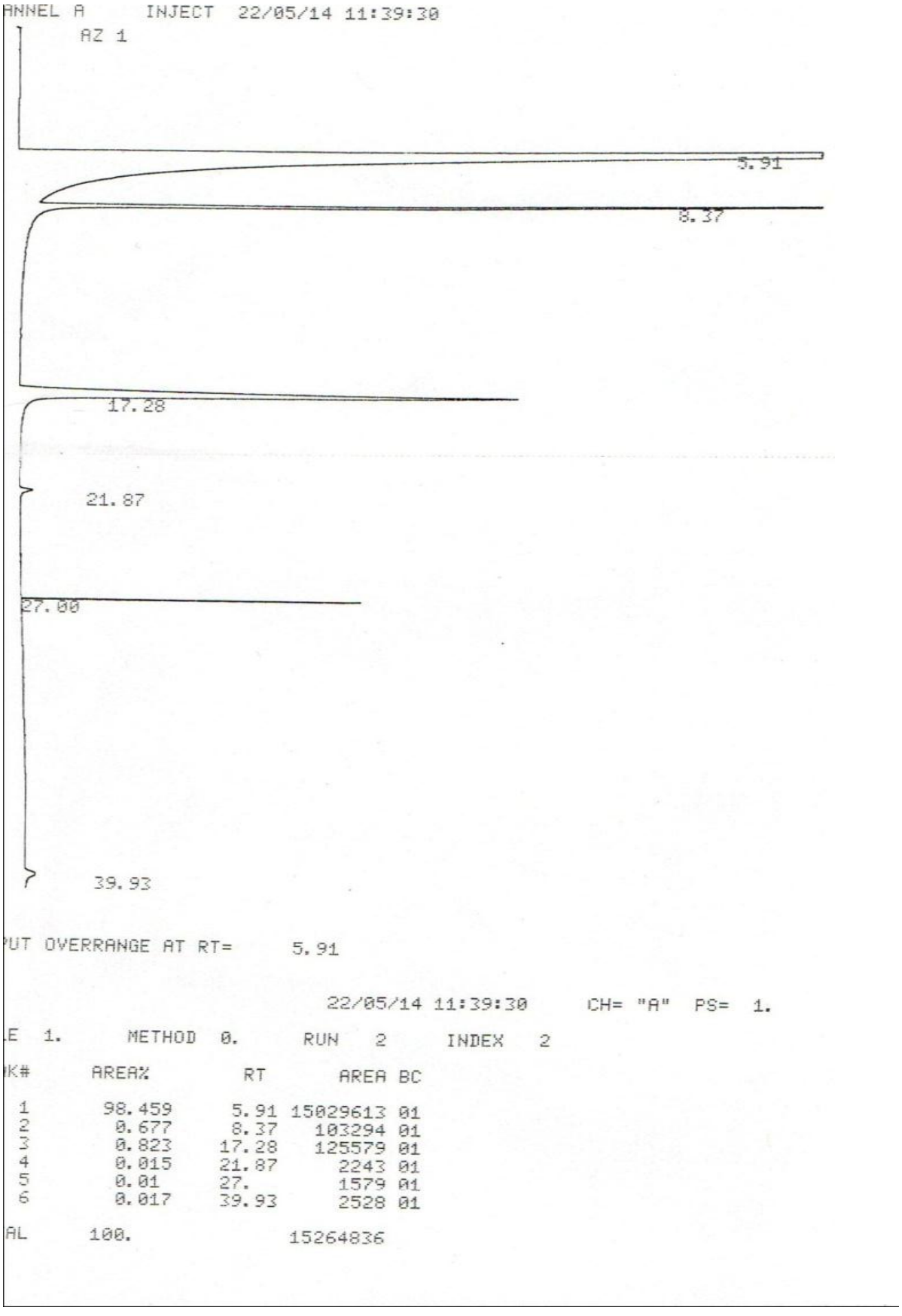
INPUT OVERRANGE AT RT= 5.92

22/05/14 12:56:44 CH= "A" PS= 1.

