

Fatty acids profile of Edible Oils and Fats in India

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About the CSE Laboratory

The Centre for Science and Environment (CSE), a non-governmental organization based in New Delhi, has set up the Pollution Monitoring Laboratory (PML) to monitor environmental pollution. PML is an ISO 9001:2000 certified laboratory accredited by SWISO, Switzerland, conducting Pollution Monitoring and Scientific Studies on Environmental Samples. The Lab has qualified and experienced staff who exercise Analytical Quality Control (AQC) and follow Good Laboratory Practices. It is equipped with state-of-art equipments for monitoring and analysis of air, water and food contamination, including Gas Chromatograph with Mass Spectrometer (GC-MS), Gas Chromatograph (GC) with ECD, NPD, FID and other detectors, High Performance Liquid Chromatograph (HPLC), Atomic Absorption Spectrometer, UV-VIS Spectrophotometer, Mercury Analyzer, Respirable Dust Sampler etc. Its main aim is to undertake scientific studies to generate public awareness about food, water and air contamination. It provides scientific services at nominal cost to communities that cannot obtain scientific evidence against polluters in their area. This is an effort to use science to achieve ecological security.

2. Introduction & Origin of the study

Fats and oils are recognized as essential nutrients in both human and animal diets. They provide the most concentrated source of energy of any foodstuff, supply essential fatty acids (which are precursors for important hormones, the prostaglandins), contribute greatly to the feeling of satiety after eating, are carriers for fat soluble vitamins, and serve to make foods more palatable. Fats and oils are present in varying amounts in many foods. The principal sources of fat in the diet are vegetable fats and oils, meats, dairy products, poultry, fish and nuts. Most vegetables and fruits consumed as such contain only small amounts of fat.

Fatty acids are the building blocks of lipids and generally comprise 90% of the fats in foods. These are compounds that are of interest when reporting lipid content labeling of fats and Oils. Saturated fatty acids – hydrocarbon chains with single bonds between each of carbon atoms – found primarily in products derived from animal sources (meat, dairy products) tend to raise the levels of low density lipoprotein (LDL) cholesterol in the blood. Unsaturated fatty acids – characterized by one (monounsaturated) or more (polyunsaturated) double bonds in the carbon chain – are found mostly in plants and sea food. Since the carbons are double-bonded to each other, there are fewer bonds available for hydrogen, so there are fewer hydrogen atoms, hence "unsaturated". *Cis* and *trans* are terms that refer to the arrangement of chains of carbon atoms across the double bond. In the *cis* arrangement, the chains are on the same side of the double bond, resulting in a kinked geometry. In the *trans* arrangement, the chains are on opposite sides of the double bond, and the chain is straight overall.

Typically, common vegetable oils, including soybean, sunflower, safflower, mustard, olive, rice bran, sesame are low in saturated fats and the double bonds within unsaturated acids are in the *cis* configuration. To improve their oxidative stability and to increase their melting points, vegetable oils are hydrogenated. The process of hydrogenation is intended to add hydrogen atoms to *cis*-unsaturated fats, eliminating a double bond and making them more saturated. Full hydrogenation would produce exclusively saturated fatty acids that are too waxy and solid to use in food production. Consequently, the process used by the industry does not eliminate all of the double

bonds and is called partial hydrogenation. Partially hydrogenated oils give foods a longer shelf life and more stable flavor. However, the process frequently has a side effect that turns some *cis*-isomers into *trans*-unsaturated fats instead of hydrogenating them. The formation of *trans* fatty acids also occurs during deodorization step of processing; it is usually carried out at temperatures ranging from 180°C to 270°C.

Human intake of *trans* fatty acids originates from foods containing industrially produced, partially hydrogenated fat, and from beef, lamb and dairy products. The majority of *trans* fat in our diet is industrially produced. It is consumed primarily as *vanaspati*, shortening and margarine, or in foods that are baked or fried using these substances, such as cakes, cookies, bread, potato chips, french fries and other fried products. The *trans* fatty acids have unfavorable effect on serum lipoprotein profiles. *Trans* fatty acids increased low density lipoprotein cholesterol levels associated with increased risk for cardiovascular and cerebrovascular diseases. They also decrease high density lipoprotein cholesterol levels (Stender and Dyerberg, 2003)

There is a mounting concern about the intake of foods containing *trans* fatty acids due to their deleterious effects on humans, although monounsaturated fatty acid is the main *trans* group ingested by most people, the presence of polyunsaturated *trans* fatty acids in significant levels has also been investigated in foods containing partially hydrogenated fats, fried food and refined oils.

The objective of this study was to determine total saturated, unsaturated and *trans* fatty acids in edible oil and fat samples widely consumed in India. PML tested the refined edible oils, *vanaspati*, *desi ghee* and butter samples with a widely and internationally used methodology of Association of Official Analytical Chemists (AOAC) for fatty acids analysis. Fatty acid methyl ester preparation was done as per the given methodology and analysis was done by Gas Chromatograph with Flame Ionization Detector (GC-FID) using a fused silica capillary column coated with a highly polar stationary phase.

3. What is a Fat or Oil?

Fats consist of a wide group of compounds that are soluble in organic solvents and insoluble in water. They have lower densities than water and at normal room temperature range in consistency from liquids to solids, depending on their structure and composition. Chemically, fats are generally triesters of glycerol and fatty acids. Although the words "oils", "fats" and "lipids" are all used to refer to fats, "oils" is usually used to refer to fats that are liquids at normal room temperature, while "fats" is usually used to refer to fats that are solids at normal room temperature. "Lipids" is used to refer to both liquid and solid fats (Anthea, 1993).

Triacylglycerols are the predominant components of most food fats and oils. The minor components include mono and diacylglycerols, free fatty acids, phosphatides, sterols, fatty alcohols, fat-soluble vitamins, and other substances (Strayer, 2006).

3.1. The Major Component – Triacylglycerols

A triacyl glycerol is composed of glycerol and three fatty acids. When all of the fatty acids in a triacylglycerol are identical, it is termed a "simple" triacylglycerol. The more common forms, however, are the "mixed" triacylglycerols in which two or three kinds of fatty acids are present in the molecule.

3.2. The Minor Components

3.2.1 Mono and Diacylglycerols

Mono and diacyl glycerols are mono and diesters of fatty acids and glycerol. They are prepared commercially by the reaction of glycerol and triacyl glycerols or by the esterification of glycerol and fatty acids. They also occur naturally in very minor amounts in both animal fats and vegetable oils.

3.2.2 Free Fatty Acids

Free fatty acids are the unattached fatty acids present in a fat. Some unrefined oils may contain as much as several percent free fatty acids. The levels of free fatty acids are reduced in the refining.

3.2.3. Phosphatides

Phosphatides consist of alcohols (usually glycerol), combined with fatty acids, phosphoric acid, and a nitrogen-containing compound. Lecithin and cephalin are common phosphatides found in edible fats. Refining removes the phosphatides from the fat or oil.

3.2.4 Sterols

Sterols, also referred to as steroid alcohols, are a class of substances that contain the common steroid nucleus plus an 8 to 10 carbon side chain and an alcohol group found in both animal fats and vegetable oils. Cholesterol is the primary animal fat sterol and is only found in vegetable oils in trace amounts. Vegetable oil sterols collectively are termed "phytosterols". Sitosterol and stigmasterol are the best-known vegetable oil sterols.

3.2.5 Fatty Alcohols

Long chain alcohols are of little importance in most edible fats. A small amount esterified with fatty acids is present in waxes found in some vegetable oils. Larger quantities are found in some marine oils.

3.2.6 Tocopherols

Tocopherols are important minor constituents of most vegetable fats described as series of organic compounds consisting of methylated phenols. They serve as antioxidants to retard rancidity and as sources of the essential nutrient vitamin E. Among tocopherols, alpha-tocopherol has the highest vitamin E activity and the lowest antioxidant activity.

3.2.7 Carotenoids and Chlorophyll

Carotenoids are color materials occurring naturally in fats and oils ranging from yellow to deep red. Chlorophyll is the green coloring matter of plants which plays an essential part in the photosynthetic process. The levels of most of these color materials are reduced during the normal processing and refining of oils.

3.2.8 Vitamins

Most fats and oils are not good sources of vitamins other than vitamin E. The fat-soluble vitamins A and D are sometimes added to foods which contain fat because they serve as good carriers and are widely consumed.

4. Fatty Acids

Triacylglycerols are comprised predominantly of fatty acids present in the form of esters of glycerol. One hundred grams of fat or oil will yield approximately 95 grams of fatty acids. Both the physical and chemical characteristics of fats are influenced greatly by the kinds and proportions of the component fatty acids and the way in which these are positioned on the glycerol molecule. The predominant fatty acids are saturated and unsaturated carbon chains with an even number of carbon atoms and a single carboxyl group as illustrated in the general structural formula for a saturated fatty acid given below:



Edible oils also contain minor amounts of branched chain and cyclic acids. Also odd number straight chain acids are typically found in animal fats.

4.1. Classification of Fatty Acids

Fatty acids occurring in edible fats and oils are classified according to their degree of saturation.

4.1.1. Saturated Fatty Acids

Those containing only single carbon-to-carbon bonds are termed “saturated” and are the least reactive chemically. The melting point of saturated fatty acids increases with chain length. All but acetic acid occur naturally in fats. Decanoic and longer chain fatty acids are solids at normal room temperatures (see Table 1).

Table 1 Saturated Fatty Acids in different oils

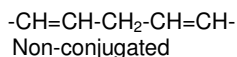
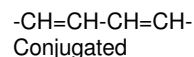
Systematic Name	Common Name	No. of Carbon Atoms*	Melting Point °C	Typical Fat Source
Ethanoic	Acetic	2	--	--
Butanoic	Butyric	4	-7.9	Butterfat
Hexanoic	Caproic	6	-3.4	Butterfat
Octanoic	Caprylic	8	16.7	Coconut oil
Decanoic	Capric	10	31.6	Coconut oil
Dodecanoic	Lauric	12	44.2	Coconut oil
Tetradecanoic	Myristic	14	54.4	Butterfat, coconut oil
Hexadecanoic	Palmitic	16	62.9	Most fats and oils
Heptadecanoic	Margaric	17	60	Animal fats
Octadecanoic	Stearic	18	69.6	Most fats and oils
Eicosanoic	Arachidic	20	75.4	Peanut oil
Docosanoic	Behenic	22	80.0	Peanut oil

Source: Strayer , 2006

*A number of saturated odd and even chain acids are present in trace quantities in many fats and oils.

4.1.2 Unsaturated Fatty Acids

Fatty acids containing one or more carbon-to-carbon double bonds are termed “unsaturated.” Oleic acid (*cis*-9-octadecenoic acid) is the fatty acid that occurs most frequently in nature. When the fatty acid contains one double bond it is called Monounsaturated Fatty Acids (MUFA). If it contains more than one double bond, it is called Polyunsaturated Fatty Acids (PUFA). Because of the presence of double bonds, unsaturated fatty acids are more reactive chemically than are saturated fatty acids. This reactivity increases as the number of double bonds increases. Although double bonds normally occur in a non-conjugated position, they can occur in a conjugated position (alternating with a single bond) as illustrated below:



With the bonds in a conjugated position, there is a further increase in certain types of chemical reactivity. For example, fats are much more subject to oxidation and polymerization when bonds are in the conjugated position (see Table 2).

Table 2 Some Unsaturated Fatty Acids In Food Fats And Oils

Systematic Name	Common Name	No. of Double Bonds	No. of Carbon Atoms	Melting Point °C	Typical Fat Source
9-Decenoic	Caproic	1	10	-	Butterfat
9-Dodecenoic	Lauroic	1	12	-	Butterfat
9-Tetradecenoic	Myristoleic	1	14	18.5	Butterfat
9-Hexadecenoic	Palmitoleic	1	16	-	Some fish oils, beef fat
9-Octadecenoic	Oleic	1	18	16.3	Most fats and oils
9-Octadecenoic*	Elaidic	1	18	43.7	Partially hydrogenated oils
11-Octadecenoic*	Vaccenic	1	18	44	Butterfat
9,12-Octadecadienoic	Linoleic	2	18	-6.5	Most vegetable oils
9,12,15-Octadecatrienoic	Linolenic	3	18	-12.8	Soybean oil, canola oil
9-Eicosenoic	Gadoleic	1	20	-	Some fish oils
5,8,11,14-Eicosatetraenoic	Arachidonic	4	20	-49.5	Lard
5,8,11,14,17-Eicosapentaenoic	-	5	20	-	Some fish oils
13-Docosenoic	Erucic	1	22	33.4	Rapeseed oil
4, 7, 10, 13, 16, 19-Docosahexaenoic acid	-	6	22	-	Some fish oils

Source: Strayer, 2006

*All double bonds are in the *cis* configuration except for elaidic acid and vaccenic acid which are *trans*.

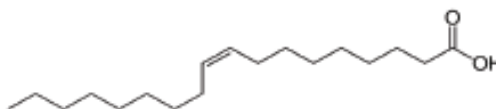
Of the polyunsaturated fatty acids, linoleic, linolenic, arachidonic, eicosapentaenoic, and docosahexaenoic acids containing respectively two, three, four, five, and six double bonds are of most interest. Vegetable oils are the principal sources of linoleic and linolenic acids. Arachidonic acid is found in small amounts in lard, which also contains about 10% of linoleic acid. Fish oils contain large quantities of a variety of longer chain fatty acids having three or more double bonds including eicosapentaenoic and docosahexaenoic acids.

4.2 Isomerism of Unsaturated Fatty Acids

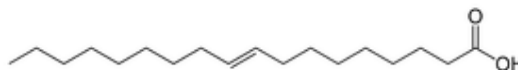
Isomers are two or more substances composed of the same elements combined in the same proportions but differing in molecular structure. The two important types of isomerism among fatty acids are (1) geometric and (2) positional.

4.2.1 Geometric Isomerism

Unsaturated fatty acids can exist in either the *cis* or *trans* form depending on the configuration of the hydrogen atoms attached to the carbon atoms joined by the double bonds. If the hydrogen atoms are on the same side of the carbon chain, the arrangement is called *cis*. If the hydrogen atoms are on opposite sides of the carbon chain, the arrangement is called *trans*, as shown by the following diagrams. Conversion of *cis* isomers to corresponding *trans* isomers result in an increase in melting points as shown in Table 2. The lower the melting point, the healthier the oil.



Oleic acid, a *cis* unsaturated fatty acid



Elaidic acid, a *trans* unsaturated fatty acid

Elaidic and oleic acids are geometric isomers; in the former, the double bond is in the *trans* configuration and in the latter, in the *cis* configuration. *Trans* isomers occur naturally in ruminant animals such as cows, sheep and goats

4.2.2 Positional Isomerism

In this case, the location of the double bond differs among the isomers. Petroselinic acid, which is present in parsley seed oil, is *cis*-6-octadecenoic acid and a positional isomer of oleic acid. *Cis*-isomers are those naturally occurring in food fats and oils. Vaccenic acid, which is a minor acid in tallow and butterfat, is *trans*-11-octadecenoic acid and is both a positional and geometric isomer of oleic acid.

5. Essential fatty acids

Fat is an important ingredient of human diet. The functional role of fat in the diet is manifold. Fat is a most concentrated source of energy. Fats are essential in the diet for the absorption and mobilization of fat-soluble vitamins such as vitamin A, vitamin E and fat-soluble antioxidants. These vitamins are not utilized by the body if fat is not available in the diet. Thus fat works as a vehicle to carry the fat-soluble vitamins, nutrients and antioxidants in the body.

Vegetable oils are the main sources of essential fatty acids for the body. Essential fatty acids (EFA) are those fatty acids, which the body cannot synthesize and need to be supplied through diet. It also helps in raising High density lipoproteins (HDL) the so-called good cholesterol. Low-fat diets can result in reduction of HDL cholesterol. Fat in the diet imparts certain textural qualities, taste and palatability to the food.

EFAs are long-chain unsaturated fatty acids derived from linolenic (which is PUFA and is also called as Omega-3), linoleic (which is PUFA and is also called as Omega-6), and oleic acids (which is MUFA and is also called as Omega-9). Omega-9 is necessary yet “non-essential” because the body can manufacture a modest amount on its own, provided essential EFAs are present.

EFAs support the cardiovascular, reproductive, immune, and nervous systems. The human body needs EFAs to manufacture and repair cell membranes, enabling the cells to obtain optimum nutrition and expel harmful waste products. A primary function of EFAs is the production of prostaglandins, which regulate body functions such as heart rate, blood pressure, blood clotting, fertility, conception, and play a role in immune function by regulating inflammation and encouraging the body to fight infection. An ideal intake ratio of Omega-6 to Omega-3 fatty acids is between 5-10:1(Simpoulos, 2002).

EFA deficiency and Omega 6/Omega 3 imbalance is linked with serious health conditions, such as heart attacks, cancer, insulin resistance, asthma, lupus, schizophrenia, depression, postpartum depression, accelerated aging, stroke, obesity, diabetes, arthritis, ADHD, and Alzheimer’s Disease, among others (Simpoulos, 1999).

5.1 Recommendations on Fat Consumption

Several Countries have recommended allowances not only on the absolute values of PUFAs, but also the balanced intake of Omega 6 and Omega 3 PUFA.

