CHAPTER II

LITERATURE REVIEW

2.0 DETERMINATION OF OIL QUALITY

The concept of halal products or foods is currently gaining attention globally because of its recognition as an alternative benchmark for safety, hygiene and quality assurance of what people consume daily (Ambali and Bakar, 2014) . Usually, the ingredient label does not list the origin of the ingredients. Muslims require some protection to ensure that information provided on food labels are true and accurate (Eliasi, 2002). Hidden ingredients from various sources present another serious problem for Muslim consumers (Riaz and Chaudry, 2003). Therefore, the increasing demand for clarity in the food industry has enhanced the development of methods for the analysis of food ingredients.

One of the main ingredients in food products or preparations of cooking ingredients are fats and oil. According to Fadzlillah et al. (2011), during the first 50 years of the twentieth century, the use of animal fats in food was very common. For instance, lard (pig fat) was the most used products for domestic frying as well as raw material in mass production of breads and cakes. Moreover, in food industry, it still serves as an important ingredient in the formulation of some food products.

In determining the quality of oil, the particular oil must be resistance from oxidation. The resistance of oil to oxidize is known as the oxidative stability (Guillen and Cabo, 2002). It can be expressed as the period of time necessary to attain the critical point of oxidation in terms of sensorial change or a sudden acceleration of the oxidative process (Silva et al., 2001).

Rancidity has always been associated with changes that lead to the undesirable flavour and odor of lipid. Generally, there are two types of rancidity which are oxidative rancidity and hydrolytic rancidity. Both of the rancidity resulted in reduction of the oil quality, but different in the mechanism. Since oxidation produces low molecular weight off flavour compounds, thus it is an important indicator of oil quality and shelf life (Hamilton, 1994). Oxidation of oil also destroys essential fatty acids and produces toxic compounds and oxidized polymers.

Oxidation of edible oils is influenced by the reaction energy, fatty acid composition, types of oxygen, and minor compounds such as metals, pigments, phospholipids, free fatty acid (FFA), mono- and diacylglycerols (MAG/DAG), thermally oxidized compounds and antioxidants (Choe et al., 2005).

2.1 FACTORS AFFECTING THE OXIDATION OF EDIBLE OIL

2.1.1 Fatty Acid (FA) Compositions of Oils.

The result from Yun and Surh (2012) research showed that fatty acid compositions can be the predictor for initial oxidation state or quality of the vegetable oils. However, the fatty acid composition might not be an appreciable contribution in determining the oxidative stability of the vegetable oil when the oil oxidation could be facilitated (ie; fry at high temperature or storing for long time after opening the oil's bottle). Unsaturated oil will oxidize more quickly than less unsaturated oil. Soybean, safflower or sunflower oil (iodine values more than 130) stored in the dark had a significantly ($P < 0.05$) shorter induction period than coconut or palm kernel oil whose iodine value is less than 20 (Tan et al., 2002). Meanwhile, high oleic and high stearic oils from gene silencing of the oilseeds or hydrogenated soybean oil had higher autoxidative stability (Liu et al., 2002).

2.1.2 Storage Temperature and Light

Ultraviolet (UV) light as well as visible (Vis) light may accelerate autoxidation process by triggering the hydrogen abstraction that will produce the formation of alkyl radicals (Choe and Min, 2006). For example in sweet fennel oil, *trans*- anethole had completely oxidised to anisaldehyde after 2 months of storage at room temperature under light (Misharina and Polshkov, 2005). It has been reported that the effect of light on oil oxidation become less as the storage temperature increases (Velasco and Dobarganes, 2002). The packaging of the oils is very important because the oxidation occurs in the presence of light.

2.1.3 Oxygen

Both concentration and type of oxygen affect the oxidation of oils. Oxygen concentration has a high impact upon the oxidation. The rate of oxidation will be affected by the concentration of dissolved oxygen and oxygen transmission rate (Kanavouras et al., 2006).

At high temperature, the oxygen concentration on the oxidation of oil increased in the presence of light and metals. It is reported that higher oxygen dependence of oil oxidation at high storage temperature is due to low oxygen solubility in the oil at high temperature (Andersson, 1998).