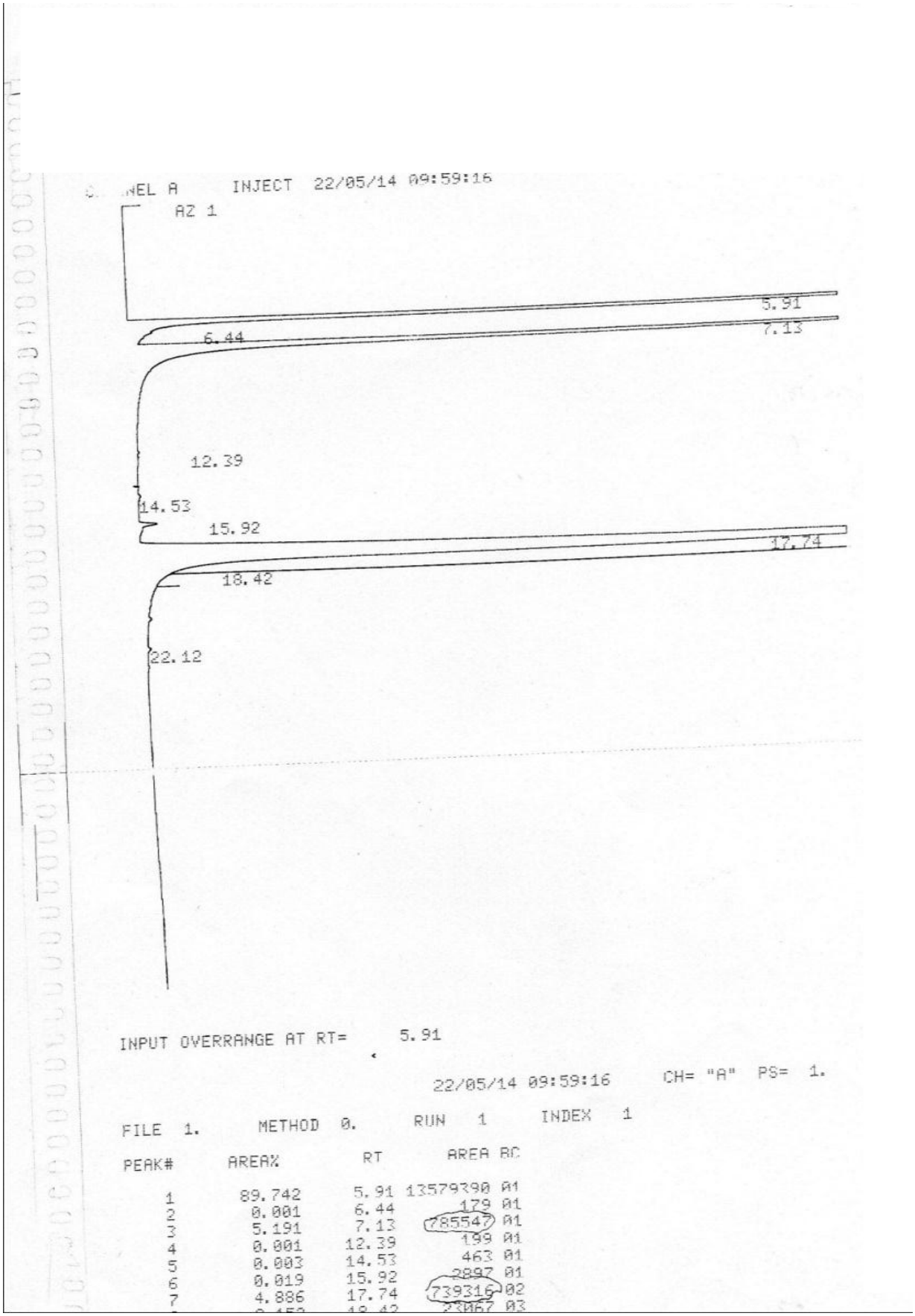
FILE 1. METHOD 0. RUN 3 INDEX 3

PEAK#	AREA%	RT	AREA	BC
1	97.909	5.92	17302530	01
2	1.01	10.8	178510	01
3	1.062	17.36	187653	01
4	0.002	20.57	317	01
5	0.003	22.09	498	01
6	0.006	28.73	1033	01
7	0.009	39.91	1515	01
TOTAL	100.		17672056	