The World Health Organization (WHO) recommends polyunsaturated fatty acid (PUFA)/saturated fatty acids (SFA) ratio of 0.8 to 1.0 and linoleic acid (Omega 6) alpha linolenic acid (Omega 3) ratio of 5-10 in the diet. (WHO/FAO, 2003)

The American Heart Association recommends total fat intake to less than 25–35 percent of total calories. A saturated fatty acid intake of not more than 10 percent of total calories, a monounsaturated fatty acid intake in the range of 10-15 percent and polyunsaturated fats upto 10 percent of total calories.

Table 3 Recommended Dietary allowance of fats in the world

Country	Total energy intake from fats ¹	Total PUFA ²	Omega 6	Omega 3	Omega 6/ Omega 3
WHO (1990)	15-30	3-7, ≤10	-	-	5-10
FAO (1994)	15-35	-	4-10	-	5-10
USA (1989)	<30	7	1-2	-	4-10
Japan (1995)	20-25	7-8	-	-	4
Canada (1990)	30 ²	≥3.5	≥3	≥0.5	4-10

(Source: Sugano and Hirahara, 2000)

Note :

1. Intake of saturated fat should be $\leq 10\%$ of energy
2. PUFA Polyunsaturated fatty acids

5.2 Which Oil is Healthy?

On the basis of these recommendations there are basically three parameters to adjudge any oil as healthy oil - ratio of saturated/ monounsaturated/ polyunsaturated fatty acid, ratio of essential fatty acids (Omega6/Omega3) and presence of natural antioxidants. It is now widely accepted that what the human body needs is a ratio varying from 5:1 to 10: 1 of omega 6 to omega 3. The ratio of polyunsaturated fatty acid (PUFA) and saturated fatty acids (SFA) should be between 0.8 to 1.0.

6. Edible Oil Industry

India is the largest importer of edible oil and the third largest consumer (after China and EU) (www.robobank.com). Of the total 5.0-5.5 million tons of vegetable oils imported by India annually, 1.3-1.5 million tons is soybean oil, imported mostly from Argentina, Brazil and US, nearly 3 million tons of palm oil is purchased from Malaysia and Indonesia. Palm oil imported into India is used in various forms - consumed directly as 'palm oil' after refining, used in the manufacture of *vanaspati*, for blending with other vegetable oil, raw oil and kernel oil for industrial purposes, etc. (Trading in agriculture products, http://www.peclimited.com/agricultural_edibleoil.htm). Per capita consumption is around 10 kg/year. Palm and soybean oil account for almost half of total edible oil consumption in India followed by mustard and groundnut oil (*Recent trend in Indian edible oil industry*, www.robobank.com).

The share of raw oil, refined oil and *vanaspati* in the total edible oil market is estimated at 42%, 48% and 10% respectively (Department of Food and Public Distribution, GOI). India has close to 9% of global oilseeds production, making it among the biggest producers of oilseeds in the world. Over 40% of the total volume of the edible oil sold in India being imported in 2005 (Huge importer of edible oil, www.euromonitor.com).

6.1. Types of Oils commonly in use in India

India is fortunate in having a wide range of oilseeds crops grown in its different agro climatic zones. Groundnut, mustard/rapeseed, sesame, safflower, linseed, nigerseed/castor are the major traditionally cultivated oilseeds (See Table 4). Soybean and sunflower have also assumed importance in recent years. Coconut is most important amongst the plantation crops. Among the non-conventional oils, rice bran oil and cottonseed oil are the most important.

India is a vast country and inhabitants of several of its regions have developed specific preference for certain oils depending upon the oils available in the region. People in the South and West prefer coconut and groundnut oil while those in the East and North use mustard/rapeseed oil. Inhabitants of northern plain are basically hard fat consumers and therefore, prefer *vanaspati*, a term used to denote a partially hydrogenated edible oil mixture.

Table 4 Market share of major edible oils in India

Oil	Percentage
Palm Oil	38
Peanut	14
Sunflower	8
Soybean Oil	21
Rapeseed	13
Cotton	6

Source: Production, Supply and Distribution database, USDA

6.2. Major players of edible oil

Edible Oil Industry is a highly fragmented industry with over 600 oil extraction units and 166 *vanaspati* manufacturing units.

In *Vanaspati* Dalda (earlier manufactured by Hindustan Lever and now by Bunge Limited) is the oldest and largest brand. Other major brands are Rath, Gemini, Raag, Jindal, Gagan, Panghat. The details of these brands are as follows:

- **Rath:** Secunderabad based Agro Tech Foods Ltd is affiliated to ConAgra Foods Inc of USA, one the world's largest food companies. It acquired Rath vanaspati brand from SIEL Ltd.
- **Dalda:** Now made by Bunge Limited, a mega-food giant US multinational. World leader in agribusiness, fertilizer and food products, bought the brand from Hindustan Lever.
- **Gemini:** By Cargill Inc, another US based company, into everything from seeds to genetic food. Largest selling edible oil brand in Maharashtra says Nielsen Company Retail audit data 2007.
- **Raag:** Is a joint venture between the Gujarat based Adani group and the Wilmar International Limited of Singapore.
- **Jindal:** Made by Indian Jindal group, into everything from steel to electric goods.
- **Gagan:** Amrit Banaspati Company is based in Chandigarh, and says it is India's largest selling vanaspati.
- **Panghat:** Part of the Siel group, which has recently changed its name to Mawana Sugars Limited.

In Edible oil The Adani-Wilmer owned Fortune brand was India's number one edible oil brand in 2006 according to A C Nielsen Retail Audit November 2006. Fortune, which is present as a refined soyabean oil, sunflower oil, groundnut oil and mustard oil, is said to have a market share of 19% in the entire edible oil market.

Marico Industries with a market share of 13-15% with brands like Saffola and Sweekar together. (<http://www.business-standard.com/india/news/marico-to-focussaffola-go-slowsweekar/03/56/150972/>).

Other important brands are:

- **Nature Fresh Actilite and Gemini** of Cargill Inc. Gemini was earlier made by Parakh Foods, largest selling brand in western India taken over by Cargill Inc.
- **Gold Winner:** brand of Kaleesuvari Refineries Pvt Ltd. established in 1995 , leading regional brand in South India.
- **Dhara** of Dhara Vegetable Oil & Foods Company Limited (DOFCO), Anand set up in 2000 as a wholly owned subsidiary of National dairy Development board (NDDB)
- **Cooklite** of Godrej Foods and **Postman** of Ahmed Nagar Mills are other well known brands.

7. Processing

Food fats and oils are derived from oilseed and animal sources. Animal fats are generally heat rendered from animal tissues to separate them from protein and other naturally occurring materials. Rendering may be either with dry heat or with steam. Vegetable fats are obtained by the extraction or the expression of the oil from the oilseed source. Historically, cold or hot expression methods were used. These methods have been replaced with solvent extraction or pre-press/solvent extraction methods which give higher oil yield. In this process the oil is extracted from the oilseed by hexane (a light petroleum fraction) and the hexane is then separated from the oil, recovered, and reused.

7.1. Hydrogenation

Hydrogenation is the process by which hydrogen is added directly to points of unsaturation in the fatty acids. Hydrogenation of fats has developed as a result of the need to (1) convert liquid oils to the semi-solid form for greater utility in certain food uses and (2) increase the oxidative and thermal stability of the fat or oil.

In the process of hydrogenation, hydrogen gas is reacted with oil at elevated temperature and pressure in the presence of a catalyst. The catalyst most widely used is nickel supported on an inert carrier which is removed from the fat after the hydrogenation processing is completed. Under these conditions, the gaseous hydrogen reacts with the double bonds of the unsaturated fatty acids as shown below:



The hydrogenation process is easily controlled and can be stopped at any desired point. As hydrogenation progresses, there is generally a gradual increase in the melting point of the fat or oil. If the hydrogenation of oil is stopped after a small amount of hydrogenation has taken place, the oils remain liquid resulting in partially hydrogenated oils used to produce institutional cooking oils, liquid shortenings and liquid margarines. Further hydrogenation can produce soft but solid appearing fats, which still contain appreciable amounts of unsaturated fatty acids and are used in solid shortenings and margarines. When oils are fully hydrogenated, many of the carbon to carbon double bonds are converted to single bonds increasing the level of saturation. This conversion also affects *trans* fatty acids eliminating them from fully hydrogenated fats and the resulting product is a hard brittle solid at room temperature.

8. *Trans* Fats

Fatty acids found in foods can be saturated (i.e. no double bonds), monounsaturated (1 double bond) or polyunsaturated (2 or more double bonds). The double bonds provide rigidity to the molecule and result in specific molecular configurations. Naturally occurring fatty acids in foods usually have the *cis* configuration, i.e. the hydrogen atoms with respect to the double bond are on the same side of the molecule resulting in the molecule having a "V" shape. In *trans* fatty acids (TFAs), the hydrogen atoms are on the opposite sides of the molecule, and the molecule assumes a nearly linear configuration similar to that for saturated fatty acids. TFAs behave more like

saturated fatty acids than unsaturated fatty acids, and this has consequences when they are incorporated into membranes and other cellular structures. Major TFAs include isomers of oleic acid (9 *cis* C18:1), including elaidic acid (9 *trans* C18:1), primarily found in partially hydrogenated vegetable oils, and vaccenic acid (11 *trans* C18:1), found in meat/ dairy products. TFAs are also present as conjugated linoleic acid (CLA, C18:2) that contains both *cis* and *trans* double bonds.

8.1 Where are *trans* fats found?

The majority of *trans* fat can be found in partially hydrogenated vegetable oils (*vanaspati*), shortenings, stick (or hard) margarine, cookies, crackers, snack foods, fried foods (including fried fast food), doughnuts, pastries, baked goods, and other processed foods made with or fried in partially hydrogenated oils. Some *trans* fat is found naturally in small amounts in various meat and dairy products.

9. Health risks associated with *trans* fats

9.1 Heart Diseases

Metabolic studies have shown that *trans* fats have adverse effects on blood lipid levels--increasing LDL ("bad") cholesterol while decreasing HDL ("good") cholesterol.

In a study conducted by Mensink *et al* 1990, 34 women (mean age, 26 years) and 25 men (mean age, 25 years) were placed on three mixed natural diets of identical nutrient composition, except that 10 percent of the daily energy intake was provided as oleic acid (which contains one double bond), *trans* isomers of oleic acid, or saturated fatty acids. The three diets were consumed for three weeks each, in random order and revealed diets high in *trans* fatty acids (11.0% energy) raised total and LDL cholesterol and lowered HDL cholesterol compared to a high oleic acid diet. The effects on lipoprotein levels did not differ between women and men. The effect of *trans* fatty acids on the serum lipoprotein profile is at least as unfavorable as that of the cholesterol-raising saturated fatty acids, because they not only raise LDL cholesterol levels but also lower HDL cholesterol levels. This combined effect on the ratio of LDL to HDL cholesterol is double that of saturated fatty acids.

The relation between *trans* fatty acids intake and risk of coronary heart disease (CHD) reported from three large cohort studies, the Health Professionals Follow-up Study (HPFS), the Alpha-Tocopherol Beta-Carotene study (ATBC) and the Nurses Health Study (NHS) provides the strongest epidemiological evidence relating dietary factors to risk of CHD. In these studies, *trans* fat consumption was assessed using detailed food frequency questionnaires (FFQ) that were validated by comparison with adipose composition or several days of diet records.

The Health Professionals Follow-up Study (HPFS) examined the association between fat intake and the incidence of coronary heart disease in men of middle age and older. 734 coronary events were documented, including 505 non-fatal myocardial infarctions and 229 deaths. After age and several coronary risk factors were controlled for, significant positive associations were observed between intake of saturated fat and risk of coronary disease. Positive associations between intake of cholesterol and risk of coronary heart disease were similarly attenuated after adjustment for fibre intake. Intake of linolenic acid was inversely associated with risk of myocardial infarction; this association became significant only after adjustment for non-dietary risk factors and was strengthened after adjustment for total fat intake (Ascherio *et al*, 1996).

In the Finnish Alpha-Tocopherol Beta-Carotene Cancer Prevention (ATBC) Study the relation of intakes of specific fatty acids and the risk of coronary heart disease was examined in a cohort of 21,930 smoking men aged 50-69 years who were initially free of diagnosed cardiovascular disease. After controlling for age, supplement group, several coronary risk factors, total energy, and fiber intake, a significant positive association between the intake of *trans* fatty acids and the risk of coronary death was observed. The intake of omega-3 fatty acids from fish was also directly related to the risk of coronary death in the multivariate model adjusting also for *trans*-saturated and *cis*-monounsaturated fatty acids. There was no association between intakes of saturated or *cis*-monounsaturated fatty acids, linoleic or linolenic acid, or dietary cholesterol and the risk of coronary deaths (Pietinen *et al* 1997).

The relation between dietary intake of specific types of fat, particularly *trans* unsaturated fat and the risk of coronary disease was studied in women enrolled in the Nurses' Health Study (NHS). 80,082 women who were 34 to 59 years of age and had no known coronary disease, stroke, cancer, hypercholesterolemia, or diabetes in 1980 were studied. The study suggested that replacing saturated and *trans* unsaturated fats with unhydrogenated monounsaturated and polyunsaturated fats is more effective in preventing coronary heart disease in women than reducing overall fat intake (Hu *et al* , 1997).

In the Zutphen Elderly Study the relation between *trans* fatty acid intake and coronary heart disease was investigated in a Dutch population with a fairly high *trans* fatty acid intake, including *trans* fatty acids from partly hydrogenated fish oils. 667 men of the Zutphen Elderly Study aged 64-84 years and free of coronary heart disease at baseline were studied. Dietary surveys were used to establish the participants' food consumption patterns. It was found that high intake of *trans* fatty acids (all types of isomers) contributes to the risk of coronary heart disease. The substantial decrease in *trans* fatty acid intake, mainly due to industrial lowering of *trans* contents in Dutch edible fats, could therefore have had a large public-health impact (Oomen *et al* , 2001).

Trans isomers of fatty acids, formed by the partial hydrogenation of vegetable oils to produce margarine and vegetable shortening, increase the ratio of plasma low-density-lipoprotein to high-density-lipoprotein cholesterol, and adversely influence risk of coronary heart disease. The largest epidemiological study, the Nurses Health study, showed a significant, positive association with the intake of industrially produced *trans* fatty acids and a non significant, inverse association between the intake of *trans* fatty acids from ruminants and the risk of heart disease. Intakes of foods that are major sources of *trans* isomers (margarine, cookies, cake, and white bread) were each significantly associated with higher risks of CHD (Willett *et al*, 1993).

9.2 Cancer

In the EURAMIC study dating from 1997, the association between *trans* fatty acid levels in adipose tissue and the incidence of cancer of the breast, prostate and large intestine was investigated in European populations with wide differences in dietary levels of *trans* fatty acids. Cancers of the breast and colon were associated negatively with *cis*-monounsaturated fatty acids however a positive association was found between *trans* fatty acid intake and the incidence of cancer of the breast and large intestine (Bakker *et al* 1997).

In another case control study investigating the association between *trans* fatty acids and cancer of the large intestine in 2000 patients and 2000 controls, signs of an increased risk of cancer related to the intake of *trans* fatty acids in subgroups of these patients were found. The association

between trans-fatty acids and colon cancer using data from a case (n = 1,993) and control (n = 2,410) study conducted in Utah, Northern California, and Minnesota. Dietary data were collected using a detailed diet history questionnaire, and nutrient values were generated from the Nutrition Coordinating Center nutrient database. After adjustment for other variables, including age at diagnosis, body size, physical activity, aspirin and/or nonsteroidal anti-inflammatory drug (referred to collectively as NSAIDs) use, energy intake, and dietary fiber and calcium, a weak association was found in women; no increased risk was observed for the *cis* form of the fatty acids. For men and women, slightly stronger associations were observed in those with 67 or more years of age. Those who did not use NSAIDs were at a 50% greater risk of developing colon cancer when they consumed high levels of *trans*-fatty acids. Women who were estrogen negative, i.e., postmenopausal not taking hormone replace therapy, had a two-fold increase in risk from high levels of *trans*-fatty acids in the diet, while women who were estrogen positive did not experience an increased risk of colon cancer, regardless of level of *trans*-fatty acids consumed. Consumption of partially hydrogenated fats should be avoided, since no increased risk was observed for the *cis* form of fatty acids, while suggestions of increased risk from *trans*-fatty acids exist for subsets of the population (Slattery, 2001).

In the Netherlands Cohort Study on Diet and Cancer, which comprised 941 cases of breast cancer, intake data derived from a validated 150-item food-frequency questionnaire were linked to an existing database with analytic data on specific fatty acids in European foods (the TRANSFAIR study). Voorrips *et al* 2002 evaluated the relation between intakes of CLA and other fatty acids and breast cancer incidence and a weak, positive relationship between CLA intake and incidence of breast cancer was found from the use of data from the TRANSFAIR study. Statistically significant positive associations were found with total *trans* fatty acids and (borderline) with saturated fatty acids. Significant inverse associations were found with monounsaturated and *cis* unsaturated fatty acids, whereas total fat and energy intake of CLA-containing food groups were not related to breast cancer incidence (Voorrips *et al* 2002).