2.2 METHODS OF DETECTION FOR ADULTERANTS

2.2.1 Differential Scanning Calorimetry (DSC)

Thermal analysis has been used for both qualitative and quantitative analysis ranging from various fields such as in pharmaceutical, biological science, minerals, metals and foods. The technique includes differential thermal analysis (DTA), dielectric thermal analysis (DEA), thermomechanical analysis (TMA) and Differential Scanning Calorimetry (DSC).

In the study done by Marina et al. (2009), they monitored the adulteration of virgin coconut with palm kernel oil (PKO) and soybean oil (SBO). They spiked the virgin coconut oil with PKO and SBO from 2 % to 40 % (w/w) separately. Using DSC, they managed to determine the fatty acid of all oil. The results show that PKO adulterated oil did not show any adulteration peak but demonstrated a gradual decrease in the peak height of the major exothermic peak.

Differential scanning calorimetry also can provides a unique thermal profiling for oil as it can be used to detect lard adulteration in virgin coconut oil (VCO). From the experiment done by Mansor et al. (2012), there was one major endothermic peak with a smaller shoulder peak that gradually smoothed out to the major peak as the percentage of lard increased during heating of the thermogram of the mixture.

Meanwhile, during the cooling phase, there was one minor peak and two major exothermic peaks which increased as percentage of lard increased and another peak which decreased in size as the percentage of lard increased. Moreover, both melting as well as cooling profile of VCO is important in determining the presence of lard adulteration. Although TAG and FA analysis by the HPLC and GC–FID are able to detect lard adulteration in VCO with high confidence, they could not provide a qualitative analysis and are limited by the use of chemicals and the requirement for highly trained personnel to operate the systems.

DSC may permits to finger print the primary crystallization of triacylglycerols (TAGs) molecules and their transitional behaviour. Dahimi et al. (2014) assessed the cross contamination caused by lard ranging from 0.5 % to 5 % concentration in the mixture containing beef tallow and chicken fat. The result obtained showed that the discrimination of lard from beef tallow and chicken fat was very obvious even when the concentration was as low as 1 %.

2.2.2 Nuclear Magnetic Resonance (NMR)

Nuclear magnetic resonance is a technique used to determine the compound structure. It is able to identify the carbon hydrogen network in the compound. NMR has many advantages although it is less sensitive than HPLC, GC and capillary electrophoresis. NMR is non-destructive, selective and capable of simultaneous detection of a great number of molecular mass components in a complex mixture. Moreover, the sample preparation is simpler and less time consuming.

NMR can be used to detect trans Fatty Acid in Virgin olive oil. (Sachi et al., 1998). The purity index for virgin olive oils is according to the absentees of trans fatty acids, meanwhile refined olive and olive pomace oils contain detectable levels of oleyl, linoleyl and linolenyl trans isomers. High resolution nuclear magnetic resonance (NMR) spectroscopy coupled with mass spectroscopy (MS) has been used to characterize food products and detects possible adulteration in fruit juices, wine and olive oil (Ogrinc et al., 2003).

There were also research done by Dais et al. (2007) showing that ^{31}P NMR Spectroscopy may be used in determining the quality control and authentication of extra virgin olive oil. The method is based on the derivatization of the labile hydrogens of functional groups, of olive oil compound with the phosphorus reagent 2-chloro- 4, 4, 5, 5-tetramethyldioxaphospholane and the use of the ^{31}P chemical shifts to identify the phosphitylated compounds.

Used frying oil is added to into the commercial qualified vegetable oil by the unscrupulous traders to seek higher profit, therefore the authenticity assurance of the commercial oil remains a challenge in terms of its health and commercial perspectives. Qing Zhang et al. (2013) have made a research which focused on using the Low Field Nuclear Magnetic Resonance to discriminate the adulteration of commercial corn, peanut, rapeseed, and soybean oil with two kinds of used frying oil.

2.2.3 Sensor Electronic Nose

Electric nose were designed to mimic the mammalian olfactory system with the advantage of repeatable measurement, allowing identification and classification of aroma mixtures (Davide et al., 1995). The system consists of multi sensor array, an information processing unit, software with digital pattern recognition algorithm as well as reference library database (Hatfield et al., 1994).