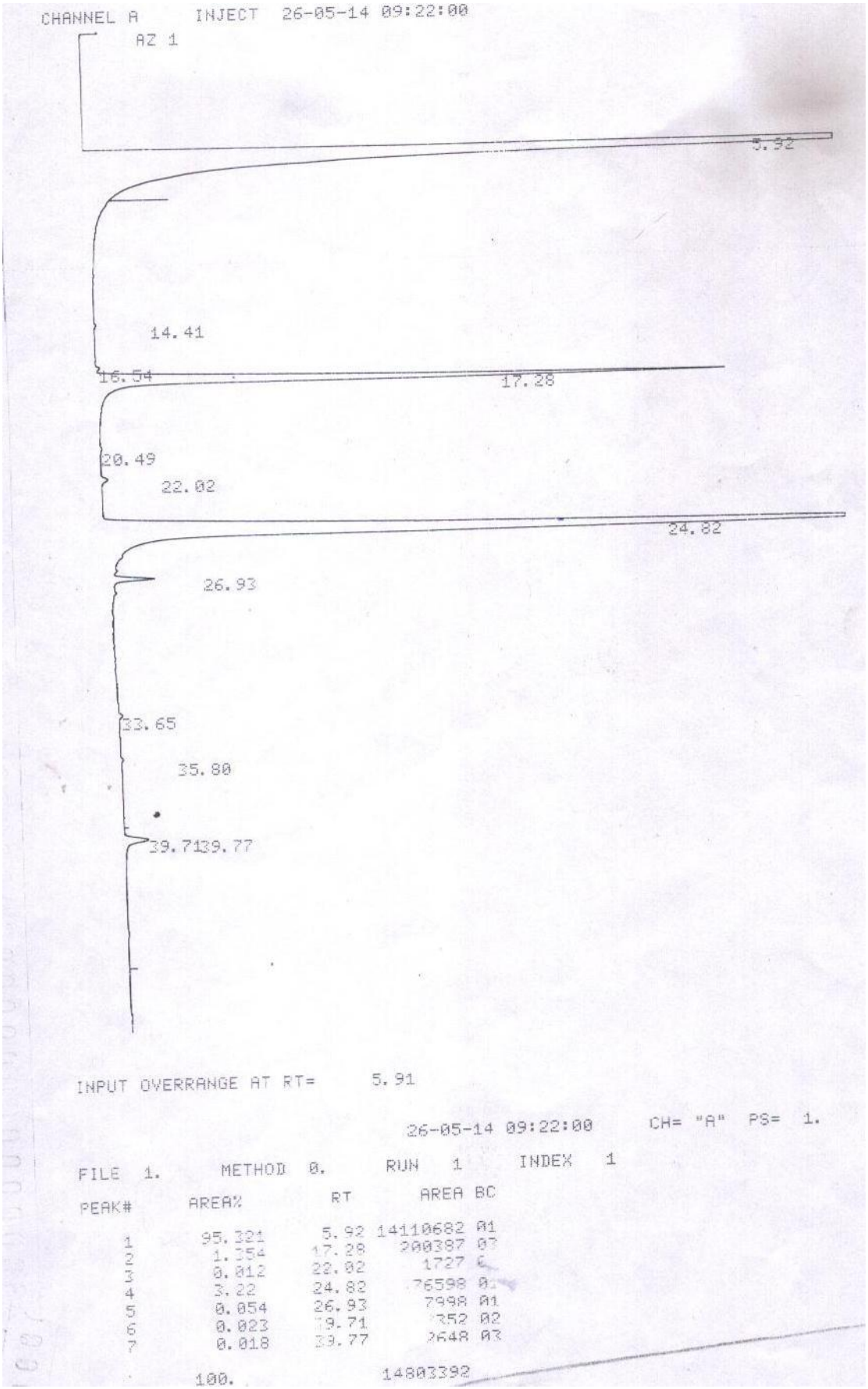
Appendix 20: Lauric acid + I.S



Appendix 21: Decanoic acid + I.S



Appendix 22: Linolenic acid + I S



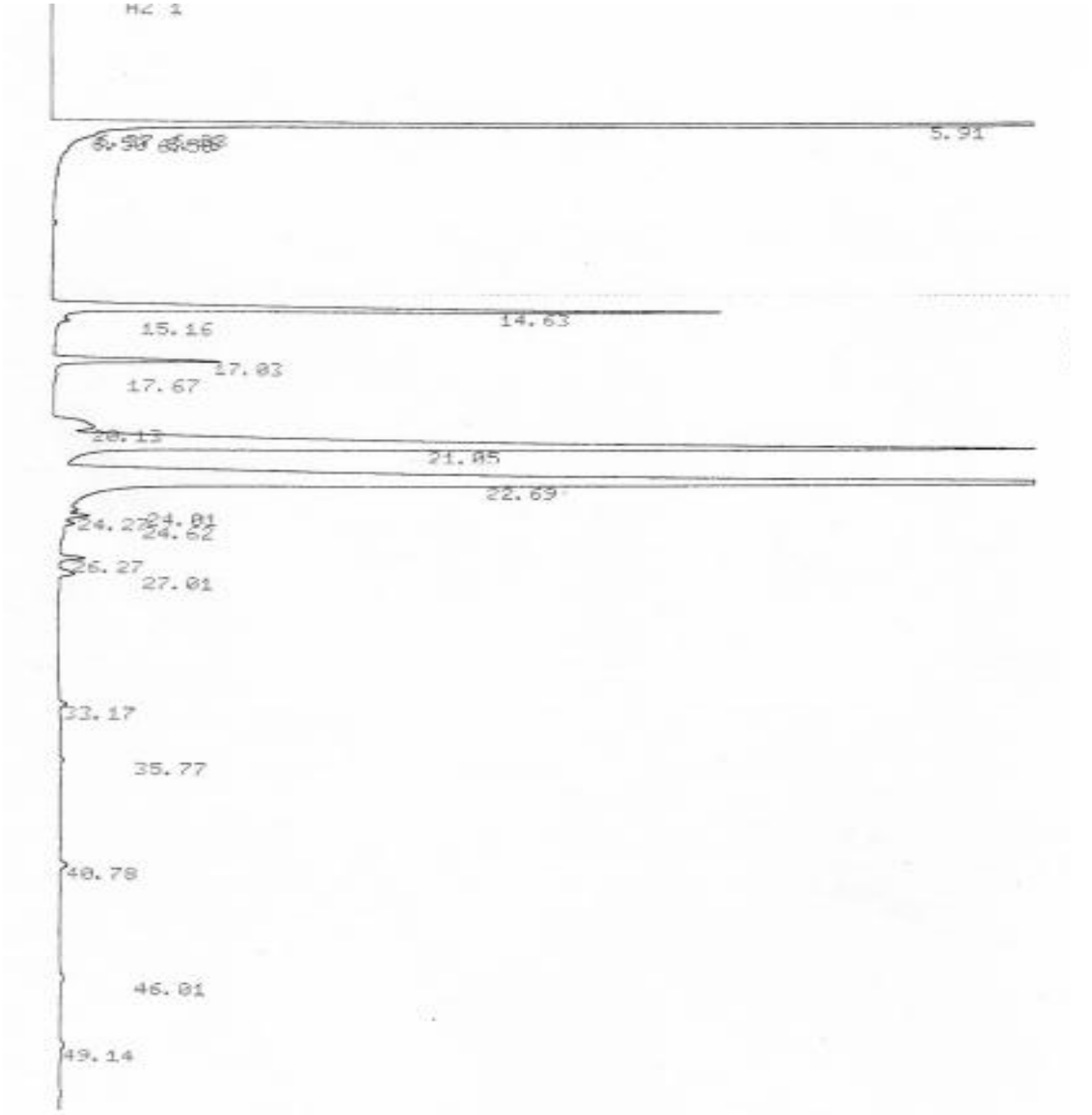
INPUT OVERRANGE AT RT= 5.91

26-05-14 09:22:00 CH= "A" PS= 1.

FILE 1. METHOD 0. RUN 1 INDEX 1

PEAK#	AREA%	RT	AREA BC
1	95.321	5.92	14110682 01
2	1.354	17.28	200387 07
3	0.012	22.02	1727 [illegible]
4	3.22	24.82	76598 01
5	0.054	26.93	7998 01
6	0.023	39.71	352 02
7	0.018	39.77	2648 03
	100.		14803392

Appendix 23: Sample 2



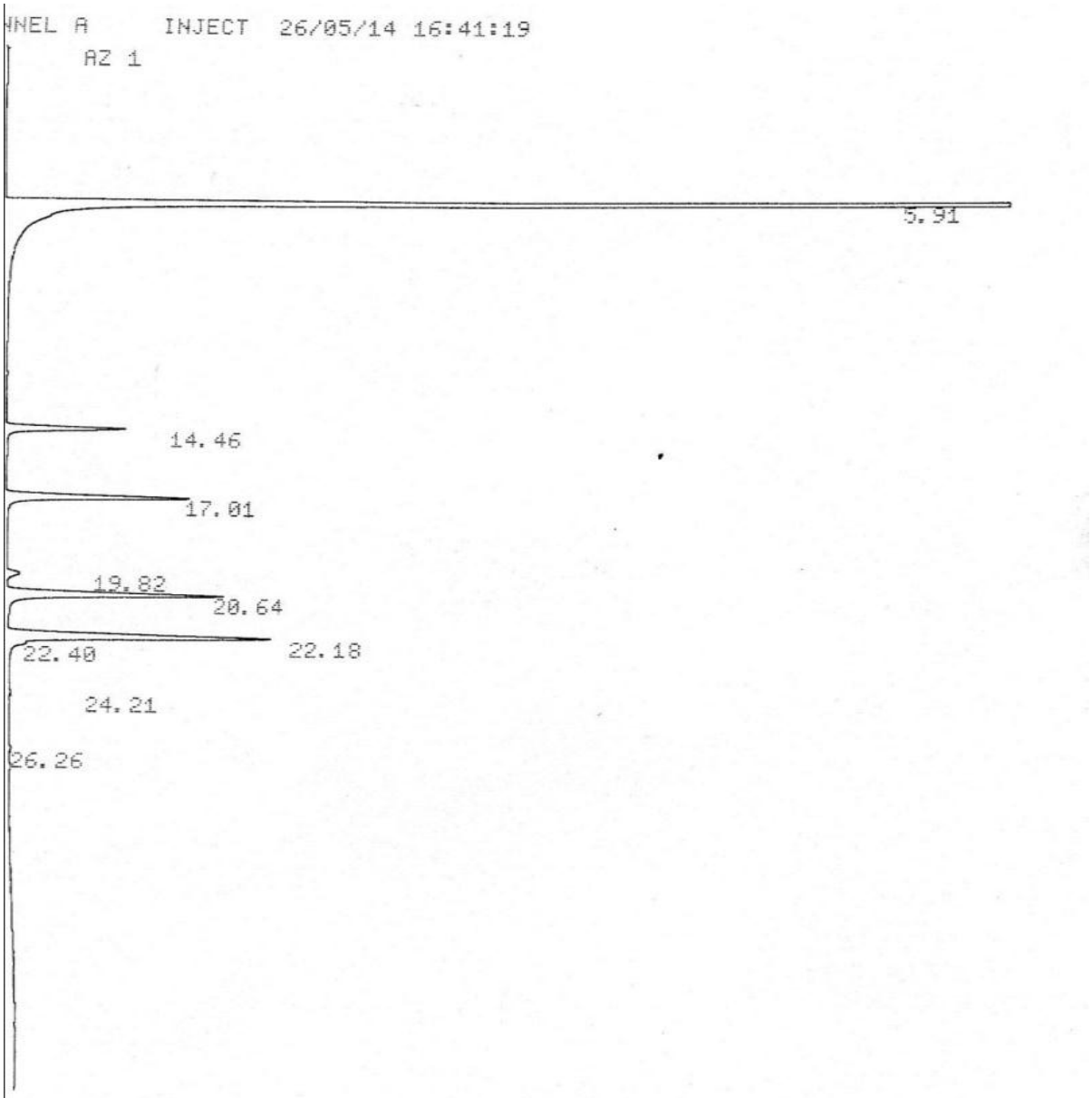
INPUT OVERRANGE AT RT= 5.91

26/05/14 15:24:06 CH= "A" PS= 1.