9.3 Diabetes

Clinical studies show that *trans* fatty acids can increase insulin resistance and that exercise can enhance the rate of adaptation to a high fat diet by increasing the rate of fat oxidation. The differences in response of inflammatory signals and of insulin resistance to different fatty acids indicate that not all fatty acids are the same. There are also experimental data showing that most, but not all, animals consuming a high fat diet will become obese. A number of mechanisms have been postulated for this difference, including differential sensitivities to neurotransmitters, to the intestinal peptide, enterostatin, and to individual fatty acids. The administration of a high-fat meal of variable fatty acid composition, elaidic acid (9 *trans*-18:1) compared with oleic acid (9 *cis*-18:1) gave rise to higher insulin levels in the blood at the same blood sugar level, which indicates that elaidic acid produces increased insulin resistance (Bray *et al*, 2002)

Analysis of the Nurses' Health Study after 14 years' observation showed relations between dietary fat intakes and the risk of type 2 diabetes. The study suggested that total fat and saturated and monounsaturated fatty acid intakes are not associated with risk of type 2 diabetes in women, but that *trans* fatty acids increase and polyunsaturated fatty acids reduce risk. Substituting non-hydrogenated polyunsaturated fatty acids for *trans* fatty acids would likely reduce the risk of type 2 diabetes substantially (Salmeron *et al*, 2001)

9.4 Effect on Fetus

Recent studies on humans have shown that *trans* fatty acids are transferred to the fetus, as they were found in the same levels in the blood of new born infants as in that of mothers. Both the fetus and the breast-fed baby are consequently exposed to *trans* fatty acids corresponding to the mother's intake. Dietary *trans* fatty acids can in part compete with essential polyunsaturated fatty acids in the body. In animal experiments, a high intake of industrially produced *trans* fatty acids inhibits the formation of long-chain polyunsaturated fatty acids (LCPUFAs) from their precursors (Pax *et al.*, 1992). LCPUFAs are important for both growth and the development of vision and the central nervous system early in life. The amount of *trans* fatty acids that must be supplied before the synthesis of LCPUFAs is affected is, however, unknown. In 1992, a study of premature babies was published in which a negative correlation was found between birth weight and *trans* fatty acid level's in plasma 4 days after birth (Koletzko 1992).

In a study published in 2001, *trans* fatty acid levels including CLA in the umbilical blood of 84 neonates reflected the mother's levels of *trans* fatty acids in the blood and thus the mother's *trans* fatty acid intake. There was an inverse relationship in the infants' blood between *trans* fatty acids and polyunsaturated fatty acids. The pregnancy period was found to be shorter in mothers with the higher *trans* fatty acid level in the infant's blood (Elias and Innis 2001).

9.5 Allergy

In an international study of asthma and allergies in childhood from 1998, the incidence of asthma, allergic cold and asthmatic eczema in children aged 13-14 years was investigated in 155 centres around the world. A positive association was found between the intake of *trans* fatty acids and these diseases. Such an association was not observed for the intake of monounsaturated and polyunsaturated fatty acids (Weiland *et al.*, 1999).

9.6 Obesity

Dietary fat and its relation to obesity has been a controversial issue for several years. Research with monkeys indicates that *trans* fat may increase weight gain and abdominal fat, despite a similar caloric intake (Goslina, Anna, 2006).

A 6-year experiment revealed that monkeys fed a *trans*-fat diet gained 7.2% of their body weight, as compared to 1.8% for monkeys on the mono-unsaturated fat diet (<http://www.newscientist.com/channel/health/mg19025565.000-six-years-of-fastfood-fats-supersizes-monkeys.html>).

10. Regulations for *trans* fats

10.1 International

The World Health Organisation (WHO) has recommended that governments around the world phase out partially hydrogenated oils if *trans*-fat labelling alone doesn't spur significant reductions. WHO also recommends that the *trans* fatty acids consumption should be less than 1% of the total daily energy intake (WHO, 2003).

The Codex Alimentarius Commission in its response to the WHO's Action plan for implementation of the global strategy on diet, physical activity and health stated that "if the provisions for labelling

of, and claims for, *trans*-fatty acids do not affect a marked reduction in the global availability of foods containing *trans*-fatty acids produced by processing of oils and by partial hydrogenation, consideration should be given to the setting of limits on the content of industrially produced *trans*-fatty acids in foods”.

10.1.1 Denmark

Denmark became the first country to introduce laws strictly regulating the sale of many foods containing *trans* fats - a move which effectively bans partially hydrogenated oils. In March 2003, following notification in 2002, the Danish food authorities, on the ground that the measure was justified on public health grounds and was aiming at minimizing the risk of cardiovascular diseases adopted legislation which introduced with effect from 1 June 2003, a limit on the level of *trans* fatty acids.

The following provisions are laid down pursuant to section 13, section 55(2) and section 78(3) of Act No. 471 of 1 July 1998 on Foods etc. (the Danish Food Act):

1. From 1 June 2003, the content of *trans* fatty acids in the oils and fats covered by this Executive Order shall not exceed 2 grams per 100 grams of oil or fat, but see subarticle (2).
2. From 1 June 2003 to 31 December 2003, the content of *trans* fatty acids in the oils and fats covered by this Executive Order which are part of processed foods in which food ingredients other than oils and fats are also contained, and which are manufactured in the food industry, the retail trade, catering businesses, restaurants, institutions, bakeries etc. may, however, be up to 5 g per 100 of oil or fat. From 1st January 2004, <2% TFAs are permitted in oils and fats used in both local and imported processed foods.
3. In products claimed to be “free of *trans* fatty acids”, the content of *trans* fatty acids shall be less than 1 gram per 100 grams of the individual oil or the individual fat in the finished product.

Trans fatty acids shall be defined as the sum of all isomeric fatty acids with 14, 16, 18, 20 and 22 carbon atoms and one or more *trans* double bonds, i.e. C14:1, C16:1, C18:1, C18:2, C18:3, C20:1, C20:2, C22:1, C22:2 *trans* isomeric fatty acids, but only polyunsaturated fatty acids with methylene-interrupted double bonds.(Source: Executive Denmark’s *trans* fat law Order No. 160 of 11 March 2003 on the Content of *Trans* Fatty Acids in Oils and Fats etc, English Translation).

10.1.2 Canada

Canada Nutrition information changes to Food and Drug Regulations were made in January 2003 requiring compliance by 12 December 2005 for large manufacturers, and by 12 December 2007 for small manufacturers. If claims are made (e.g reduced TFA levels) immediate compliance is required.

All vegetable oils and spreadable margarines have *trans* fat content limited to 2% of total fat content. A limit of 5% *trans* fat of total fat content in all products sold to Consumers.

(Canada Agricultural Situation Nutrition labeling Regulations 2005 available at <http://www.fas.usda.gov/gainfiles/200502/146118917.doc>)

10.1.3 USA

On 11th July 2003, US Food and Drug Administration(FDA) published a final rule in the Federal Register that amended its regulations on food labeling to require *trans* fatty acids be declared in the nutrition label of conventional foods and dietary supplements (68FR 41434) (effective from January

1 2006). In August 2003 FDA issued a detailed guidance document on interpretation of the regulations "Guidance for Industry: Food labeling: Trans Fatty acids in Nutrition labeling, Nutrient content Claims, and Health Claims." In April 2004 the FDA Advisory Committee recommended that TFA intake be reduced to "...<1% of energy intake..." Subsequently (January 2006), the FDA required TFA levels also to be included on food labels, with % saturated fat equal to the sum of saturated and *trans* fatty acids.

The FDA decided not to distinguish between industrially produced TFAs and those derived from rumen hydrogenation, thus dairy products must be labeled with TFA levels. This rule took effect on 1 January 2006.

Trans fatty acids should be listed as "trans fat" or "trans" on a separate line under the listing of saturated fat in the nutrition label. Trans fat content must be expressed as grams per serving to the nearest 0.5 g increment below 5 grams and to the nearest gram above 5 grams.

If a serving contains less than 0.5 g per serving to be labeled as 0 gram per serving, or trans fat free and foods containing greater than 4 g saturated+ trans fatty acids cannot carry a health claim (<http://vm.cfsan.fda.gov/~dms/transqui.html>).

10.1.4 European Union

The European Food Safety Authority was asked to produce a scientific opinion on *trans* fats (*European Food Safety Authority (2004)*). Declarations of the amount of TFA in a food are subject to the rules on nutrition labeling harmonized at EU level. Nutrition labeling is voluntary unless a nutrition claim is made. Separate identification of the amount of TFA, as a component of the total fat content of the food is only required if a TFA nutrition claim is made. The commission has announced that the Directive on nutrition labeling will soon be amended. In the UK and many other European countries the situation is complicated. Although there is no specific requirement for the labeling of trans-fats on food labels, some manufacturers have started to do so voluntarily.

10.1.5 Australia/New Zealand

Mandatory TFA labeling was considered during a comprehensive review of the Food Standards Code in 1999-2002, however Food Standards Australia New Zealand (FSANZ) and its precursor decided not to mandate labeling of TFAs as it was believed that TFA consumption was relatively low; and that similar reductions in saturated fat intake would be more likely to have a greater impact. TFA contents are required on labels only if a nutrition claim is made with respect to cholesterol, saturated or unsaturated fatty acids or TFAs. Voluntary labeling is permitted and many vegetable oil spread manufacturers include TFA levels on labels. The Australian Heart Foundation recommends that saturated and TFAs be 8% of the total energy intake.

10.2 Regulations for Edible oil in India

Four major types of regulatory policy instruments are used in India for enhancing food safety: mandatory product standards; mandatory process standards; licensing and prohibitions and voluntary product and process standards. Edible Oil Industry is regulated under different standards.

10.2.1 Prevention of Food Adulteration Act, 1954

Includes specific standard on edible oils (Appendix B) giving broad specification for different oils – cottonseed, coconut, groundnut, linseed, mahua, rapeseed, olive (revised), sunflower etc. Includes standard for blended vegetable oil, which allows different oils to be blended and sold. The

specifications do not lay down guidelines on fatty acid composition of different oils. In addition, there are specifications for *vanaspati* (hydrogenated vegetable oil). Under this standard, companies can mix any quantity of any 'harmless' vegetable oil in their brand and can vary it as well.

In September 2008, the ministry issued notification for labelling of food, under the PFA. This notification includes for the first time labelling for nutrition and health claims. For edible oil, if the company makes nutrition or health claims, then it is required to: provide information on its package about the amount or type of fatty acids, including cholesterol, SFA, MUFA, PUFA and *trans* fats.

10.2.2 Bureau of Indian Standards

The Bureau of Indian Standards (BIS) lays down different specifications for Edible Oils and *vanaspati*. Giving requirements for physical and chemical tests for moisture and insoluble impurities (percent by mass), Colour, Refractive index at 40°C, Iodine value, unsaponifiable matter (percent by mass), Flash Point (°C), heavy metals, aflatoxins and pesticides, free fatty acid value expressed as oleic acid (maximum 5.0 and 0.25 percent by mass for raw and refined grades of materials), but no standard for fatty acid composition or *trans* fats.

10.2.3 AGMARK

The Agmark is a voluntary standard for Vegetable Oils and *vanaspati* governed by the Directorate of Marketing and inspection of the Ministry of Agriculture (Government of India) as per the Agricultural produce Grading and Marking Act (1937). Blended Edible Vegetable oil and fat spread are compulsorily required to be certified under AGMARK. Does not have any standards for fatty acids or *trans* fat.

11. Review Of Literature

Fatty acid patterns were determined in 83 brands of margarine, 9 brands of low-calorie margarine and 18 brands of shortening, frying and cooking fat purchased at random from the retail market in the Federal Republic of Germany in 1973/1974, and a second time in 1976. As a result of gas-liquid chromatographic analyses on a Silar 10 C coated packed column, complemented in some cases by the values recorded on a highly selective SP 2340 capillary column, *trans*-octadecenoic acids ranging from 53.2 to 0.1% were measured. None of the products examined was completely free of *trans*-fatty acids. High values of *trans*- octadecenoate were always accompanied by positional isomers of *cis*- octadecenoate, by 9 *trans*, 12 *trans*-octadecadienoate and by 9*cis*,12 *trans*-octadecadienoate and 9 *trans*,12*cis*-octadecadienoate. Furthermore, two mixed geometric isomers derived from linolenic acid (probably 9*cis*, 12*cis*, 15 *trans*-octadecatrienoate and 9 *trans*,12*cis*,5*cis*-octadecatrienoate) could be identified, provided that the individual brand contained sufficient linolenic acid. Following partial hydrogenation, *trans* hexadecenoate, 0.1 to 0.2%, was detected in some of the edible fats (Heckers and Melcher1978).

A study examined the concentration (mg/g) of *trans* polyunsaturated fatty acid (TPFA) in five soybean oil brands by gas-liquid chromatography. Tricosanoic acid methyl ester was used as the internal standard. All samples analyzed presented *trans* 18:2 fatty and *trans* 18:3 acids in detectable amounts. The concentration of TPFA ranged from 5.8 to 30.2 mg/g, with a mean concentration value of 18.4 mg/g. *trans* 18:3 fatty acids had the highest TPFA group concentrations, which ranged from 3.9 to 16.3 mg/g. The main isomer of this group presented the

9c, 12c, 15t configuration. For *trans* 18:2 fatty acids, concentrations ranged from 1.9 to 14.0 mg/g with a mean value of 8.1 mg/g. Alpha-linolenic acid (all *cis*) concentrations ranged from 30.7 to 60.6 mg/g and their degree of isomerization ranged from 6.0 to 31.5, indicating that the deodorization process varies from one producer to another. From per capita consumption of soybean oil brands in Brazil and their TPHA concentrations, it is possible to conclude that their contribution to the average TPHA intake per person in Brazil is 0.4 g/d (Martin et al, 2006).

Estimated dietary intake of TFA for the Australian population ranged between 1.2 and 1.6g/day and for New Zealanders ranged between 1.6 and 2.0 g/day at the mean level of intake. Major contributors to the intake of Australia were dairy products (26-44%), pastry and pastry based mixed foods (8-17%), fats and oils (8-18%) meat and poultry (9-15%), cereal and cereal products (10-13%) and cereal based products (10-13%) and cereal based mixed foods (6-12%) depending on the population group assessed. Major contributors to the intake of TFA for New Zealand were fats and oils (30-44%), dairy products (19-21%) cereal and cereal based products (9-10%) pastry and pastry based mixed foods (8-11%), meat and poultry (8-10%).

(Review Report on Trans fatty acids in the New Zealand and Australian Food Supply available at http://www.foodstandards.gov.au/srcfiles/Transfat%20report_CLEARED.pdf)

A maximum of 100 foods per country were sampled and centrally analyzed. Each country calculated the intake of individual *trans* and other fatty acids, clusters of fatty acids and total fat in adults and/or the total population using the best available national food consumption data set. A wide variation was observed in the intake of total fat and (clusters) of fatty acids in absolute amounts. The variation in proportion of energy derived from total fat and from clusters of fatty acids was less. Only in Finland, Italy, Norway and Portugal total fat did provide on average less than 35% of energy intake. Saturated fatty acids (SFA) provided on average between 10% and 19% of total energy intake, with the lowest contribution in most Mediterranean countries. TFA intake ranged from 0.5% (Greece, Italy) to 2.1% (Iceland) of energy intake among men and from 0.8% (Greece) to 1.9% among women (Iceland) (1.2-6.7 g/d and 1.7-4.1 g/d, respectively). The TFA intake was lowest in Mediterranean countries (0.5-0.8 en%) but was also below 1% of energy in Finland and Germany. Moderate intakes were seen in Belgium, The Netherlands, Norway and UK and highest intake in Iceland. *trans* isomers of C18: 1 were the most TFA in the diet. Monounsaturated fatty acids contributed 9-12% of mean daily energy intake (except for Greece, nearly 18%) and polyunsaturated fatty acids 37%. The current intake of TFA in most Western European countries does not appear to be a reason for major concern. In several countries a considerable proportion of energy was derived from SFA. It would therefore be prudent to reduce intake of all cholesterol-raising fatty acids, TFA included (Hulshof *et al* 1999).

Trans fatty acids (TFA) comprise a variety of positional isomers, mainly with 18 carbon atoms and one double bond (C18:1). They are found in foods of ruminant animal origin and in partially hydrogenated vegetable oils. The isomeric composition of TFA in animal and vegetable foods differs, but no definite differences have been documented between the metabolic and health effects of the different isomers. In the Nordic countries the intake of TFA has declined during the past 10-15 years, mainly through reduced use of partially hydrogenated vegetable oils. TFA are mainly found in foods that contain far higher amounts of saturated fatty acids (SFA). The proportion of SFA

plus TFA should be kept to one-third of total dietary fatty acids. The problem of excessive consumption of these unfavorable fatty acids should be managed with food-based dietary guidelines in agreement with the Nordic Nutrition Recommendations (Aro *et al*, 2006).

Detailed fatty acid analysis of over 200 foods was undertaken for the purpose of determining the variability in fatty acid content among foods within a product category and the significance of this variability to the estimation of *trans* fatty acids intakes from analysis of dietary intake data. The results show that reflecting differences in the fats and oils in the manufacturing or preparation processes. Level of *trans* fatty acids in 16 breads (whole wheat and white) ranged from 1% to over 30% of total fatty acids. In hard margarine it was 31.1 to 44.6 % of total fatty acids. In soft margarines 1.1 to 44.4 % of total fatty acids (Innis *et al*, 1999).