Hai and Wang (2006) used electric nose to detect maize oil adulteration in camellia seed oil and sesame oil. However, the PCA result showed that it cannot be used to discriminate the adulteration of camellia seed oil but can be used for adulteration in sesame oil. Moreover, using ANN (artificial neuro network) model, the electric nose cannot predict the percentage of adulteration in camellia seed oil but it is able to do it in sesame oil.

In the experiment done by Marina et al. (2010), virgin coconut oil was mixed with refined, bleached and deodorized (RBD) palm kernel olein at different level of adulteration. The adulteration peaks were identified using linear regression from the chromatogram profile. The electric nose based on the acoustic wave sensor, was used to generate a pattern of volatile compound present in the sample. Principal component analysis (PCA) was used to differentiate between pure and adulterated samples. The PCA provided good differentiation of samples with 74 % of the variation accounted by PC1 and 17 % accounted by PC2. Pure samples formed a separate cluster from all of the adulterated samples.

2.2.4 Fluorescence Spectroscopy

Fluorescent spectroscopy has the advantages of having high sensitivity and selectivity. Thus, it may be used to investigate real time structure and dynamics in solution as well as under microscopes. It works by measuring the intensity of photons that being emitted from sample after it has absorbed photons.

In the paper written by Poulli et al. (2006), they managed to demonstrate the potential of total synchronous fluorescence (TSyF) spectra to differentiate virgin olive oil from sunflower oil and synchronous fluorescence (SyF) spectra combined with multivariate analysis. The spectrum was acquired by varying the excitation wavelength in the region 270 until 720 nm while the wavelength interval was in the region from 20 to 120 nm. As a result, the TSyF contour plots for sunflower in contrast to virgin olive oil, showed fluorescence region in the excitation wavelength which range from 325 until 385 nm.

The potential application of fluorescence spectroscopy in detecting the adulteration of milk fat with vegetable oil and characterizing the samples according to the source of fat has been studied. Ntakatsane et al. (2013) have adulterated pure butterfat with different vegetable oils at various concentrations which are 0, 5, 10, 15, 20, 30 and 40 %. The 2- and 3- dimensional front face fluorescence spectroscopy and gas chromatography were used to obtain the fluorescence spectra and fatty acid profile based on the total concentration of saturated fatty acid and unsaturated fatty acids, and also on the 3 major fluorophores which are tryptophan, tocopherols and riboflavin. The result showed that fluorescence spectroscopy was able to detect up to 5 % of adulteration of vegetable oil in the butterfat.

2.2.5 Fourier Transform Infrared (FTIR) Spectroscopy

FTIR spectroscopy works according to Beer's law which stated that the intensities of the bands in the spectrum are proportional to the concentration of the corresponding samples (Vlanchos et al., 2006). Over the years in the field of fats and oils, FTIR spectroscopy has received great attention in quantitative analysis because of the 'green analytical chemistry'. The method has been known of the easy sample preparation with reduced or no sample pre- treatment steps (Sherazi et al., 2010).

When combined with partial least square (PLS) model, it has been used in the quantitative analysis of lard in mixture with other animal fats (Che Man and Mirghani, 2001), cake formulation (Syahariza et al., 2005) and chocolate products (Che Man et al., 2005). FTIR has been widely used in researches including vegetable oil. For instance in the authentication of virgin olive oil (Lai et al., 1995) and extra virgin olive oil (Alam and Hamid, 2007).

Rohman et al. (2011) managed to develop a fast technique of FTIR spectroscopy for the detection and quantification of lard in Canola oil, Corn oil, Extra Virgin Olive Oil, Soybean oil, and Sunflower oil. At fingerprints of 1500-1000 cm^{-1}, they are able to quantify and classify the lard in the mixtures with vegetables oil. A minor difference of peak heights between lard and vegetable oils were observed at 1117 cm^{-1} and 1097 cm^{-1} corresponding to C-H bending vibration and C-H deformation vibrations of fatty acids. According to Bendini et al. (2007), in fats and oils, most of the peaks and shoulders of spectrum indicates specific functional groups. Since the main components of both lard and vegetable oil are triglycerides, thus, their spectra look very similar. However, due to the fingerprint technique (no two compounds having the same spectra in terms of amount and intensity of peak) FTIR spectroscopy can be used to extract difference among these oils.