FILE 1. METHOD 0. RUN 2 INDEX 2

PEAK#	AREA%	RT	AREA	BC
1	92.862	5.91	13908097	02
2	0.012	6.33	1807	02
3	0.03	6.37	4451	02
4	0.018	6.5	2726	02
5	0.132	6.58	19785	03
6	0.787	14.63	117912	01
7	0.188	17.03	28161	01
8	0.121	20.13	18151	02
9	2.366	21.05	354334	02
10	3.396	22.69	508655	00
11	0.007	24.01	1022	06
12	0.02	24.27	2977	07
13	0.027	26.27	4043	01
14	0.025	27.01	3688	01

Appendix 24: Sample 3



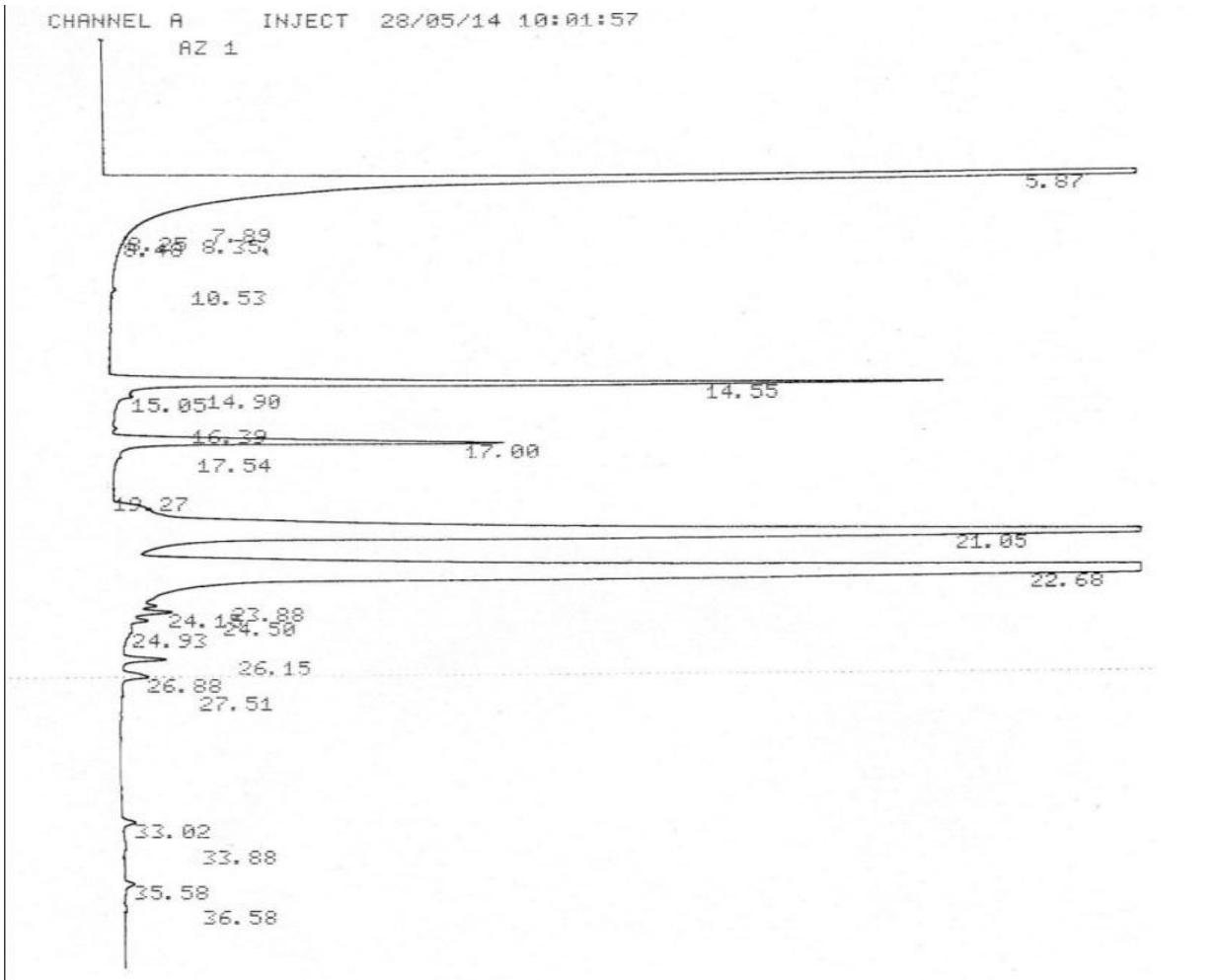
UT OVERRANGE AT RT= 5.91

26/05/14 16:41:19 CH= "A" PS= 1.

E 1. METHOD 0. RUN 3 INDEX 3

K#	AREA%	RT	AREA	BC
1	99.087	5.91	14081871	01
2	0.104	14.46	14723	01
3	0.186	17.01	26403	01
4	0.013	19.82	1850	01
5	0.268	20.64	38096	01
6	0.329	22.18	46700	02
7	0.014	22.4	1919	03
AL	100.		14211562	