A study was conducted to examine the trends in dietary intake of trans fatty acids from 1980-82 and 1995-1997 using data collected as part of the Minnesota Heart Survey. Downward trend in dietary intake of trans-fatty acids were found between 1980-82 and 1995-1997. for men mean intake of total trans fatty acids declined from 83 g per day in 1980-82 to 6.2 g/day in 1995-97 (Harnack *et al* 2003).

The fatty acid composition of chips, cakes and ice creams was determined with particular attention to their *trans* fatty acid content. The *trans* C18:1 content was determined by a combined capillary gas-liquid chromatography (GLC) and silver thin-layer chromatography (Ag-TLC). Six of ten types of chips examined contained more than 10% *trans* C18:1 (in the range of 10.3 to 17.3% of the total fatty acids), and the other four had below 0.5%. In the lipids of cakes *trans* C18:1 isomers occurred at 1.49 to 41.44% and only four types of cakes contained less than 5% of *trans* C18:1. The *cis-trans* and *trans-cis* C18:2 isomers were present among the fatty acids of the majority of chips and cakes investigated. Six types of chips contained *trans-trans* C18:2 in the 1.2–1.6% range. *trans* fatty acids were absent in the lipids of 6 types of ice cream, but two types contained 11.3 and 19.4% *trans* C18:1 (Izegarska *et al* 2001).

Edible oils sold in the capital cities of eight provinces were purchased. One hundred twenty-six samples, representing 14 different oils according to their labels, were assayed for their fatty acid content in 2001. Fatty acids were analyzed by standard gas chromatographic methods. More than 76% of households in China consume edible oil, providing an average of 29.6 g of edible oil per day to persons aged two years or older. Rapeseed was consumed by one-quarter of individuals. Rapeseed is rich in C22:1n9 *cis* (erucic acid). About 33% of edible oils differed from their labeled identification. Rapeseed oil, identified by the presence of C22:1n9 (erucic acid), was most frequently not labeled as such. In another 28% of the samples, trans isomers of linolenic acid were detected. Deviations from the label identification were more common in southern than in northern provinces. Regulations requiring complete labeling of mixed edible oils in China might help prevent unintentional consumption of fatty acids associated with adverse health outcomes. In particular, consumption of erucic acid and trans fatty acids might be reduced. The results suggest the need for closer control of food oil labeling in China, especially in the South (Wallingford *et al*, 2004).

The contents of *trans* fatty acids (TFA) in 139 German foods were analyzed by capillary gas chromatography. The TFA analysis included myristelaidic acid (C14:1 t9), palmitelaidic acid (C16:1

t9), petroselaidic acid (C18:1 t6), elaidic acid (C18:1 t9), *trans* vaccenic acid (C18:1 t11), octadecadienoic acid isomers (C18:2 c9t12, C18:2 t9t12, and C18:2 9tc12), and 9 geometrical octadecatrienoic acid isomers (C18:3 n-3). Maximum TFA contents occurred in French fries (up to 34%), crisps (up to 22%) and in instant sauces (up to 22%). The TFA content in dairy products ranged from 2.0% (blue cheese) to 6.1% (Jurassic cheese), in meat products from 0.2% (cooked ham) to 8.6% (lamb), in fish from 0.4% (pollock) to 1.0% (carp), and in vegetable fats and oils and margarines from <0.01% (edible oils) to 4.1% (sunflower margarines). The average estimated TFA intake was calculated to 1.9g TFA/d for women and 2.3g TFA/d for men (Fritsche and Steinhar, 1997).

The fatty acid composition of 11 brands of peanut butter and paste freshly prepared from roasted peanuts was analyzed with emphasis on isomeric *trans*-fatty acids. No *trans*-fatty acids were detected in any of the samples in an analytical system with a detection threshold of 0.01% of the sample weight. Hydrogenated vegetable oils are added to peanut butters at levels of 1-2% to prevent oil separation. Some hydrogenated vegetable oils are known to be sources of *trans*-fatty acids in the human diet. The addition of these products was not found to result in measurable amounts of *trans*-fatty acids in the peanut butters analyzed. (Sanders, 2001)

In 1992 margarines were examined, and in 1995 brands covering the entire Danish market were examined. Significant amounts of *trans*-18:1 were found only in hard margarines (mean: $4.2 \pm 2.8\%$) and shortenings (mean: $6.8 \pm 3.1\%$), whereas the semi-soft and soft margarines contained substantially less *trans*-18:1 in 1995 than in 1992. Where marine oils had been used to a larger degree the mean *trans*-monoenoic content was about 15%, of which close to 50% was made up of long-chain (C₂₀ and C₂₂) *trans* fatty acids. A note-worthy decrease in the content of *trans*-18:1 had occurred for the semi-soft margarines, from $9.8 \pm 6.1\%$ in 1992 to $1.2 \pm 2.2\%$ in 1995. Calculated from sales figures, the supply of *trans*-18:1 plus saturated fatty acids from margarines had decreased over this three-year period by 1.4 g/day, which has been replaced by *cis* monounsaturated and polyunsaturated fatty acids (Ngeh-Ngwainbi *et al*, 1997).

Fifteen laboratories participated in a collaborative study to determine total, saturated, unsaturated, and monounsaturated fats in cereal products by gas chromatographic (GC) analysis of fatty acid methyl esters (FAMES). Cereal products, representing a wide range of cereal grains and processes, were hydrolyzed in 8N HCl and extracted with ethyl and petroleum ethers. FAMES were produced by the reaction of the mixed ether extracts with sodium hydroxide in methanol (NaOH/MeOH) and then with boron trifluoride reagent (14% BF₃ in MeOH). Capillary GC quantitatively determined them. Total fat was calculated as the sum of individual fatty acids expressed as tri-acyl glycerol equivalents in accordance with nutrition labeling guidelines. Saturated, unsaturated, and monounsaturated fats were calculated as sums of individual fatty acids. The total fat contents of samples ranged from 0.56 to 12.64%. A split design was used to determine performance parameters of results obtained by 15 laboratories on 24 samples. Of the 24 samples, 7 were blind duplicates and 5 were independent materials. Statistical analysis for total fat yielded a relative standard deviation for repeatability (RSD_r) range of 1.32 to 13.30% and a relative standard deviation for reproducibility (RSD_R) range of 4.42 to 22.82%. The goal of this study was to determine total fat, saturated fat, unsaturated, and monounsaturated fat in cereal-based products

by complete extraction, methylation, and quantitation of total fatty acids. The acid hydrolysis-capillary GC method for determining total, saturated, unsaturated, and monounsaturated fats in cereal products has been adopted by AOAC INTERNATIONAL (Ovesen *et al*, 1996).

This study presents the FA composition and *trans* FA (TFA) contents of different hydrogenated vegetable oils and blended fats marketed in Pakistan. Thirty-four vanaspati (vegetable ghee), 11 shortenings, and 11 margarines were analyzed. The contents of saturated FA, *cis* monounsaturated FA, and *cis* PUFA were in the following ranges: vanaspati 27.8–49.5, 22.2–27.5, 9.3–13.1%; vegetable shortenings 37.1–55.5, 15.8–36.0, 2.7–7.0%; and margarines 44.2–55.8, 21.7–39.9, 2.9–20.5%, respectively. Results showed significantly higher amounts of TFA in vanaspati samples, from 14.2 to 34.3%. Shortenings contained TFA proportions of 7.3–31.7%. The contents of TFA in hard-type margarines were in the range of 1.6–23.1%, whereas soft margarines contained less than 4.1% TFA (Bhanger and Anwar, 2004).

12. Materials and Methods

12.1. Sampling methodology

Edible Oil samples of different brands were purchased from various markets in Delhi in the month of April 2007. Details of the samples are in Annexure I. 30 samples of different edible oils and fats classified as follows: Edible oils (21 samples) - soybean, sunflower, safflower, ground nut, mustard, coconut, olive, sesame oil, rice bran and palm oil and blended Oil (safflower+rice bran oil) - Vanaspati (7 samples), desi ghee (1 sample) and butter (1 sample) were analyzed for total saturated, total unsaturated and trans fatty acids methyl esters. Each sample was analysed separately and in duplicate.

12.2. Apparatus

Gas Chromatographs-Thermoquest-Trace GC equipped with Split/Splitless Injection system - with Flame Ionization Detector with advanced software (Chromcard-32 bit Ver 1.06 October 98). A fused silica capillary column coated with a highly polar stationary phase, HP-88 (88%-Cyanopropyl)-methylarylpolysiloxane, - 100 m x 0.25 mm i.d x 0.20 μ m with oven temperature programme - Initial temperature 140^oC, hold time of 5 minutes; ramp, 1^oC/min; final temperature 250^oC; hold time 5 minutes. Total run time 120 minutes. Injector port, 225^oC; Detector 260^o C; The gas flow rates used were 0.3ml /min carrier gas (Nitrogen), 15ml/min make up gas (Nitrogen) and 35 and 350 ml/min flame gases (Hydrogen and Air, respectively). A 10- μ l syringe from Hamilton Co. was employed for injection.

Reaction flasks - 125 ml flasks with outer standard tapered joints. Condenser-water cooled, reflux with 20-30 cm jacket and standard taper inner joint.

12.3 Reagents

All the reagents and solvents used were of AR grade.

- Boron trifluoride Reagent (125 g BF₃/L methyl alcohol)
- Methanolic sodium hydroxide solution (0.5 M)
- n-Heptane –AR grade

12.4. Standardsa). The standard fatty acid methyl ester (FAME) mixture of 37 components used for calibration was obtained from Supelco, USA details given below:

S. No	Trivial Name	IUPAC Name	Percent	Carbon Number	Retention time (minutes)
1	Butyric acid Methyl ester	methyl butanoate	4	C4:0	21.89
2	Caproic acid Methyl ester	methyl hexanoate	4	C6:0	22.32
3	Caprylic acid Methyl ester	methyl octanoate	4	C8:0	24.79
4	Capric acid methyl ester	methyl decanoate	4	C10:0	29.13
5	Undecanoic acid methyl ester	methyl undecanoate	2	C11:0	32.23
6	Lauric acid methyl ester	methyl dodecanoate	4	C12:0	36.41
7	Tridecanoic acid methyl ester	methyl tridecanoate	2	C13:0	40.69
8	Myristic Acid methyl ester	methyl tetradecanoate	4	C14:0	45.79
9	Myristoleic Acid methyl ester	methyl tetradecenoate	2	C14:1	49.74
10	Pentadecanoic acid methyl ester	methyl pentadecanoate	2	C15:0	51.06
11	<i>Cis</i> -10-Pentadecenoic acid methyl ester	<i>cis</i> -methyl pentadec-10-enoate	2	C15:1	55.39
12	Palmitic acid Methyl ester	methyl hexadecanoate	6	C16:0	56.98
13	Palmitoleic acid methyl ester	<i>cis</i> -methyl hexadec-9-enoate	2	C16:1	60.40
14	Heptadecanoic acid methyl ester	methyl heptadecanoate	2	C17:0	62.52
15	<i>Cis</i> -10-Heptadecenoic acid methyl ester	<i>cis</i> -methyl heptadec-10-enoate	2	C17:1	66.27
16	Stearic acid methyl ester	methyl octadecanoate	4	C18:0	68.66
17	Elaidic acid methyl ester	<i>trans</i> -methyl octadec-9-enoate	2	C18:1 9 <i>t</i>	70.75
18	Oleic Acid methyl ester	<i>cis</i> -methyl octadec-9-enoate	4	C18:1 9 <i>c</i>	71.83
19	Linoelaidic acid methyl ester	<i>cis, trans</i> -methyl octadeca-2,6-dienoate	2	C18:2 6 <i>t</i>	74.24
20	Linoleic acid methyl ester	<i>cis, trans</i> -methyl octadeca-2,6-dienoate	2	C18:2 6 <i>c</i>	76.58
21	Arachidic acid methyl ester	methyl eicosanoate	4	C20:0	80.09
22	γ -Linoleic acid methyl ester	<i>cis</i> -6,9,12-octadecatrienoate	2	C18:3n6	82.29
23	<i>Cis</i> -11-Eicosenoic acid methyl ester	<i>cis</i> -methyl eicos-11-enoate	2	C20:1	83.07
24	Linolenic acid methyl ester	<i>cis</i> -methyl octadeca-9,12,15-trienoate	2	C18:3n3	83.11
25	Heneicosanoic acid methyl ester	methyl heneicosanoate	2	C21:0	85.74
26	<i>Cis</i> -11,14-Eicosadienoic acid methyl ester	<i>cis</i> -methyl icoso-11,14-dienoate	2	C20:2	87.94
27	Behenic acid methyl ester	methyl docosanoate	4	C22:0	91.33
28	<i>Cis</i> -8,11,14-Eicosatrienoic acid methyl ester	<i>cis</i> -methyl icoso-8,11,14-trienoate	2	C20:3n6	93.57
29	Erucic acid methyl ester	<i>cis</i> -methyl docos-13-enoate	2	C22:1n9	93.92
30	<i>Cis</i> -11,14, 17-Eicosatrienoic acid methyl ester	<i>cis, cis, cis</i> -methyl icoso-11,14,17-trienoate	2	C20:3n3	94.11
31	Arachidonic acid methyl ester	<i>cis</i> -methyl icoso-5,8,11,14-tetraenoate	2	C20:4n6	96.33
32	Tricosanoic acid methyl ester	methyl tricosanoate	2	C23:0	96.41
33	<i>Cis</i> -13,16-Docosadienoic acid methyl ester	<i>cis</i> -methyl docosa-13,16-dienoate	2	C22:2	98.70
34	Lignoceric acid methyl ester	methyl tetracosanoate	4	C24:0	99.44
35	<i>Cis</i> -5,8,11,14, 17-Eicosapentaenoic acid methyl ester	<i>cis</i> -methyl icoso-5,8,11,14,17-pentaenoate	2	C20:5n3	101.63
36	Nervonic acid methyl ester	<i>cis</i> -methyl tetracos-15-enoate	2	C24:1	104.35
37	<i>Cis</i> 4,7,10,13, 16,19-Docosahexaenoic acid methyl ester	<i>cis</i> -methyl docosa-4,7,10,13,16,19-hexaenoate	2	C22:6n3	112.32

b) The *trans* fatty acid methyl ester standards were obtained from Sigma Chemicals and were prepared at concentrations mentioned below:

S. No	Trivial Name	IUPAC Name	Percentage	Carbon Number	Retention time (minutes)
1	Internal Standard Triundecanoin methyl ester		2		32.41
2	Myristelaidic acid methyl ester	<i>trans</i> -methyl tetradec-9-enoate	2	C14: 1 9 <i>t</i>	48.15
3	Palmitelaidic acid methyl ester	<i>trans</i> -methyl hexadec-6-enoate	2	C16: 1 6 <i>t</i>	59.00
4	Petroselaaidic acid methyl ester	<i>trans</i> -methyl Octadec-6-enoate	2	C18: 1 6 <i>t</i>	70.30
5	Elaidic acid methyl ester	<i>trans</i> -methyl octadec-9-enoate	1.5	C18: 1, 9 <i>t</i>	70.53
6	Vaccenic Acid methyl ester	<i>trans</i> -methyl octadec-11-enoate	1.5	C18: 1 11 <i>t</i>	70.76
7	Linoleic acid methyl ester	<i>trans, trans</i> -methyl octadeca-9,12-dienoate (50%)	2.5	C18: 2 9 <i>t</i> ,12 <i>t</i>	74.20
8		<i>cis, trans</i> -methyl octadeca-12-enoate (20%)	1	C18: 2 12 <i>t</i>	75.36
9		<i>trans, cis</i> -methyl octadeca-9,12-dienoate (20%)	1	C18: 2 9 <i>t</i>	75.84
10		<i>cis, cis</i> -methyl octadeca-9,12-dienoate (10%)	0.5	C18: 2 9 <i>c</i> ,12 <i>c</i>	76.41
11	Linolenic acid methyl ester	<i>trans, trans, trans</i> -methyl octadeca-9,12,15-trienoate (30%)	1.5	C18:3 9 <i>t</i> ,12 <i>t</i> ,15 <i>t</i>	78.22
12		<i>trans, trans, cis</i> -methyl octadeca-9,12,15-trienoate (15%)	0.75	C18:3 9 <i>t</i> ,12 <i>t</i> ,15 <i>c</i>	79.51
13		<i>trans, cis, trans</i> -methyl octadeca-9,12,15-trienoate (15%)	0.75	C18:3 9 <i>t</i> , 12 <i>c</i> ,15 <i>t</i>	79.67
14		<i>cCis, trans, trans</i> -methyl octadeca-9,12,15-trienoate (15%)	0.75	C18:3 9 <i>c</i> , 12 <i>t</i> , 15 <i>t</i>	80.14
15		<i>cis, cis, trans</i> -methyl octadeca-9, 12, 15-trienoate(7%)	0.35	C18:3 9 <i>c</i> , 12 <i>c</i> , 15 <i>t</i>	80.21
16		<i>cis, trans, cis</i> -methyl octadeca-9,12,15-trienoate (7%)	0.35	C18:3 9 <i>c</i> ,12 <i>t</i> ,15 <i>c</i>	81.17
17		<i>trans, cis, cis</i> -methyl octadeca-9,12,15-trienoate (7%)	0.35	C18:3 9 <i>t</i>	81.36
18	<i>cis, cis, cis</i> -methyl octadeca-9,12,15-trienoate (4%)	0.2	C18:3 9 <i>c</i> , 12 <i>c</i> ,15 <i>c</i>	81.60	
19	Eicosenoic acid methyl ester	<i>trans</i> -methyl Eicos-11-enoate	5	C20:1 11 <i>t</i>	82.01
20	Brassicidic acid methyl ester	<i>trans</i> -methyl docos-13-enoate	5	C22:1 13 <i>t</i>	92.81

c) Triglyceride internal standard solution- C11:0 – triundecanoin; 5mg/ml in CHCl₃ was prepared

12.5 Preparation of Fatty acid Methyl esters (FAME)

FAME of the samples were prepared according to AOAC Official Method 969.33 Fatty acids in Oils and Fats. Preparation of methyl esters by Boron Trifluoride method. Glycerides and phospholipids are saponified and fatty acids are esterified in the presence of BF₃ catalyst and analysed by GC-FID.