Che Man et al. (2011a) used FTIR spectroscopy to detect the presence of lard in French fries pre-fried in palm oil adulterated with lard. The result showed that the spectra of palm oil and lard were different at frequency 3006 cm^{-1} and the frequency region of 1120-1095cm^{-1}. They exhibit a shoulder band at 3006cm^{-1}. It was associated with the stretching vibration of cis olefinic double bonds (Guillen and Cabo, 1997). The results also showed that lard contained linolenic acyl groups twice as much as palm oil which were later reflected in the spectrum for lard because sharper band at frequency 3006cm^{-1} was observed compared to lard spectrum. Moreover, it was also found that as the ratio of lard in palm oil was increased, the peak area and height will also gradually increased.

FTIR spectroscopy can be used to determine the authentication of *Nigella sativa* oil when combine with chemometrics. Nurrulhidayah et al. (2011b) prepared the binary mixtures of *Nigella sativa* oil and grape seed oil in the concentration ranges of 0.5 %- 60 % (v/v). Eventhough both oils FTIR spectra appear quite similar, there were actually some significant differences either in number of peaks at region 1750cm^{-1} – 1700 cm^{-1}. It was also observed that *Nigella sativa* oil has two sharp peaks in 1744 cm^{-1} and 1710 cm^{-1} while grape seed oil has only one peak at 1744 cm^{-1}. These peak were associated with carbonyl (C=O) stretching vibration (Rohman and Che Man, 2009a).

2.2.6 Gas Chromatography (GC)

Currently, chromatographic techniques such as gas chromatography-mass spectrometry (GS-MS) (Park et al., 2010) gradually become the most important and common techniques to detect adulteration. According to Xie et al. (2013), GC-MS is a common tool for the analysis of separation of fatty acids and sterols.

Gas chromatography equipped with flame ionization detector was used in the experiment to detect the adulteration of olive oil with relatively cheap oil such as soybean oil, sunflower oil and canola oil. The iodine value and the refractive index in the two samples of adulterated oil were significantly higher ($P < 0.01$) when compared to the reference (genuine) olive oil. Using the GC method, it showed that fatty acid (FA) profiles in the two samples exhibited higher amounts of linoleic and linolenic acids but significantly lower amounts for oleic acid (Jafari et al., 2009).

Seo et al. (2010) performed a method for the identification and detection of corn oil in adulterated sesame oil. The fatty acids compositions were determine using GC-FID and IRMS. The result showed that the content of palmitic, linoleic and linolenic acid increased gradually as the mixture rate of corn oil was increased, while the content of stearic acid and oleic acid decreased.

Xie et al. (2013) develop a method of detecting adulteration of camellia seeds oil. They randomly selected camellia seed oil with different compositions of eight soybean oil (particularly the oleic acid content) with different levels of adulteration mixtures at 1, 2, 3, 4, 5, 10, 15, 20, 35, 45 and 50 %. Later, all samples were esterified and analyzed using GC-MS. The result showed that oleic acid (C18:1) and linoleic acids (C18:2) were the predominant fatty acids in all oils samples. Higher oleic acids were found in camellia seed oil (78.19 %-85.63 %) than in soybean oils (23.88 %-27.69 %). As for the linoleic acid, soybean oil contain higher value (50.60 %- 52.23 %) compared to camellia seed oil (6.53 %- 9.49 %).

2.2.7 Headspace

Headspace analysis is normally defined as vapor-phase extraction which involve the partitioning of analytes between a non volatile liquid or solid phase and the vapor phase above the liquid or solid. It is also expected that the vapor phase mixture contains less components than the usual complex liquid or solid sample and this mixture is transferred to a GC (Snow and Slack, 2002).