Appendix 25: sample 5



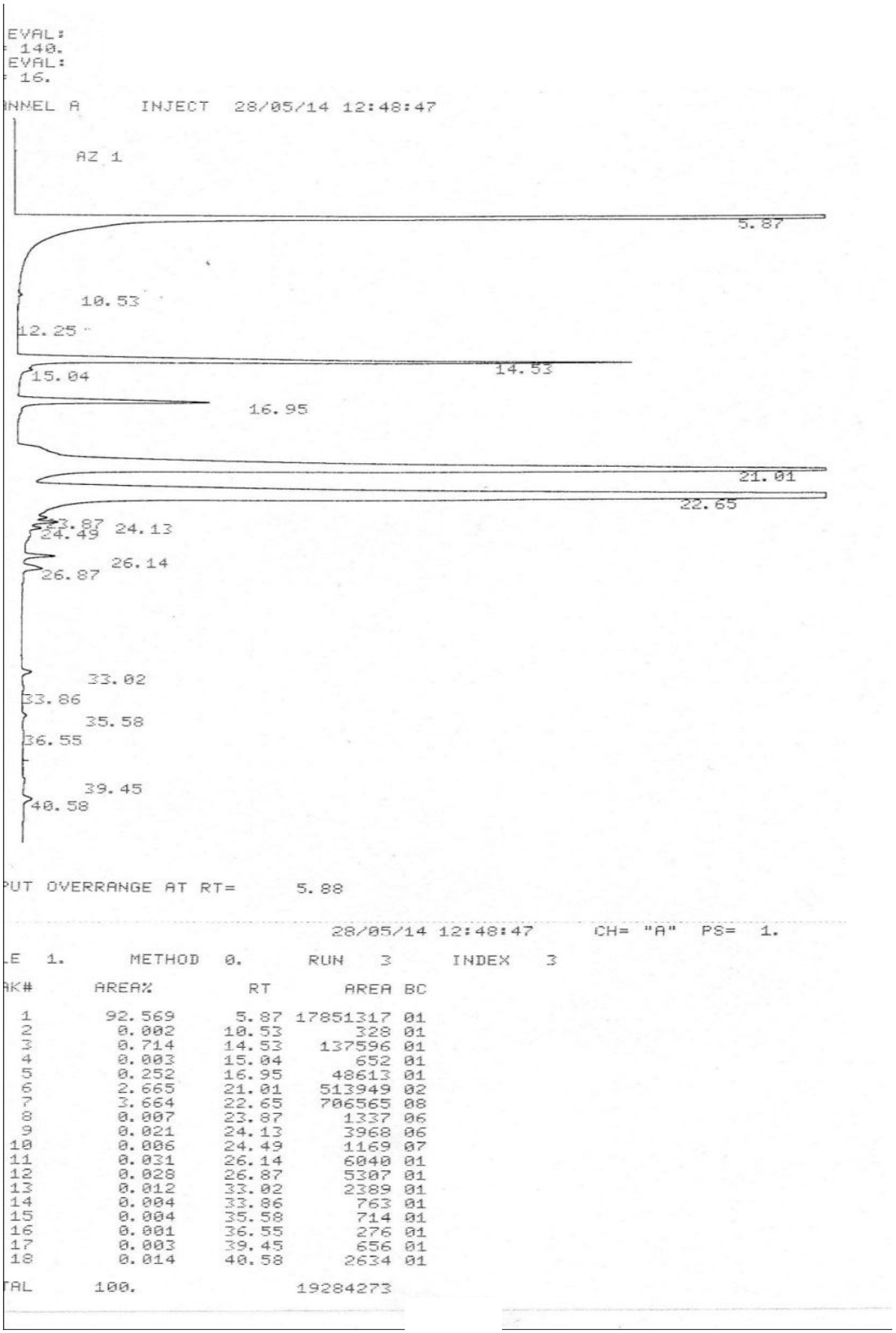
INPUT OVERRANGE AT RT= 5.88

28/05/14 10:01:57 CH= "A" PS= 1.

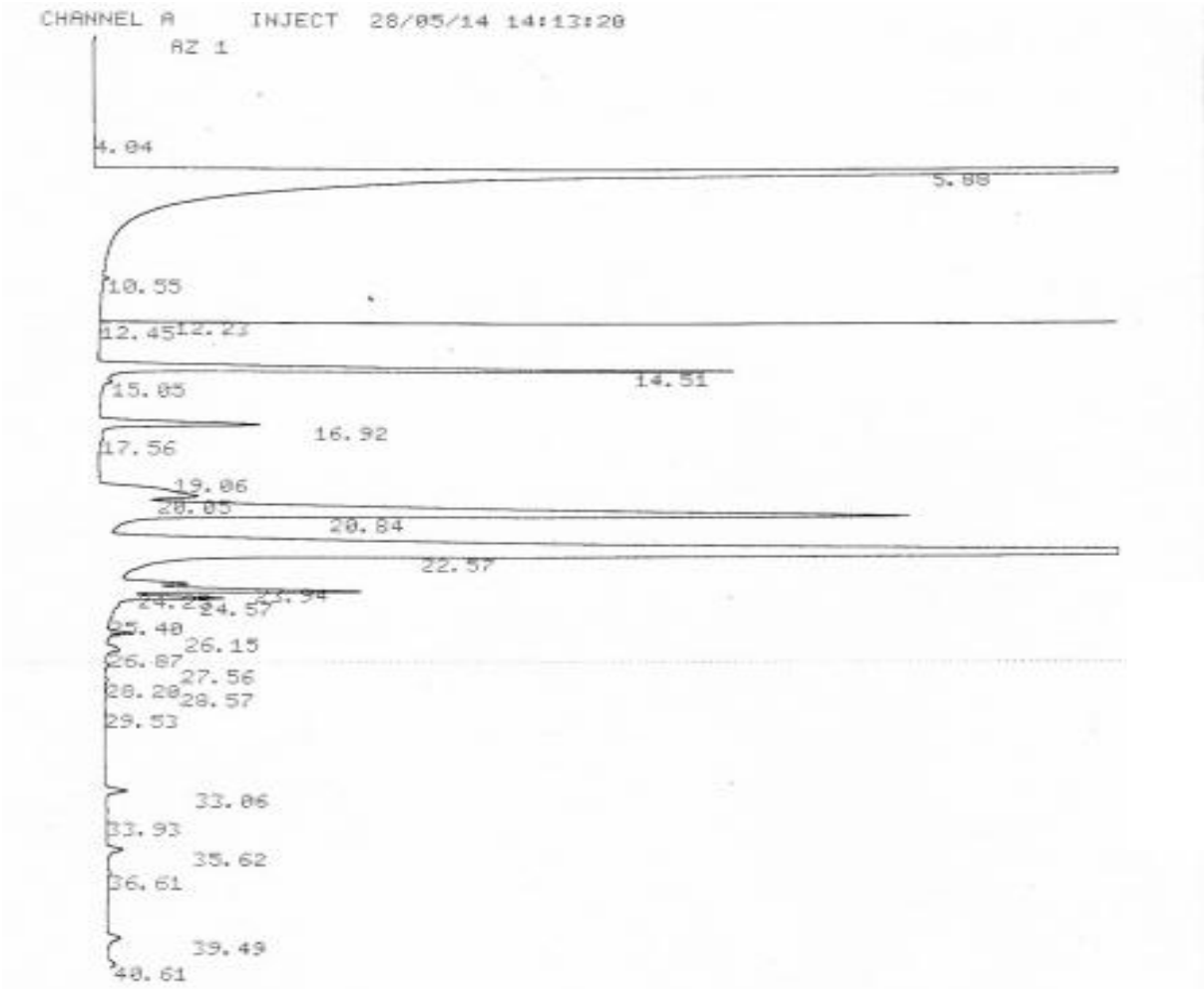
FILE 1. METHOD 0. RUN 1 INDEX 1

PEAK#	AREA%	RT	AREA	BC
1	91.457	5.87	17601370	02
2	0.02	7.89	3780	02
3	0.002	8.25	469	02
4	0.002	8.35	438	03
5	0.002	10.53	386	01
6	0.826	14.55	159051	09
7	0.003	15.05	661	05
8	0.001	16.39	151	01
9	0.425	17.	81727	08
10	0.001	17.54	267	05
11	2.971	21.05	571744	02
12	4.159	22.68	800397	08
13	0.007	23.88	1425	06
14	0.022	24.15	4275	06
15	0.009	24.5	1784	06
16	0.002	24.93	364	07
17	0.033	26.15	6409	01
18	0.031	26.88	5914	01
19	0.001	27.51	214	01
20	0.012	33.02	2309	01
21	0.003	33.88	498	01
22	0.009	35.58	1658	01
23	0.001	36.58	240	01
TOTAL	100.		19245531	