200 mg of oil sample was taken in a flask and then added 4 ml of methanolic Sodium Hydroxide (0.5M) and boiling chip. Attached condenser, and refluxed until fat globules disappear (10 min). 5.0

ml of BF₃ solution from auto pipette was added through condenser and boiled for 2 minutes. 2ml heptane was added through condenser and boiled for 1 minute. Removed heat and then added 15 ml saturated NaCl solution. Stoppered flask and shook vigorously for 15 seconds solution was tepid. Added additional saturated NaCl solution to float heptane solution into neck of flask. Transferred upper heptane solution into glass stoppered test tube and added small portion of Na₂SO₄ to remove H₂O. Diluted solutions to concentration of 10% for GC determination.

12.6 Chromatography

Obtained relative retention times (vs FAME of triglyceride internal standard solution) and response factors of individual FAMES by GC analysis of individual FAME standard solutions and mixed FAME standard solutions.

Injected 1µl each of individual FAME standard solutions and 1µl each of mixed FAMES standard solution (saturated, unsaturated and *trans*). Used mixed FAME standard solutions to optimise chromatographic response before injecting the test solution. Injected 1 µl of test solution into GC column.

12.7 Calculations

(Note: For any unknown or uncalibrated peaks, use the nearest calibrated fatty acid response factors and conversion factors to calculate total, saturated and monounsaturated fats.)

Calculate the empirical response factor for each fatty acid as follows:

$$R_i = \frac{P_{S_i} \times W_{C11:0}}{P_{SC11:0} \times W_i}$$

P_{S_i} = Peak Area of individual fatty acids in mixed FAMES standard solution

P_{S_{C11:0}} = Peak area of C11:0 fatty acid in mixed FAMES standard solution

W_{C11:0} = Weight of internal standard in mixed FAMES standard solution and

W_i = Weight of individual FAME in mixed FAMES standard solution.

Determine relative retention times for each FAME in FAMES standard solution relative to C11:0 and use them to identify various FAMES in test samples

Calculate amount of individual triglycerides) (W_{TGi}) in test portion as follows

$$W_{FAMEi} = \frac{P_{t_i} \times W_{t_{C11:0}} \times 1.0067}{P_{t_{C11:0}} \times R_i}$$

$$W_{TGi} = W_{FAMEi} \times f_{TGi}$$

Where P_{t_i} = peak area of fatty acid i in test portion

W_{t_{C11:0}} = weight of C11:0 internal standard added to test portion, g

1.0067 = conversion of internal standard from triglyceride to FAME

P_{t_{C11:0}} = peak area of C11:0 internal standard in test solution

f_{TGi} = conversion factors for FAME to triglycerides for individual fatty acids

Calculate weight of each fatty acid (W_i) as follows

$$W_i = W_{FAMEi} \times f_{FAi}$$

f_{Fai} = conversion factor for conversion of FAMES to their corresponding fatty acids

Calculate amount of saturated fat (expressed as fatty acids; (w/w; expressed as saturated fatty acids; sum of C4:0, C6:0, C8:0 etc) as follows:

$$\text{Saturated fat, \%} = (\sum \text{saturated } W_i / W) \times 100$$

Calculate amount of unsaturated fat (expressed as fatty acids; C14:1, C16:1, C18:1, cis and trans, C18:2, C18:3, C20:1 and C22:1) in test solution as follows

$$\text{Unsaturated fat \%} = (\sum \text{unsaturated } W_i / W) \times 100$$

13. Results and Discussion

Total fatty acids profile (saturated and unsaturated fatty acids) comprising 37 components and trans fatty acids pattern (9 trans fatty acid methyl esters) were analyzed in 30 samples of edible oil and fats comprising - edible oil (21 sample), vanaspati (7), desi ghee (1), butter (1).

The total fat content (g/100g of oil or fat or %) and the percentage of saturated fatty acids, unsaturated fatty acids (mono and poly unsaturated) and *trans* fatty acids of 30 samples is provided in Annexure II.

13.1 Fatty acids (saturated and unsaturated)

The results of determination of fatty acid composition detected in edible oil and fat samples compared with the range of standard composition available in literature (Annexure II) indicate that the predominant fatty acid is linoleic acid (C18:2 6c) in soybean (28.1 - 54.3%), sunflower (27.2 - 38.8%) and safflower oils (30.5%). The levels detected are lower than the appropriate ranges for linoleic acid specified in the Codex Standard for named Vegetable oils (Codex Standard 210 (amended 2003, 2005) for soyabean (48-59%), sunflower (48.3 - 74%) and safflower (67.8-73.2%).

Oleic acid (C18:1 9c) was predominant in ground nut (40.6%) and olive oil samples (65.56%). Ground nut sample was below the range specified in the Codex standard for oleic acid for groundnut oil (55- 83%) and the oleic acid content in the olive oil sample analyzed was within the range specified in Codex Standard for Olive oil (55-83%). From nutritional viewpoint, the presence of oleic acid in diet is very useful. It has been shown that oleic acid is effective in lowering LDL content and LDL cholesterol content (Grundy, 1989).

Erucic acid (C22:1 9c) was the predominant fatty acid in the mustard oil samples detected in the range of (24.3-37.2%). The level of this fatty acid was within the range specified for erucic acid content in mustard oil in the Codex Standard (22-50%)

Lauric acid (C12:0) was the predominant fatty acid in the coconut samples detected at the level of 44.6% which is almost close to the range specified for lauric acid content in coconut oil in the Codex Standard (45.1- 53.2%).

Oleic (C18: 1 9c) and Linoleic acid (C18:2 6c) are the predominant acids in sesame oil samples and rice bran oil. The level of oleic acid and linoleic acid in the sesame oil sample was 39% and 38% respectively which is within the range specified for oleic (34.4 - 45.5 %) and linoleic acid (36.9 - 47.9%) in sesame oil in the Codex standard. The level of oleic and linoleic acid in the rice bran

sample was 36.4% and 24.7% respectively which is lower than the range specified for oleic (38 - 46 %) and linoleic acid (33 - 40%) in rice bran oil in the codex standard.

Palmitic (C16:0) and oleic acid (C18:1 9c) are predominant acids in Palm Oil detected in the range of 33.2 - 35.8% and 31.9 - 35.5% respectively. The levels detected are lower than the range specified for Oleic (39.3 - 47.5 %) and linoleic acids (36 to 44 %) in the Codex Standard.

Among all the oils tested high content of alpha-Linolenic acid (Omega 3) was detected in the soyabean oil about 10.9% (range 6.3 - 14.2%) and mustard oil about 7.1% (2.9 -10.5%), which improves the Omega 6/Omega 3 ratio in the diet. Omega 3 may contribute to a reduced risk of fatal Ischemic Heart Disease (IHD) through its antiarrhythmic effect. In cell culture studies Omega 3 was shown to slow the beating rate of isolated neonatal rat cardiac myocytes (Siebert *et al* 1993). Excessive amounts of Omega 6 polyunsaturated fatty acids and very high Omega 6/Omega 3 ratio promote the pathogenesis of many diseases, including cardiovascular diseases, cancer, and inflammatory diseases and autoimmune diseases (Simopoulos, 2002).

13.2 Trans fatty acids

Nine *trans* fatty acids: Myristelaidic acid (*trans* -tetradec-9-enoic acid), Palmitelaidic acid (*trans* -hexadec-6-enoic acid), Petroso elaidic acid (*trans* -octadec-6-enoic acid), Elaidic acid (*trans* -octadec-9-enoic acid), Vaccenic Acid (*trans* - octadec-11-enoic acid, Linoleic acid – 4 isomers (*cis*, *cis*-9,12-octadecadienoic acid *trans,trans*-9,12; *cis,trans*-9,12; *trans, cis*- 9,12 Octadecadienoic acid), Linolenic acid 8 isomers (*trans, trans, trans*- 9,12,15; *trans, trans, cis* -9,12,15; *trans, cis trans* -9,12,15; *cis,trans, trans*- 9,12,15, *cis,cis,trans*- 9,12,15; *cis trans, cis*- 9,12,15; *trans,cis, cis*- 9,12,15; *cis, cis, cis* - 9,12,15 octadecatrienoic acid), Eicosenoic acid (*trans*- eicos-11-enoic acid), Brassidic acid (*trans* - docos-13-enoic acid) in different oils and fats are given in Annexure III.

None of the samples analysed were completely free of *trans* components. In 21 refined edible oil samples analysed for *trans* fats; *trans* fat content was in the range of 0.08 - 3.3%. Most of the samples were within the *trans* fat limit of Denmark of 2% (2 g per 100 g of oil or fat) except Saffola gold (Fortified Blended oil with the goodness of kardi oil) in which the *trans* fat content was 2.5% (1.2 times above the limit) and Shalimar's Classic Basmati (Refined Rice Bran oil). Elaidic acid (9 *trans* -octadec-9-enoic acid, 18:1 9f) was detected in the range of 0.3 -1.5 % accompanied by positional isomers of *cis, cis*-9,12-octadecadienoic acid *trans,trans*-9,12; *cis,trans*-9,12; *trans, cis*- 9,12 Octadecadienoic acid. Mixed geometric isomers derived from linolenic acid (*trans,trans, cis* - 9,12,15 octadecatrienoic acid ; *cis,,trans, cis* - 9,12,15 octadecatrienoic acid ; *trans,cis, cis* - 9,12,15 octadecatrienoic acid; *cis, cis,,trans* - 9,12,15 octadecatrienoic acid; *trans, cis, trans* - 9,12,15 octadecatrienoic acid; *cis,,trans, trans* - 9,12,15 octadecatrienoic acid were also identified in refined oil samples. In natural vegetable oils, the unsaturated acids are present in the *cis* form. Small amounts of *trans* fatty acid isomers are formed from the natural *cis* isomers in refined edible oils due to high temperatures used during the deodorization process. *Trans* fatty acid content in refined edible oil can be upto 3% (Ackman *et al* 1998).

In Vanaspati, the total *trans* fats in the 7 vanaspati samples ranged from 9.4 - 23.7%. It is 4.7-11.9 times the *trans* fat limit of Denmark of 2%. Elaidic acid (18:1 9f) is the major *trans* fatty acid formed in industrial hydrogenation. The *trans* -octadec-9-enoic acid (Elaidic acid) ranged from 8.5 - 22.0%.

Upto 30% *trans* elaidic acid has been reported in Vanaspati samples (Jeyarani *et al*, 2005). *Trans* unsaturated acids formed during catalytic hydrogenation modify the chemical and nutritional

properties of fats and oils considerably. It has serum cholesterol increasing effect in the presence of dietary cholesterol.

In the one sample of Desi ghee analysed ; *trans, trans, cis* -9,12,15octadeca-trienoic acid (Linolenic acid) was 4.1%. Total *trans* fat content in Desi ghee was 5.3% (2.7 times the limit for *trans* fats in Denmark of 2%) TFA content in Desi ghee is reported upto 2% (Ackman *et al* 1998).

In butter total *trans* fat content was 3.7%, which is 1.9 times the limit for *trans* fats in Denmark of 2%. The *trans* - octadec-11-enoic acid (Vaccenic acid) content was predominant in butter 2.8% and lesser amounts of *trans*-tetradec-9-enoic acid (myristelaidic acid) (0.3%), *trans* -hexadec-6-enoic acid (palmitelaidic acid) was (0.1%), *trans* -octadeca-6-dienoic acid (linoelaidic acid) was 0.1 %, *trans* – octadeca-6-dienoic acid (linoleic acid) was 0.1% and *trans*- eicos-1-enoic acid was 0.2%

Out of 30 samples analysed none of the samples (edible oil, vanaspati, desi ghee and butter) were free of *trans* fats. Highest concentration of *trans* fats was found in vanaspati samples followed by desi ghee, butter and least in edible oils.

13.3 Recommended dietary ratios of Omega 6 to Omega 3 and PUFA to SFA

The World Health Organization (WHO) recommends polyunsaturated fatty acid (PUFA)/saturated fatty acids (SFA) ratio of 0.8 to 1.0 and linoleic acid (Omega6) /alpha linolenic acid (Omega3) ratio of 5-10 in the diet. Polyunsaturated to saturated fatty acid ratio and Omega 6 to Omega 3 ratio of the 30 oil and fat samples has been shown in Annexure II.

Polyunsaturated to saturated fatty acid ratio obtained for soybean oil was 2.6 - 4.1; sunflower oil: 1.7 - 4.0; groundnut oil: 1.8; mustard oil: 4.6 - 8.6; safflower oil: 5.9; Blend (Safflower +Rice bran oil): 2.2; coconut oil: nil; olive oil: 0.6; sesame oil: 2.3; rice bran oil: 1.2; and palm oil was 0.2.

Out of all the oils tested PUFA /SFA intake of 1.2 in the Rice bran oil was optimum and also closest to WHO recommendation of 0.8 to 1.

Omega6 to omega 3 for soybean oil was 2.0 - 8.6; sunflower oil: 55.3 - 76.3; groundnut oil: 7.5; mustard oil: 0.02 - 4.1; safflower oil: 88.2; Blend (Safflower +Rice bran oil): 117.3; coconut oil: nil; olive oil: nil; sesame oil: 88.5; rice bran oil: 4.4; and, palm oil: 23.0- 24.8.

Only soyabean (2 - 8.6) and groundnut oil (7.5) provide the optimal ratio of Omega 6 to Omega 3 ratio of 5-10 recommended by WHO. Remaining oils have either very high or low omega 6 to omega 3 ratio.

Monounsaturated fats are highest in Olive oil 67.0% followed by mustard oil 26.9- 67.6% and groundnut oil 43.1%. In recent years there has been an interest in monounsaturated fatty acids as a suitable replacement for saturated fatty acids. Their net effect on serum lipids and proteins is not much different from polyunsaturated fatty acids but they are not as susceptible to oxidation which may play a role in arterogenesis(Krauss, 2000).

No single oil could meet all the requirements of these recommendations but some are nearer to the recommendations.

14. Conclusions

- Total fatty acids profile (saturated and unsaturated fatty acids) comprising 37 components and *trans* fatty acids pattern (9 *trans* fatty acid methyl esters) were analyzed in edible oil (21 sample), vanaspati (7), desi ghee(1), butter (1).
- Variations in the different brands and also detected differences between the labelling claims made by the companies.
- All edible oils contained small amounts of *trans* fatty acids in the range of 0.1 - 3.3%.
- The total *trans* fats in the 7 Vanaspati samples ranged from 9.4 - 23.7% (4.7-11.9 times the *trans* fat limit of Denmark of 2% for fats and oils). Elaidic acid (9 *trans* octadecenoate, 18:1n9t) is the major *trans* fatty acid formed in industrial hydrogenation. The *trans* 9 octadecenoate (Elaidic acid) ranged from 8.5 -22%.
- Total *trans* fat content in Desi ghee was 5.3% which is 2.7 times the limit for *trans* fats in Denmark of 2% for fats and oils.
- In butter total *trans* fat content in was 3.7% which is 1.9 times the limit for *trans* fats in Denmark of 2% for fats and oils.
- Highest concentration of *trans* fatty acids was obtained in Vanaspati (7) samples analysed followed by desi ghee, butter and least in edible oils.
- Industrially produced *trans* fatty acids have an adverse effect on health and increase the development of heart diseases, cancer, diabetes, allergic disorders and obesity. They should not be used in food and should be phased out as soon as possible.
- It is advisable to include a variety of oils in the diet and in moderation.