There are several techniques that could be describes as GC- headspace which are static (vapor- phase extraction) and dynamic (purge and trap) headspace. HS-GC consist of two steps. Firstly, the sample will be placed in a vial that has a gas volume above it, Later this vial will be thermostatted at a constant temperature until they reach the equilibrium. Then, an aliquot of the vial's gas phase will be introduced into the carrier gas stream which will carry it into the column to be analyse. A newer technique such as solid phase microextraction (SPME) uses traps that will help to separate the volatile analytes from the excess of the diluted headspace gas. The dynamic headspace technique is a continuous method of gas extraction and separated the volatile components from the matrix by continuous flow of an inert gas above the sample and it is known as purge and trap (Kolb and Ettre, 2006).

One of the main advantages of using headspace method is the speed of the analysis since no prior sample preparation steps are required, and the simplicity of the measuring process. When coupling a headspace sampler to a mass spectrometer (HS-MS), it is able to recognise complex mixture of volatile compounds, without the associated with gas sensors (Zubritsky, 2000).

Morales et al. (1994) did a research on determining the volatiles in virgin olive oil by using dynamic headspace gas chromatography. It has the advantage of concentrating sample which will make it possible to detect compounds that are present but at low concentration but contribute significantly to the flavour. Tenax TA was used as adsorbent material, thermal desorption and cryofocusing prior to capillary GC to avoid undesirable peak broadening. The result of gas chromatogram of virgin olive oil showed the presence of 100 components, 56 of which were identified in their work. The volatiles identified corresponded to different chemical families such as 7 hydrocarbons, 9 alcohols, 9 aldehydes, 9 ketones, 1 acid, 12 esters, and 2 furans.

Lorenzo et al. (2002) proposed the use of direct coupling of a headspace sampler to a mass spectrometer for the detection of adulterants in olive oil using the application of the linear discriminate analysis technique. They performed the experiment for three different tasks; non adulterated olive oil/ adulterated olive oil, olive oil adulterated with sunflower oil/olive oil adulterated with olive-pomace oil, and non adulterated olive oil /olive oil adulterated with sunflower oil/olive oil adulterated with olive-pomace oil. The result showed that this method has been very good for the three classification tasks addressed, thus it might be used as a screening method.

Pena et al. (2005) develop an analytical method to detect adulteration of virgin olive oils and olive oils with hazelnut oil by a headspace autosampler directly coupled to a mass spectrometer. The end result showed that a minimum adulteration level of 7 % and 15 % can be detected in refined and virgin olive oil respectively.

Headspace analyzer could also be used for the identification of pork for halal authentication. Nurjuliana et al. (2011) study the aroma profiling and identify the components that contribute to the flavour of pork by employing gas chromatography mass spectrometer with headspace analyzer. The result showed that there were a total of 43 volatile components of porks identified by the GC-MS-HS. The majority of the compounds are well known lipid oxidation products including ketones, aldehydes as well as alcohols. Whereas the most detected compounds were aldehydes and ketones. The result obtained also showed that the volatile profile of pork contained a higher proportion of heptanal, which correlated with the experiment done by Shahidi (1994) that aldehydes are the major components identified in the volatiles of cooked pork.

HS-SPME-GC-MS could be used to differentiate between the volatile compound of sunflower oil and high oleic sunflower oil. Hexanal, E-2 heptanal, E-2 decenal and E,E-2,4-nonadienal were the most suitable compounds in differentiating the two oil varieties from each other (Peterson, 2012).

In the study done by Zhao et al. (2013), they did a preliminary investigation to examine and compare the performance of flavours between three types of pure vegetable oils and two types of adulterated oils using HS-GC x GC –TOF/MS. They detected the volatile profile of three types of pure vegetables oils and two adulterated oils which were sesame oil and peanut oil adulterated with soybean oils at six different levels of adulterations 5 %, 10 %, 30 %, 50 %, 70 % and 90 % respectively. They identified 30 common volatile components of the flavour among the three types of vegetable oils such as aldehydes, alcohols, ketones, acids, esters and hydrocarbon, heterocyclic compounds and benzene rings. Meanwhile, in soybean oil, the main volatile compound was E-2-penten-1-ol and E, E-2-4-hexadienal while no particular flavour was obtained from peanut oil compared with another two oil samples.