Appendix 26: Sample 6



Appendix 27: Sample 7



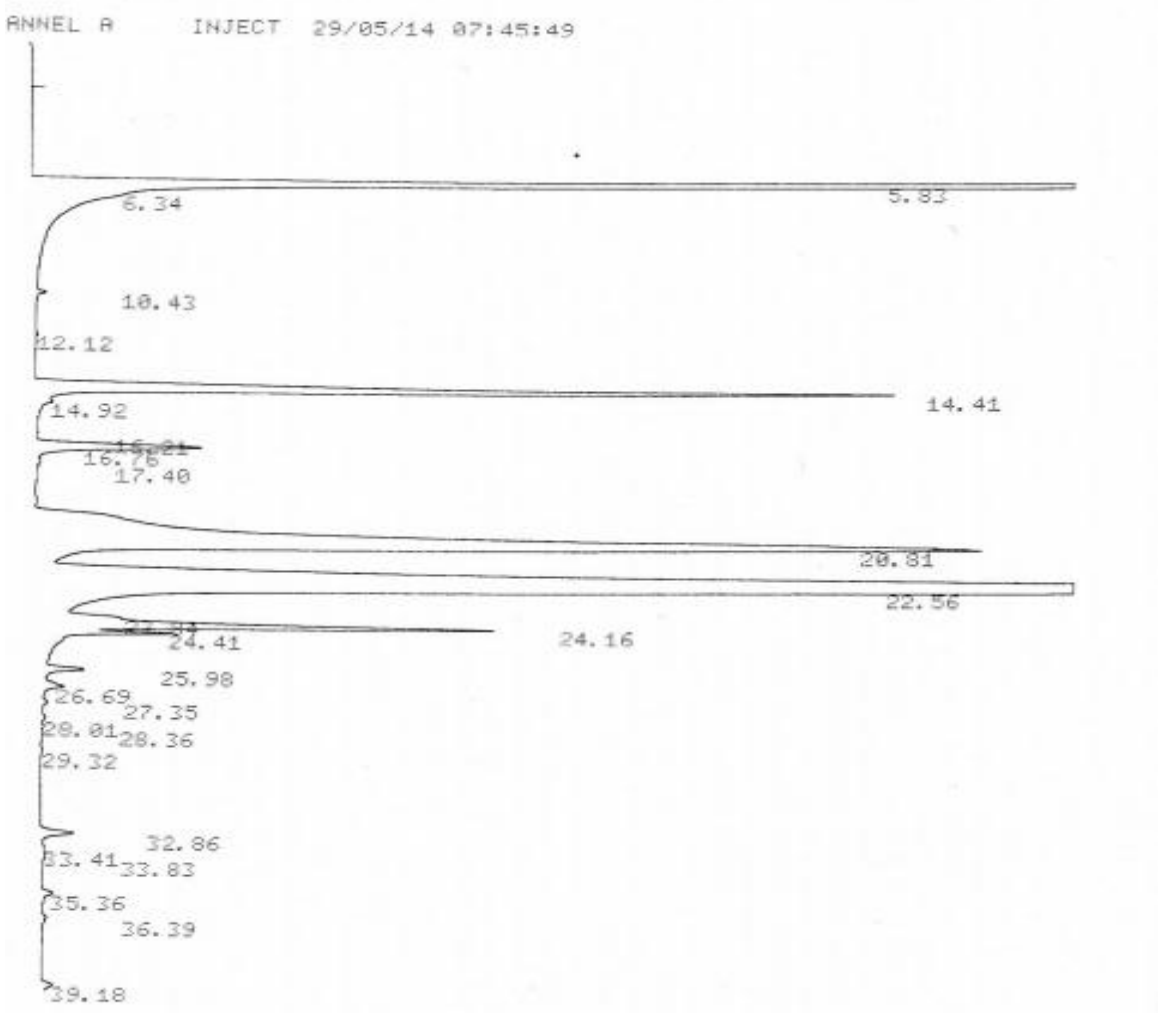
INPUT OVERRANGE AT RT= 5.89

28/05/14 14:13:20 CH= "A" PS= 1.

FILE 1. METHOD 0. RUN 4 INDEX 4

PEAK#	AREA%	RT	AREA	BC
1	0.003	4.04	633	01
2	93.645	5.88	18240774	01
3	0.003	10.55	575	01
4	0.001	12.23	132	02
5	0.019	12.45	3623	03
6	0.547	14.51	106484	01
7	0.003	15.85	490	01
8	0.148	16.92	28868	01
9	0.002	17.56	320	01
10	0.001	19.06	219	02
11	0.217	20.05	42280	02
12	1.499	20.84	291985	02
13	3.491	22.57	680016	08
14	0.054	23.94	10533	06
15	0.195	24.25	37929	06
16	0.071	24.57	13805	07
17	0.001	25.4	130	05
18	0.021	26.15	4007	06
19	0.018	26.87	3458	06
20	0.003	27.56	607	06
21	0.003	28.2	615	06
22	0.001	28.57	143	07
23	0.002	29.53	336	01
24	0.019	33.06	3742	01
25	0.001	33.93	200	01
26	0.015	35.62	2902	01
27	0.002	36.61	436	01
28	0.014	39.49	2731	01
29	0.003	40.61	667	01