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Annexure I: Details of the Edible oil samples analysed for fatty acid composition

S. No.	Code No	Brand Name	Oil Type	Company	Place of Manufacture	Batch Number	Date of manufacture	Expiry date	Remarks
1	001	Fortune	Refined Soyabean Oil	Adani Wilmer Ltd	Adani Wilmer Ltd, (Unit No. II), Vill - Dhrub, Taluka Mundra, District Kutch, Gujarat	B.NO (AT) SB08	Nov-06	Best before nine months from packaging.	Made from imported oil, free from Argemone oil, Contains permitted antioxidants and OMG3-Omrga 3 essential PUFA.
2	002	Nature Fresh	Refined Imported Soyabean oil	Cargill India Pvt Ltd	Cargill India Pvt Ltd, Vill - Bhimasar, Kutch, Gujarat - 370240	B.NO 223413 ROLL NO. 838 AK1	Sep-06	Best before nine months from packaging.	Free from Argemone oil, Nutrition Facts (Approximate composition per 100g): Calories-884, Saturates-14.4%, MUS-23.3%, PUS-57.9%.
3	003	Dalda	Refined Soyabean oil	Bunge India Pvt Ltd	JMD oils Pvt Ltd, Survey No. 320, Village Bhimasar, Gandhidham, Kutch, Gujarat - 370110	B.NO GJ 12101	Oct-06	Best before six months from packaging.	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigarseed, Palm, Rice Bran, Salseed, Sesame, Sunflower, Watermelon, Solvent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil; free from Argemone oil.
4	004	Gemini	Refined Imported Soyabean Oil	Cargill India Pvt Ltd	Cargill India Pvt Ltd, Vill - Bhimasar, Kutch, Gujarat - 370240	B.NO 123417 Roll No. 459 M AK1	Nov-06	Best before nine months from packaging.	Free from Argemone oil, Nutrition Facts (Approximate composition per 100g): Calories-884, Saturates-14.4%, MUS-23.3%, PUS-57.9%
5	005	Sweekar	Refined Sunflower Oil	Marico Ltd	A.P organics Pvt Ltd, Vill - Maanwala Saron Road, Dhuri - 148024 (Punjab)	B.NO LOT No JX 149 M/C.A SR No. 387548	Sep-06	Best before six months from manufacture	Low absorb oil, Free from Argemone oil, Contains permitted antioxidants, antifoaming agents.
6	006	Sundrop	Refined Sunflower Oil	Agrotech Foods Ltd	Premier Soya oil Ltd, 100, Industrial Area, Jhotwara - 302012	B.NO 03 E1E302 (JA)	Oct-06	Best before six months from manufacture	Free from Argemone oil, Contains permitted antioxidants (TBHQ), Nutrition Facts (Approximate composition per 20g): Calories-180, Saturates-12%, Mono & Poly Saturates-88%, Added Vitamin A-534 I.U., Added Vitamin D-116 I.U., Vitamin E-10 I.U.
7	007	Gemini	Refined Sunflower Oil	Cargill India Pvt Ltd	Cargill Foods India Ltd, E-4, E-45, MIDC Kurkumbh - 413802, Maharashtra	B.NO 003 A KK	Jul-06	Best before six months from packaging.	Free from Argemone oil, Nutrition Facts (Approximate composition per 100g): Calories-884, Saturates-10.1%, MUS 45.4%, PUS 40.1%.
8	008	Nature Fresh	Refined Imported Sunflower Oil	Cargill India Pvt Ltd	Cargill Foods India Ltd, E-4, E-45, MIDC Kurkumbh - 413802, Maharashtra	B.NO 012 A KK	Sep-06	Best before nine months from manufacture.	Free from Argemone oil, Nutrition Facts (Approximate composition per 100g): Calories-884, Saturates-10.1%, MUS 45.4%, PUS 40.1%.
9	009	RR Primio	Refined Groundnut Oil	RR OOMERBHOY Pvt Ltd,	Elite Tradex Pvt, A-8, Supa-Parner, MIDC, Dist - Ahmednagar - 414301	B.NO E-90	Nov-06	Best before twelve months from manufacture.	Nutritional Facts: MUFA-54%, PUFA-29%, SFA-7%, Calories/10g-88.
10	010	Dhara	Refined Mustard oil	Dhara vegetable oil and foods company Ltd, NDDDB campus, Anand - 388001	Shree Banaskantha oil seeds Growers Cooperative Union Ltd, Palanpur-Deesa Highway, Badarpura, Distt. Banaskantha, Gujarat - 385510	B.NO P1V01	Sep-06	Best before eight months from packaging.	Free from Argemone oil, Rich in content of natural A.L.A.; Representative samples tested in competent laboratories and certified unadulterated.

S. No.	Code No.	Brand Name	Oil Type	Company	Place of Manufacture	Batch Number	Date of manufacture	Expiry date	Remarks
11	011	Dhara	Kachhi Ghani Mustard oil	Dhara vegetable oil and foods company Ltd,	Dhara vegetable oil and foods company Ltd, NDDB campus, Anand - 388001	B.NO A1KO7	Oct-06	Best before eight months from packaging.	Representative samples tested in competent laboratories and certified unadulterated.
12	012	Fortune	Pure Kachhi Ghani Mustard	Adani Wilmer Ltd	SK Solvex Pvt Ltd, E-118-119 Industrial Area, Sama Dungar, Jaipur, Rajasthan	B.NO AJKM 0618	Sep-06	Best before twelve months from packaging.	Representative samples tested in competent laboratories and certified unadulterated.
13	013	Saffola	Refined Kardi oil	Marico Ltd	E-10, MIDC Area, Ajanta Road, Jalgaon - 425003	B.NO LOT J-061	Jun-06	Best before nine months from packaging.	Contains permitted antioxidants and antifoaming agents; Free from Argemone oil.
14	014	Saffola Gold	Fortified Blended oil with the goodness of kardi oil	Marico Ltd	E-10, MIDC Area, Ajanta Road, Jalgaon - 425003	B.NO L.NO JF 379	Sep-06	Best before six months from packaging.	Fortified blended oil with the goodness of Kardi with Vitamin and oryzanol; Contains an admixture of refined Rice Bran oil-70% by weight and Kardi oil-30% by weight, Contains permitted antioxidants and antifoaming agents; Vitamin E-250ppm, Free from Argemone oil.
15	015	Dalda	Vanaspati	Bunge India Pvt Ltd	B-16, Bulandshahar Road, Industrial Area, Site No. 1, Ghaziabad (UP)	B.NO GO 2091	Sep-06	Best before six months from packaging.	Free from Argemone oil, Contains permitted antioxidants (TBHQ), Typical Nutrient analysis: Energy-9KCalory/g, Cholesterol-Nil, PUS-60%, MUS-25%, Saturates-15%, VitaminE-900ppm.
16	016	Rath	Vanaspati	Agrotech Foods Ltd	M/S Kundan Edible products Ltd, Plot No. 1, RIICO Industrial Area, Khushkhera, Bhairwadi, Rajasthan - 301019	B.NO (KE) 119EDE225	Aug-06	Best before six months from manufacture.	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigerseed, Coconut, Palm, Rice Bran, Sesame, Sunflower, Watermelon, Solvent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil, Salseed oil(upto 10%); free from Argemone oil, Contains Vitamin A-25 I.U. and D-2 I.U. when packed.
17	017	Gemini	Vanaspati	Cargill India Pvt Ltd	Global oils & fats Ltd. Post Bhamisar Tq Anjar, Kutch, Gujarat - 370240	B.NO 293427 AK2 Roll No. 472 J	Aug-06	Best before twelve months from packaging.	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigerseed, Palm, Rice Bran, Sesame, Sunflower, Watermelon, Solvent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil, Salseed oil(upto 10%); free from Argemone oil, Contains Vitamin A-25 I.U. and D-2 I.U. when packed.
18	018	Raag	Vanaspati	Adani Wilmer Ltd	Adani Wilmer Ltd, Dhruv Taluka Mundra Kutch, Gujarat	B.NO (AO) VA 05	Sep-06	Best before nine months from packaging.	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigerseed, Palm, Rice Bran, Sesame, Sunflower, Watermelon, Solvent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil

S. No.		Brand Name	Oil Type	Company	Place of Manufacture	Batch Number	Date of manufacture	Expiry date	Remarks
19	019	Jindal	Vanaspati	Jindal oil and fats	Jindal oil & fats Ltd, 9th Km Stone, Chandigarh Road, Talwandi Rana, Hissar - 125001	B.NO 05	Oct-06	Best before six months from packaging.	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigerseed, Palm, Rice Bran, Sesame, Sunflower, Watermelon, Sovent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil
20	020	Mik Food	Desi Ghee	Milk Food Ltd, Punjab	Milk Food Ltd, Village - Agwanpur, Kanth Road 244001, Muradabad	B.NO M157B97	Nov-06	Best before twelve months from manufacture.	
21	021	Panghat	Kachhi Ghani Mustard Oil Grade - 1	Seil Edible oils Ltd	Ganga Industries 1668 -C/2, Thana Road, Najafgarh, New Delhi - 110043	B.NO TF No/LOT 08 NR/2/MO/2006.0	Jul-06	Best before nine months from packaging.	Representative samples tested in competent laboratories and certified unadulterated
22	023	Amul	Pasteurised Butter	Gujarat Co-op. Milk Marketing Federation Ltd., Anand	Mehsana District Co-operative Milk Producers' Union Ltd., Mehsana 384002	MC326	Mar-07	Best before twelve months from packaging when stored refrigerated at 4°C or below.	Ingredient: Butter, Common Salt, Contains permitted natural colour (Annatto). Nutritional Facts per 100g: Energy720kcal, Total Fat-80g, Saturated Fat 51g, Cholesterol 0.18g, sodium 836mg, Total Carbohydrate-0g, Protein-0g, Not a significant source of Dietary Fibre, Sugars, Vitamin C, Calcium and Iron.
23	024	Gagan	Vanaspati	Amrit Banaspati Company Limited, Amrit Nagar, G.T.Road, Ghaziabad – 201 009, Uttar Pradesh	Chandigarh Road, Rajpura – 140 401, Punjab	52	April-07	Best before nine months from manufacture.	Made from any or all of the following permissible vegetable oil: Cottonseed, Mahua, Maize, Palm, Rice Bran, Sesame, Sunflower, Sovent Extracted Ground Nut, Mustard, Rapeseed. Nutritional Information (100g contains):Energy-900K.Cal; Carbohydrates-0g; Proteins-0g; Cholesterol-0g; SFA-40-50g; MUFA-45-65g; PUFA-5-7g (Values are approx.); free from Argemone oil; Contains VitaminA-25IU/g, Vitamin D-2IU/g; Enriched with Til Oil.
24	025	Parachute	Coconut oil	Marico Ltd., Rang Sharda, K.C. Marg, Bandra (W), Mumbai-400050	Plot No.-SP 15, Pipdic Electronics Park, Mannadipet, Thirubhuvanai, Puducherry-605107	PM007	April-07	Best before 15 months from manufacture.	A premium quality 100% pure edible oil made from the finest coconuts.
25	026	Figaro	Olive Oil	Consumer Marketing (India) Pvt. Ltd., 201 Dhantak Plaza, Makwana Road, Marol, Andheri East, Mumbai	Made and Packed in Spain	Lot No. 6252368	Aug-06	Best before 12 months from the date of manufacture	Ingredients: Refined Olive Oil and Extra Virgin Olive oil, 0 mg Cholesterol.
26	027	TilSona	Til Oil	Recon Oil Industries Private Ltd., 5 Chunawala Estate, Kondivitta Road, Mumbai-400059	C/o Modern Goyal Oil Mills, 18-2-45/2, Chandrayngutta, hyderabad-500005	Lot No. 036	May-07	Best before 12 months from the date of manufacture	100% natural and pure, double filtered.

S. No.		Brand Name	Oil Type	Company	Place of Manufacture	Batch Number	Date of manufacture	Expiry date	Remarks
27	028	Panghat	Vanaspati	Siel Edible Oils Ltd., 5th Floor, Kirti Mahal, 19, Rajendra Place, New Delhi-110008	Sangrur Agro Ltd., Rural Focal Point, Village Bhindran, Distt. Sangrur-148001 (Punjab).	B.No. 09	April-07	Best before six months from manufacture	Made from any or all of the following permissible vegetable oil: Cottonseed, Kardi, Mahua, Maize, Nigerseed, Palm, Rice Bran, Sesame, Sunflower, Watermelon, Sovent Extracted Mustard/Rapeseed/Ground Nut/Sesame Oil, Salseed oil(upto 10%), Vegetable oils imported for edible purpose.; free from Argemone oil, %FFA-0.25(Max); Test for mineral oil- negative.
28	029	Shalimar's Classic Basmati	Refined Rice Bran Oil	Shalimar Agro Tech Pvt. Ltd., Bahadurpura, Hyderabad-500264	Shalimar Agro Tech Pvt. Ltd., bahadurpura, Hyderabad-500264	B.No. 904)	April -07	Best before six months from manufacture	Healthy Cooking Oil. SatUraed Fatty Acid: 24.0%, MUFA:43.0%; PUFA-33.3%. Free from Argemone oil.
29	030	Palm Gold- Active	Refined Palmolein Oil	Sarda Agro Oil Ltd., H.Office: Satamrai, Goganpahad, R.R.Disst., Hyderabad (A.P); B.Office:S.No.287, Thammavaram Village, Near light house, Kakinada-533005	Sarda Agro Oil Ltd., H.Office: Satamrai, Goganpahad, R.R.Disst., Hyderabad (A.P); B.Office:S.No.287, Thammavaram Village, Near light house, Kakinada-533005	B.No. SAOL/07	April -07	Best before eight months from packaging.	Free from Argemone oil
30	031	Ruchi Gold	Refined Palmolein Oil	Ruchi Infrastructure Ltd., 706, Tulsiani Chambur, Nariman Point, Mumbai-400024	Manufactured By: Ruchi Infrastructure Ltd., New Port Area, Beach Road, Kakinada-533008 (A.P.); Packed By: PAMBAN AGRO FOODS Pvt. Ltd., New Port Area, Beach Road, Kakinada-533008 (A.P.).	B.No. 707072	July-07	Best before six months from packaging.	Made from Imported Oil. Free from Argemone oil

Annexure II: Fatty acid profile of 30 edible oil and fat samples

S.No.	Code	Brand	Type	Percentage (g/100g oil or fat)									Trans fats compared to Denmark standards of 2% (no. of times)	PUFA/SFA (WHO: 0.8-1)	Omega6/Omega3 (WHO: 5-10)	Predominant fatty acid
				SFA	MUFA	PUFA	TUFA	Omega 9	Omega 6	Omega 3	Total trans fatty acids	Total fatty acids				
Soyabean			Standard Composition	10.1-17.1	17-31.1	52.5-70.1		17-30%	48-59%	4.5-11%						
1	001	Fortune	Refined Soyabean Oil	16.1	22.5	66.3	88.8	22.2	54.3	6.3	1.5	106.4	0.7	4.1	8.6	Linoleic acid
2	002	Nature Fresh	Refined Imported Soyabean Oil	15.3	16.7	47.5	64.2	16.7	29.7	14.2	1.5	81.0	0.7	3.1	2.1	
3	003	Dalda	Refined Soyabean Oil	17.1	17.4	44.7	62.1	17.4	28.1	13.7	0.4	79.6	0.2	2.6	2.1	
4	004	Gemini	Refined Soyabean Oil	13.5	16.2	54.3	70.5	16.2	44.2	9.3	1.5	85.5	0.8	4.0	4.7	
Sunflower			Standard Composition	8.1-17.1	14-40.4	48.3-74.6		14-39.4%	48.3-74.0%	0-0.3%						
5	005	Sweekar	Refined Sunflower Oil	16.5	47.1	27.6	74.7	47.1	27.3	0.4	0.1	91.4	0.0	1.7	76.3	Linoleic acid
6	006	Sundrop	Refined Sunflower Oil	8.8	47.7	35.4	83.2	47.4	34.9	0.5	0.3	92.2	0.1	4.0	65.3	
7	007	Gemini	Refined Sunflower Oil	11.7	23.7	39.4	63.1	23.7	38.8	0.6	1.5	76.3	0.7	3.4	69.2	
8	008	Nature Fresh	Refined Imported Sunflower Oil	16.8	22.8	32.0	54.8	22.8	31.3	0.6	0.9	72.5	0.4	1.9	55.7	
Groundnut			Standard Composition	12-27.8	35.7-71.6	12-43.3		35.0-69.0%	12.0-43.0%	0.0-0.3%						
9	009	RR Pmio	Refined Groundnut Oil	16.1	43.1	28.6	71.7	40.6	24.7	3.3	0.9	88.7	0.5	1.8	7.5	Oleic acid

S.No.	Code	Brand	Type	Percentage (g/100g oil or fat)									Trans fats compared to Denmark standards of 2% (no. of times)	PUFA/SFA (WHO: 0.8-1)	Omega6/Omega3 (WHO: 5-10)	Predominant fatty acid
				SFA	MUFA	PUFA	TUFA	Omega 9	Omega 6	Omega 3	Total trans fatty acids	Total fatty acids				
Mustard			Standard Composition	1.2 - 12.0	35.5- 89.0	16-44		8.0- 23.0%	10.0- 24.0%	6.0- 18.0%						
10	010	Dhaara	Refined Mustard Oil	3.6	62.6	24.3	86.8	9.4	11.9	2.9	0.6	91.1	0.3	6.8	4.1	Erucic acid
11	011	Dhaara	Kacchi Ghaani Mustard Oil	5.0	42.9	39.7	82.6	11.6	14.9	10.5	1.0	88.5	0.5	8.0	1.4	
12	012	Fortune	Pure Kacchi Ghaani Mustard Oil	5.6	67.6	26.0	93.6	8.8	13.0	9.0	0.4	99.7	0.2	4.6	1.4	
13	021	Panghat	Kachhi Ghani Mustard Oil Grade - 1	3.6	26.9	31.3	58.2	0.7	0.1	6.0	0.2	62.0	0.1	8.6	0.0	
Safflower			Standard Composition	7.4- 12.8	8.5- 23.9	67.8- 83.3		8.4- 21.3%	67.8- 83.2%	0.0-0.1%						
14	013	Saffola	Refined Kardi oil	8.5	11.4	50.1	61.5	11.4	49.4	0.6	1.0	71.1	0.5	5.9	88.2	Linoleic acid
Blended Oil																
15	014	Saffola Gold	Fortified Blended oil with the goodness of kardi oil	15.8	25.3	34.6	59.9	24.9	30.5	0.3	2.4	78.1	1.2	2.2	117.3	Oleic and linoleic acid
Coconut			Standard Composition	81.0- 107.3	5.0- 10.2	1.0-2.7		5.0- 10.0%	1-2.5%	0.0-0.2%						
16	025	Parachute	Coconut oil	88.9	6.7	1.0	7.8	6.7	1.0	0.0	0.3	96.9	0.1	0.0	0.0	Lauric acid
Olive			Standard Composition	8-17.1	55.0- 84.6	3.5- 22.5		55-83%	3.5- 21.0%	0.0-1.5%						
17	026	Figaro	Olive Oil	14.2	67.0	7.9	74.9	65.6	6.9	0.0	0.9	89.9	0.4	0.6	0.0	Oleic acid
Sesame			Standard Composition	12.7- 17.1	34.4- 46.1	37.1- 48.9		34.4- 45.5%	36.9- 47.9%	0.2-1.0%						
18	027	TilSonna	Sesame Oil	16.8	39.0	39.2	78.2	39.0	38.1	0.4	1.3	96.3	0.7	2.3	88.5	Oleic and linoleic acid
Rice bran			Standard Composition	15.3- 27.8	38.0- 47.0	33.2- 42.9		38.0- 46.0%	33.0- 40.0%	0.2-2.9%						
19	029	Shalimar's Classic Basmati	Refined Rice Bran Oil	25.4	36.6	31.4	68.0	36.4	24.7	6.2	3.3	96.7	1.7	1.2	4.0	Oleic and linoleic acid