Appendix 28: Sample 12



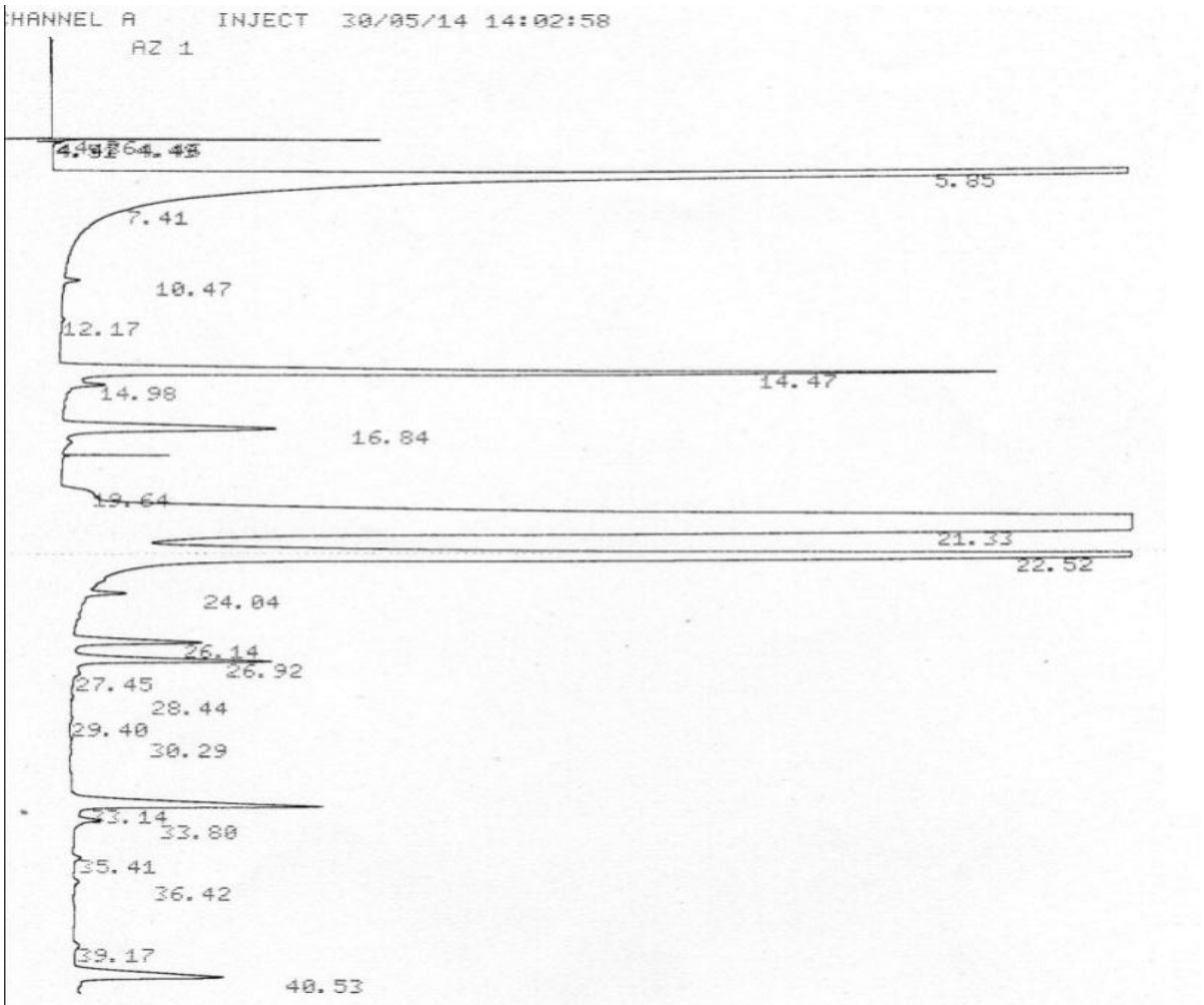
PUT OVERRANGE AT RT= 5.83

29/05/14 07:45:49 CH= "A" PS= 1.

LE 1. METHOD 0. RUN 5 INDEX 5

AK#	AREA%	RT	AREA	BC
1	91.299	5.83	17644972	02
2	0.202	6.34	39020	03
3	0.005	10.43	895	01
4	0.001	12.12	202	01
5	0.796	14.41	153867	01
6	0.003	14.92	542	01
7	0.001	16.21	176	02
8	0.191	16.76	36926	08
9	0.002	17.4	371	05
10	2.354	20.81	455006	02
11	4.544	22.56	878103	03
12	0.04	23.84	7748	02
13	0.383	24.16	74064	02
14	0.069	24.41	13265	03
15	0.027	25.98	5278	01
16	0.02	26.69	3944	01
17	0.003	27.35	621	01
18	0.003	28.01	571	02
19	0.002	28.36	355	03
20	0.001	29.32	207	01
21	0.03	32.86	5803	01
22	0.003	33.83	493	01
23	0.01	35.36	1904	01
24	0.003	36.39	661	01
25	0.008	39.18	1567	01
TAL	100.		19326561	

Appendix 29: Sample 15



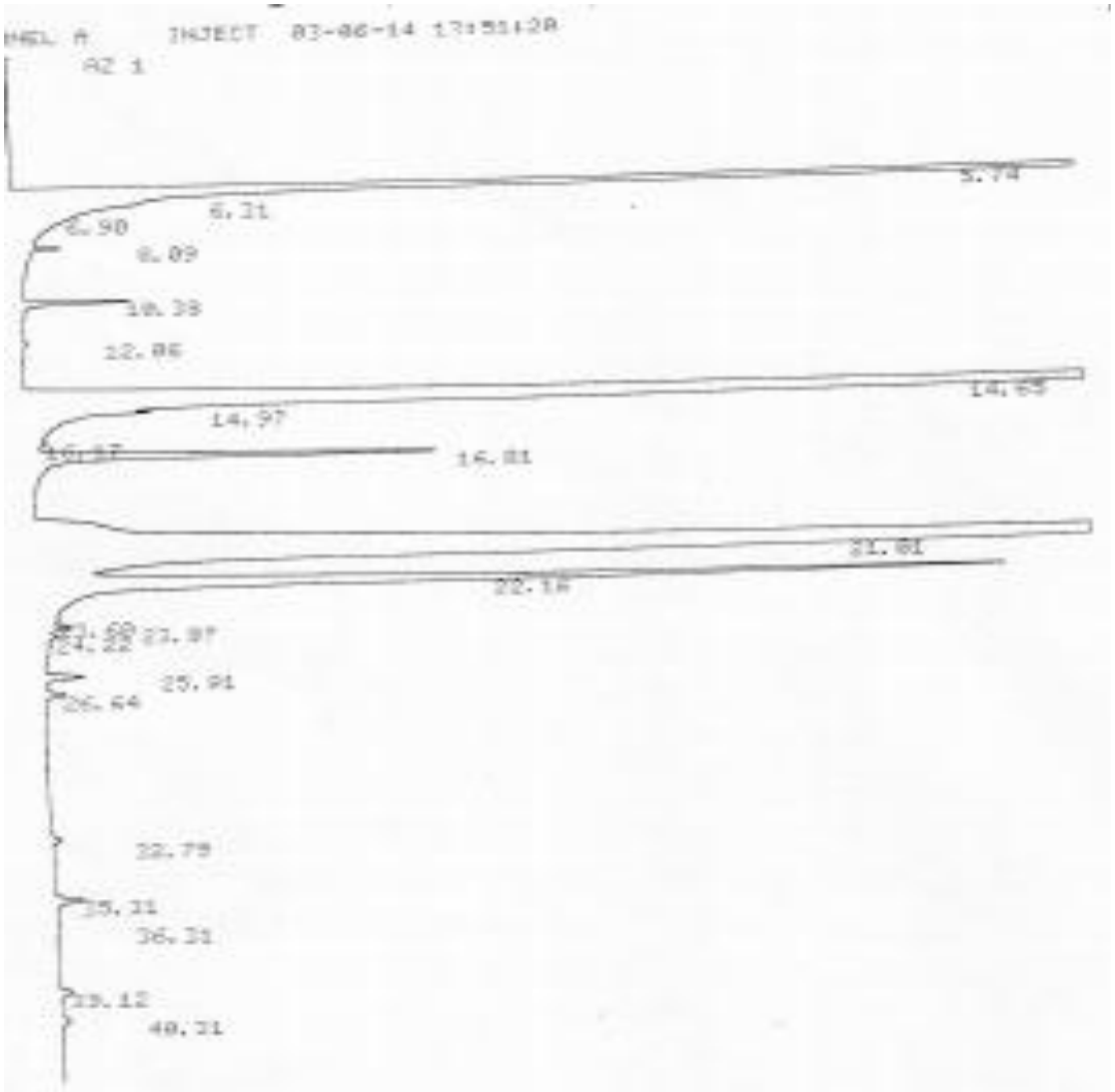
INPUT OVERRANGE AT RT= 5.85

30/05/14 14:02:58 CH= "A" PS= 1.

FILE 1. METHOD 0. RUN 3 INDEX 3

PEAK#	AREA%	RT	AREA	BC
1	0.056	4.36	11377	02
2	0.004	4.46	754	02
3	0.006	4.47	1268	02
4	0.003	4.49	710	02
5	0.01	4.51	2122	03
6	88.369	5.85	18096554	09
7	0.008	10.47	1611	01
8	0.002	12.17	400	01
9	0.843	14.47	172677	08
10	0.018	14.98	3624	05
11	0.238	16.84	48734	01
12	0.059	19.64	12074	02
13	7.282	21.33	1491263	02
14	2.328	22.52	476776	03
15	0.02	24.04	4097	01
16	0.105	26.14	21597	01
17	0.156	26.92	31847	01
18	0.004	27.45	815	01
19	0.002	28.44	309	01
20	0.002	29.4	439	01
21	0.001	30.29	261	01
22	0.272	33.14	55651	02
23	0.03	33.8	6167	03
24	0.006	35.41	1168	01
25	0.005	36.42	967	01
26	0.005	39.17	1028	01
27	0.166	40.53	34064	01
TOTAL	100.		20478354	

Appendix 30: Sample 18



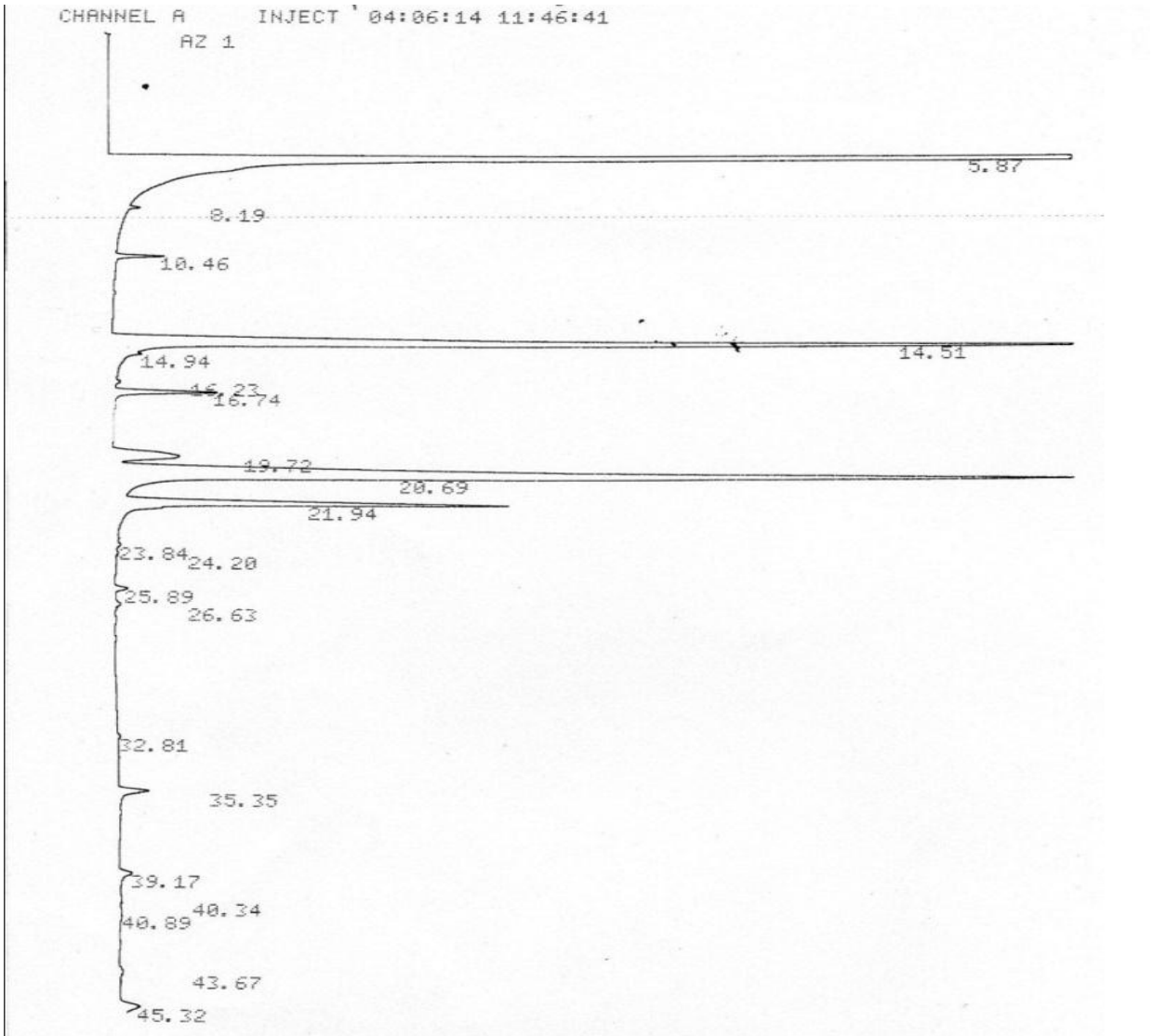
INPUT OVERRANGE AT RT= 5.74

83-06-14 13:51:20 CH= "A" FS= 1.

FILE 1- METHOD 0. RUN 5 INDEX 5

PEAK#	AREA%	RT	AREA	BC
1	89.71	5.74	185119717	89
2	0.011	6.89	2283	01
3	0.061	10.33	12677	01
4	0.002	12.86	422	01
5	3.466	14.65	715478	88
6	0.009	14.97	1947	05
7	0.002	16.17	486	01
8	0.357	16.81	73786	01
9	5.184	21.81	1070891	88
10	1.086	22.16	224248	05
11	0.002	23.6	485	02
12	0.011	23.87	2256	03
13	0.003	24.22	516	01
14	0.028	25.91	5782	01
15	0.016	26.64	3311	01
16	0.009	32.79	1885	01
17	0.021	35.31	4379	01
18	0.002	36.21	356	01
19	0.011	39.12	2271	01
20	0.009	40.31	1873	01
TOTAL	100.		206440803	

Appendix 31: Sample 22



INPUT OVERRANGE AT RT= 5.87

04:06:14 11:46:41 CH= "A" PS= 1.

FILE 1. METHOD 0. RUN 3 INDEX 3

PEAK#	AREA%	RT	AREA	BC
1	96.252	5.87	18281813	01
2	0.004	8.19	831	01
3	0.025	10.46	4814	01
4	1.273	14.51	241819	08
5	0.004	14.94	680	05
6	0.002	16.23	412	01
7	0.077	16.74	14622	01
8	0.125	19.72	23800	02
9	1.727	20.69	327941	08
10	0.417	21.94	79263	05
11	0.002	23.84	454	01
12	0.001	24.2	198	01
13	0.011	25.89	2176	01
14	0.007	26.63	1303	01
15	0.002	32.81	471	01
16	0.031	35.35	5864	01
17	0.013	39.17	2458	01
18	0.002	40.34	416	01
19	0.002	40.89	314	01
20	0.004	43.67	769	01
21	0.018	45.32	3330	01
TOTAL	100.		18993748	

Appendix 33: Publication