S.No.	Code	Brand	Type	Percentage (g/100g oil or fat)									Trans fats compared to Denmark standards of 2% (no. of times)	PUFA/SFA (WHO: 0.8-1)	Omega6/Omega3 (WHO: 5-10)	Predominant fatty acid
				SFA	MUFA	PUFA	TUFA	Omega 9	Omega 6	Omega 3	Total trans fatty acids	Total fatty acids				
Palm oil			Standard Composition	43.3-57.4	36.0-45.0	9.0-12.5		36.0-44.0%	9.0-12.0%	0.0-0.5%						
20	030	Palm Gold-Active	Refined Palmolein Oil	49.1	35.5	10.7	46.3	35.5	10.2	0.4	0.9	96.3	0.5	0.2	24.8	Palmitic +Oleic acid
21	031	Ruchi Gold	Refined Palmolein Oil	42.5	31.9	10.3	42.2	31.9	9.7	0.4	1.0	85.7	0.5	0.2	23.0	
Vanaspati																
22	015	Dalda	Vanaspati	28.7	8.4	40.7	49.0	6.3	2.8	0.3	9.4	87.1	4.7	1.4	10.4	Palmitic+Eicosatrienoic acid
23	016	Rath	Vanaspati	38.5	21.6	2.4	24.0	21.4	2.2	0.2	15.9	78.4	7.9	0.1	10.4	Palmitic+Oleic acid
24	017	Gemin	Vanaspati	44.5	37.6	3.5	41.1	37.6	2.5	0.1	12.7	98.4	6.4	0.1	23.2	Palmitic+Oleic acid
25	018	Raag	Vanaspati	46.1	17.6	3.2	20.7	17.5	3.2	0.0	23.3	90.2	11.7	0.1	0.0	Palmitic+Oleic acid
26	019	Jindal	Vanaspati	46.1	4.2	1.0	5.2	4.2	1.0	0.0	13.8	65.0	6.9	0.0	0.0	Palmitic
27	024	Gagan	Vanaspati	43.2	20.0	4.8	24.8	20.0	4.7	0.0	14.8	82.8	7.4	0.1	0.0	Palmitic+Oleic acid
28	028	Panghat	vanaspati	41.6	21.3	1.7	23.0	21.2	0.2	0.7	23.7	88.3	11.9	0.0	0.3	Palmitic+Oleic acid
Desi Ghee																
29	020	Milk Food	Desi Ghee	55.9	21.1	10.6	31.7	17.8	6.9	1.0	5.3	93.0	2.7	0.2	6.7	Palmitic + Oleic acid
Butter																
30	023	Butter	Amul	59.4	19.0	4.1	23.0	15.2	1.3	2.4	3.7	86.2	1.9	0.1	0.5	Palmitic + Oleic acid

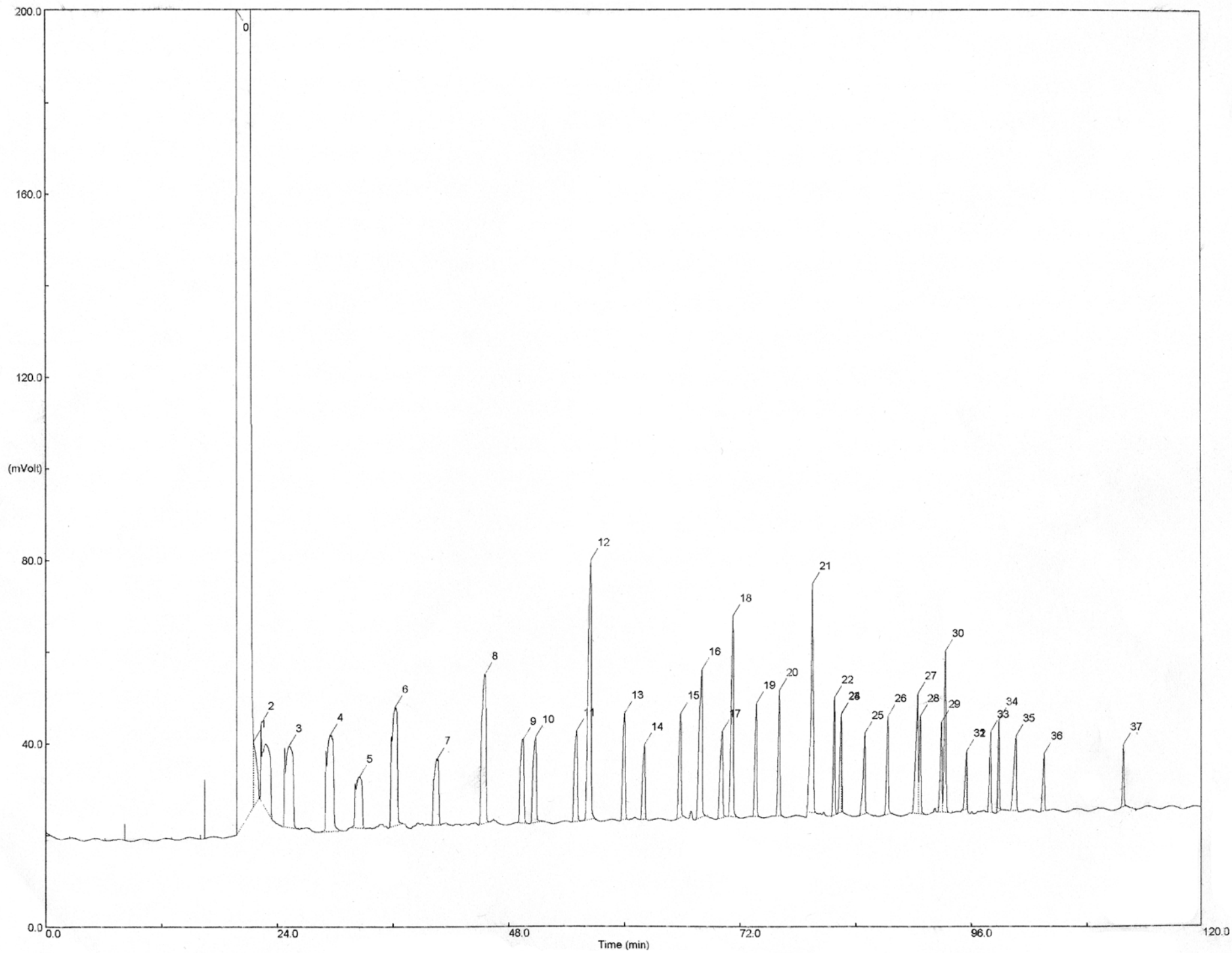
Note:

1. Standard composition for edible oil from Codex Standard for named vegetable Oils -Codex -Stan 210 (Amended 2003,2005)
2. SFA denote 'Saturated fatty acids'; MUFA denote 'Monounsaturated fatty acids'; PUFA denote 'Polyunsaturated fatty acids'; TUFA denote 'Total unsaturated fatty acids'
3. Omega 9 is Oleic acid C18:1 9c; Omega 6 is Linoleic acid C18:2 6c; Omega 3 is Linolenic acid 18:3 3c

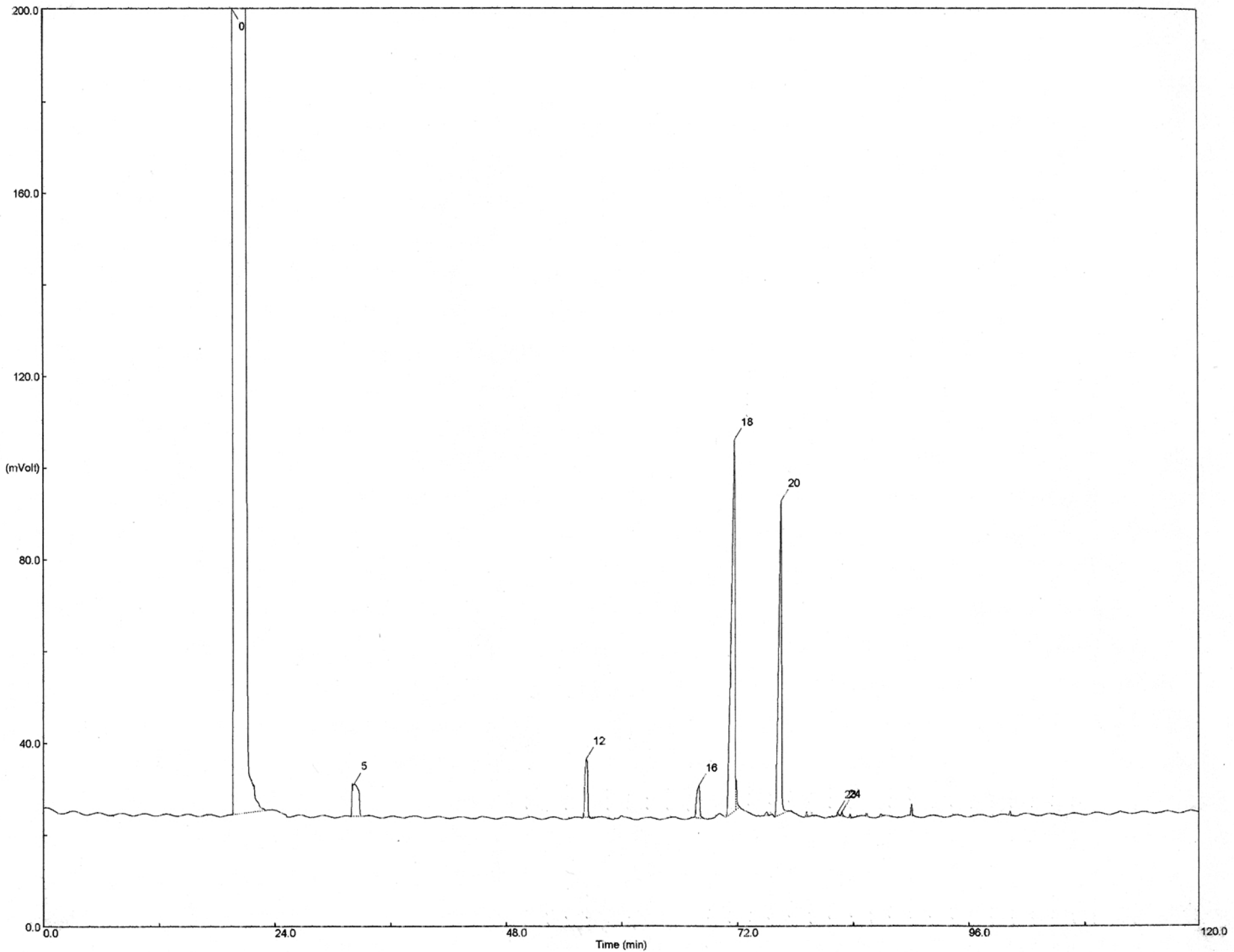
Annexure III: Trans fatty acids profile of edible oils and fats

S.No.	Code No.	Food Item	Type	Trans fatty acids (gm per 100 gm oil or fat/ percentage)								
				Myristelaiddic acid	Palmitelaiddic acid	Elaidic acid	Vaccenic acid	Linoleic acid	Linoleic acid	Linoleic acid	Linoleic acid	Linoleic acid
			Carbon number	C14:19t	C16:1 6t	C18:19t	C18:1 11t	C18:2 6t	C18:2 9t	C18:212t	C18:2 9t,12t	C18:2 9t,15t
1	001	Fortune	Refined Soyabean Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
2	002	Nature Fresh	Refined Imported Soyabean Oil	0.0	0.0	0.4	0.0	0.2	0.0	0.0	0.0	0.0
3	003	Dalda	Refined Soyabean Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0
4	004	Gemini	Refined Soyabean Oil	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0
5	005	Sweekar	Refined Sunflower Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
6	006	Sundrop	Refined Sunflower Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.0
7	007	Gemini	Refined Sunflower Oil	0.0	0.0	0.9	0.0	0.2	0.0	0.2	0.2	0.0
8	008	Nature Fresh	Refined Imported Sunflower Oil	0.0	0.0	0.7	0.0	0.0	0.0	0.1	0.0	0.0
9	009	RR Primio	Refined Groundnut Oil	0.0	0.0	0.4	0.0	0.0	0.0	0.0	0.0	0.0
10	010	Dhaara	Refined Mustard Oil	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0
11	011	Dhaara	Kacchi Ghaani Mustard Oil	0.0	0.0	0.4	0.0	0.0	0.0	0.0	0.0	0.0
12	012	Fortune	Pure Kacchi Ghaani Mustard Oil	0.0	0.0	0.3	0.0	0.0	0.0	0.0	0.0	0.0
13	021	Panghat	Kachhi Ghani Mustard Oil Grade - 1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
14	013	Saffola	Refined Kardi oil	0.0	0.0	0.7	0.0	0.0	0.0	0.1	0.1	0.0
15	014	Saffola gold	Fortified Blended oil with the goodness of kardi oil	0.0	0.0	0.8	0.0	0.0	0.0	0.8	0.0	0.0
16	025	Parachute	Coconut oil	0.0	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0
17	026	Figaro	Olive Oil	0.0	0.0	0.6	0.0	0.0	0.0	0.0	0.0	0.0
18	027	TilSonna	Sesame Oil	0.0	0.0	1.3	0.0	0.0	0.0	0.0	0.0	0.0
19	029	Shalimar's Classic Basmati	Refined Rice Bran Oil	0.0	0.0	1.5	0.0	0.0	0.8	0.4	0.0	0.0
20	030	Palm Gold-Active	Refined Palmolein Oil	0.0	0.0	0.4	0.0	0.0	0.2	0.1	0.0	0.0
21	0.31	Ruchi Gold	Refined Palmolein Oil	0.0	0.0	0.6	0.0	0.0	0.1	0.1	0.0	0.0
22	015	Dalda	Vanaspati	0.0	0.0	8.5	0.0	0.0	0.2	0.1	0.0	0.0
23	016	Rath	Vanaspati	0.0	0.0	14.6	0.0	0.2	0.4	0.3	0.2	0.0
24	017	Gemin	Vanaspati	0.0	0.0	12.5	0.0	0.0	0.0	0.1	0.1	0.0
25	018	Raag	Vanaspati	0.0	0.0	18.5	0.0	0.0	0.3	0.2	2.0	0.0
26	019	Jindal	Vanaspati	0.0	0.0	13.6	0.0	0.0	0.0	0.1	0.0	0.0
27	024	Gagan	Vanaspati	0.0	0.0	14.3	0.0	0.1	0.1	0.0	0.0	0.0
28	028	Panghat	vanaspati	0.0	0.0	22.1	0.0	0.2	0.8	0.3	0.3	0.0
29	020	Milk Food	Desi Ghee	0.5	0.0	0.0	0.0	0.1	0.0	0.0	0.4	0.0
30	023	Butter	Amul	0.3	0.1	0.0	2.8	0.1	0.1	0.0	0.0	0.0

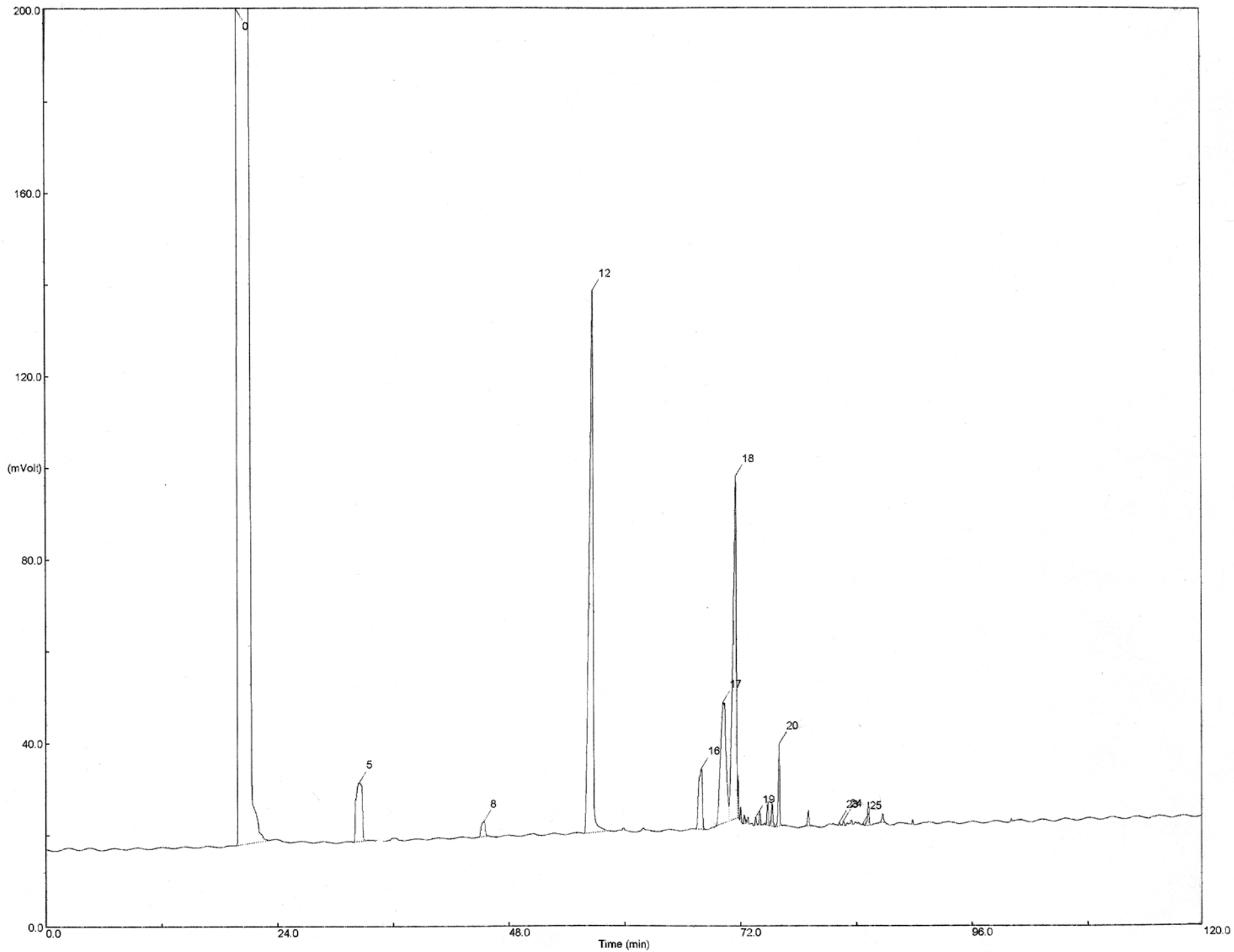
			Trans fatty acids (gm per 100 gm oil or fat/ percentage)									
S.No.	Code No.	Food Item	Type	Linolenic acid	Linolenic acid	Linolenic acid	Linolenic acid	Linolenic acid	Linolenic acid	Eicosenoic acid	Brassicidic acid	Total trans fatty acids
			Carbon number	C18:3 9t,12t	C18:3 9t	C18:3 12t	C18:3 15t	C18:3 9t,15t	C18:3 12t,15t	C20:1 11t	C22:1 13t	
1	001	Fortune	Refined Soyabean Oil	0.2	0.0	0.8	0.0	0.0	0.4	0.0	0.0	1.5
2	002	Nature Fresh	Refined Imported Soyabean Oil	0.0	0.0	0.0	0.6	0.3	0.0	0.0	0.0	1.5
3	003	Dalda	Refined Soyabean Oil	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.4
4	004	Gemini	Refined Soyabean Oil	0.0	0.0	0.0	0.6	0.2	0.0	0.3	0.0	1.5
6	005	Sweekar	Refined Sunflower Oil	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.1
7	006	Sundrop	Refined Sunflower Oil	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3
8	007	Gemini	Refined Sunflower Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.5
9	008	Nature Fresh	Refined Imported Sunflower Oil	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.9
10	009	RR Pmio	Refined Groundnut Oil	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.9
11	010	Dhaara	Refined Mustard Oil	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.6
12	011	Dhaara	Kacchi Ghaani Mustard Oil	0.5	0.0	0.0	0.0	0.0	0.1	0.0	0.1	1.0
13	012	Fortune	Pure Kacchi Ghaani Mustard Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.4
14	021	Panghat	Kachhi Ghani Mustard Oil Grade - 1	0.0	0.0	0.0	0.0	0.0	0.1	0.0	0.2	0.2
15	013	Saffola	Refined Kardi oil	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.0	1.0
16	014	Saffola gold	Fortified Blended oil with the goodness of kardi oil	0.4	0.2	0.0	0.0	0.0	0.1	0.1	0.0	2.4
17	025	Parachute	Coconut oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2
18	026	Figaro	Olive Oil	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.9
19	027	TilSonna	Sesame Oil	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.3
20	029	Shalimar's Classic Basmati	Refined Rice Bran Oil	0.6	0.0	0.1	0.0	0.0	0.0	0.0	0.0	3.3
21	030	Palm Gold-Active	Refined Palmolein Oil	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.9
22	031	Ruchi Gold	Refined Palmolein Oil	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.0
23	015	Dalda	Vanaspati	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	9.4
24	016	Rath	Vanaspati	0.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	15.9
25	017	Gemin	Vanaspati	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	12.7
26	018	Raag	Vanaspati	2.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	23.3
27	019	Jindal	Vanaspati	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	13.8
28	024	Gagan	Vanaspati	0.2	0.0	0.0	0.0	0.0	0.0	0.0	0.0	14.8
29	028	Panghat	vanaspati	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	23.7
30	020	Milk Food	Desi Ghee	4.1	0.0	0.2	0.0	0.0	0.0	0.0	0.0	5.3
31	023	Butter	Amul	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	3.7

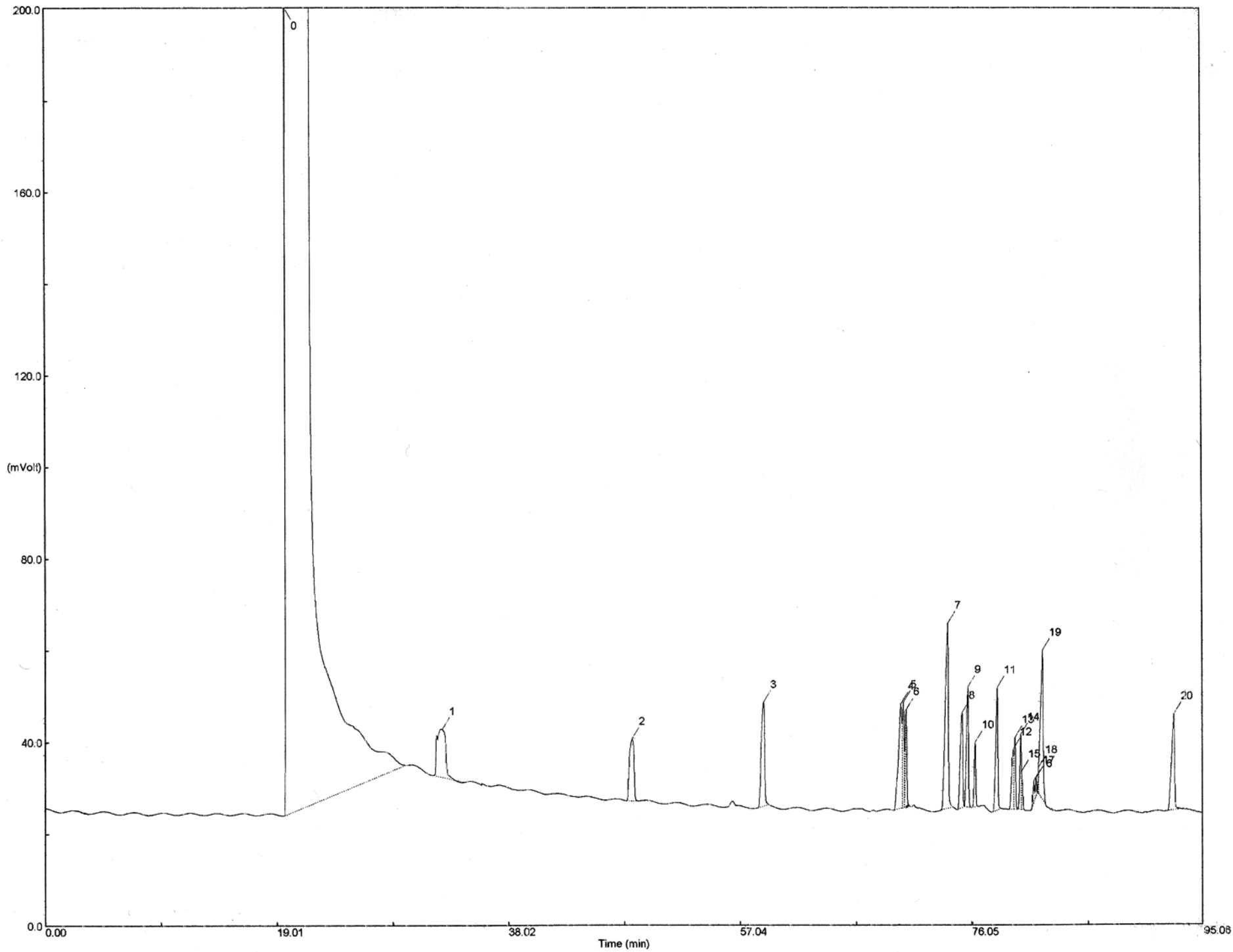


Chromatogram 1: FAME standard mix.

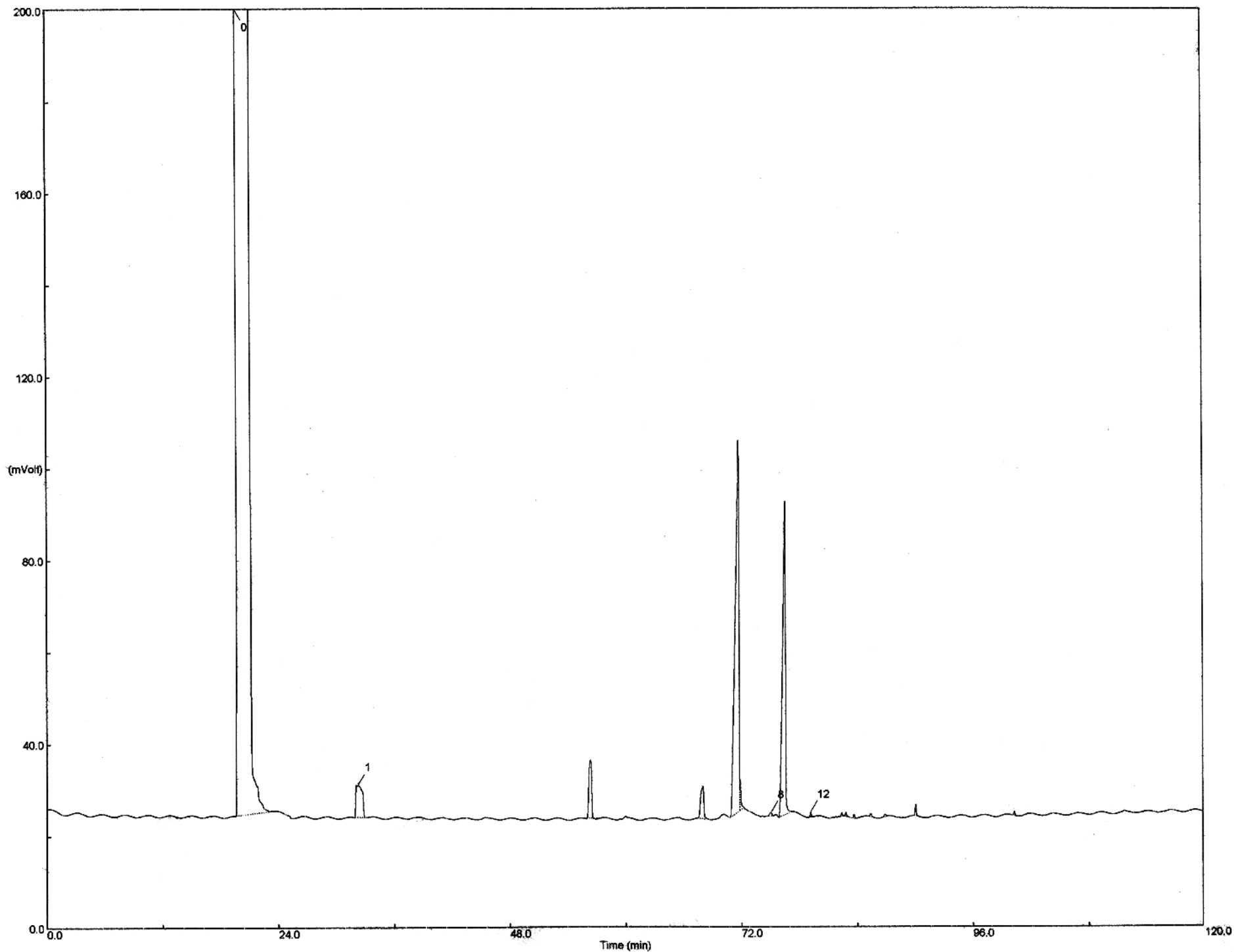


Chromatogram 2: Fatty acid profile of Sundrop-Refined Sunflower Oil

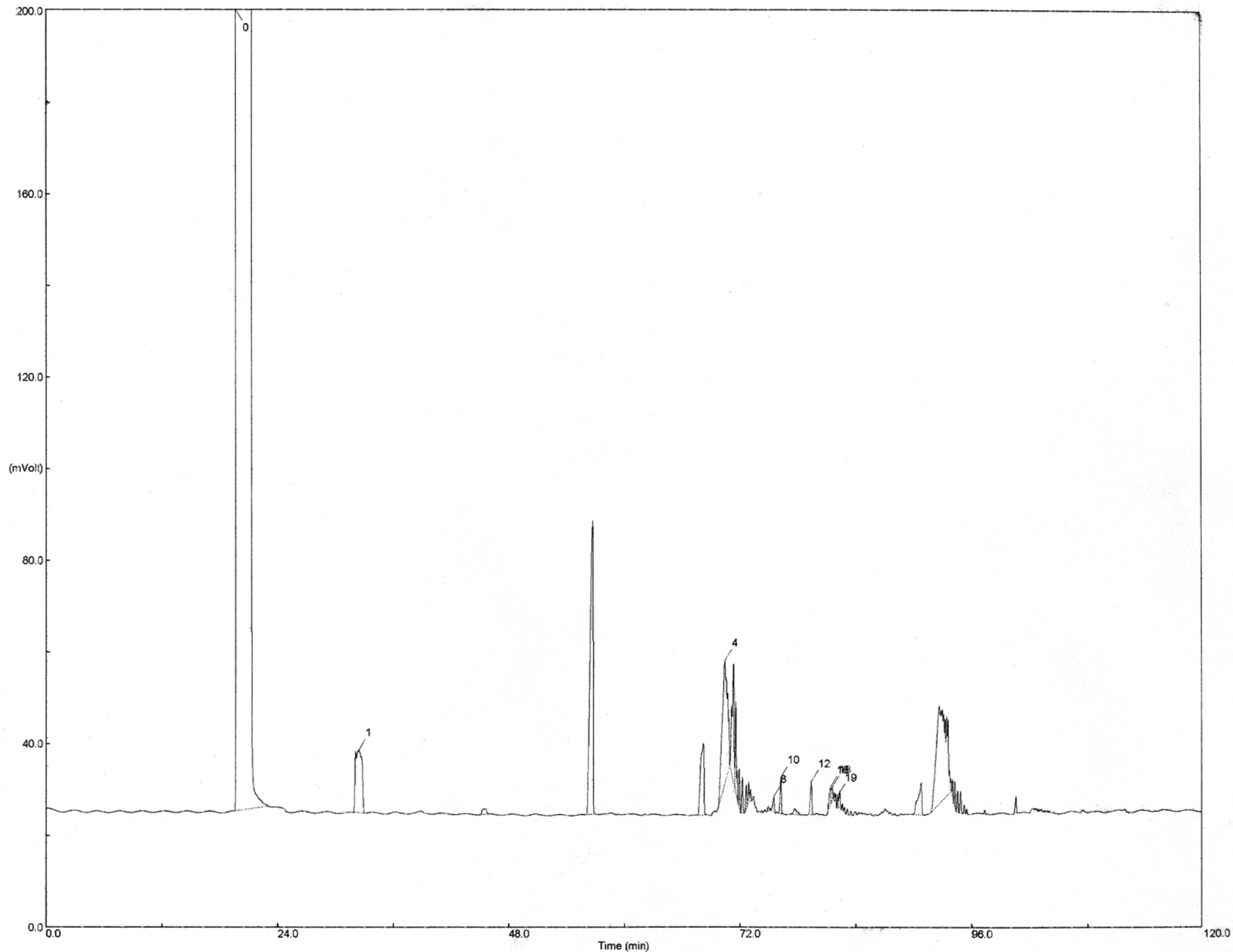




Chromatogram 4: trans FAME standard mix.



Chromatogram 5: trans Fatty acid profile Sundrop-Refined Sunflower Oil.



Chromatogram 6. trans Fatty acid profile of Rath- vanaspati.