2.3 PROFILING

2.3.1 RBD Palm Oil Profiling

The colour of refined, bleached and deodorized (RBD) palm oil is normally very light yellow. However, during processing, various components can affect the colour stability of the finished materials and can be a major quality characteristic. The temperature where the oil began to smoke indicates how well it can tolerate heating and reheating. The higher the smoke point, the better it is. Typical frying temperature is about 360 °F (180 °C). During this temperature, unsaturated oils tend to break down or polymerized quickly. Palm oil with a smoke point of 437 °F (225 °C), well above normal frying temperatures, makes it an ideal frying oil. Palm oil has relatively high melting point (Fife, 2007).

In the research done by Sarma et al. (2011), they analyzed for different physicochemical parameters between texturized RBD palm oil and partially hydrogenated RBD palm oil. As for the result for texturized RBD palm oil, it were reported that the moisture and volatile matter was 0.04, the iodine value was 49.61, the acid value was 0.14, the saponification value was 200 and the melting point was 38 °C.

Using GC-MS, Dirinck et al. (1977), were able to examine the volatile trace constituents isolated from palm oil. The result showed that trans-2-octenal, n-nonanal, truns-2-decenal, trans-2-undcenal, b-ionone, cis-2,4-decadienal and trans 2,4-decadienal as important contributors to palm oil.

Nor et al. (2007) did a research on the changes of headspace volatile constituents of palm olein, soybean oil, corn oil and sunflower oil while frying from 2 hours to 40 hours. They found that in palm olein, the 2t, 4t-decadienal content decreased from 15.9 μg g^{-1} (2 hours) to 3.2 μg g^{-1} after 40 hours of frying while hexanal increased from 11.3 μg g^{-1} (2 hours) to 33.8 μg g^{-1} after 40 hours of frying. The low number of hexanal content indicated that palm olein was more stable towards degradation at higher temperature because of the lower content of linoleic acid. Palm olein was expected to produce lower quantity of decadienal as compared to other liquid oils (Boskou et al., 2006).

Petersen et al. (2013) evaluate the volatile compounds that could be the marker for the edible oil deterioration during the production of deep fried French fries. They compare the result between the sensory characteristic and the volatile compounds. 32 hours frying were performed on sunflower oil, rapeseed oil, high oleic rapeseed oil, high oleic sunflower oil and palm olein. They found that after 3 hours of deep frying, E, E- 2, 4-decadienal and heptanal showed the ability to differentiate between sample of various oxidative state, while E, E,2, 4-heptadienal and E, 2-decenal showed a positive correlation with well known lipid oxidation parameters.

Alireza et al. (2010) investigate the effect of frying media and storage time on fatty acids compositions and iodine value (IV) of deep fat fried potato chips. All experiments were conducted at 180 ^{o}C for 5 days. The result for fatty acids analysis showed a decreased in linolenic (C18:3) and linoleic acids (C18:2), while the value for palmitic acid (C16:0) increased when the heating time was prolonged for palm olein. Moreover, there was a significant difference ($p < 0.05$) in terms of IV for each frying oil during the 5 days of heating.

2.3.2 Lard Profiling

Animal fat such as lard and vegetable oils were composed of triacylglycerols (TAG), diacylglycerols (DAG), free fatty acids and other minor components including sterols, caratenoids and fat soluble vitamins (Gunston, 2004). However, according to Andrikopolous et al. (2002), the main classes found in fats and oils are TAGS. Therefore, Rohman et al. (2012) have done an experiment using high liquid performance chromatography with refractive index detector to differentiate the composition of TAG between lard, and other animal fats such as mutton, chicken fat and beef as well as cod liver oil. They found that the main TAG composing lard are palmitoolein (POO), palmitooleostearin (POS) and palmitooleopalmitin (POP) accounting of 21.55 ± 0.08, 14.08 ± 0.04 and 5.10 ± 0.04% respectively.

Dahimi et al. (2013) investigate the use of Gas Chromatography with Flame Ionization Detector (GC-FID) coupled with chemometrics techniques to differentiate lard at very low concentrations in beef and chicken fats. The results obtained showed that lard contains higher fatty acid (FA) of linoleic acid (C18: 2cis) and low palmitic acid (C16:0) but the result was opposite for beef tallow and chicken fats.

The majority of the compounds are well known lipid oxidation products including ketones, aldehydes as well as alcohols. Whereas the most detected compounds were aldehydes and ketones. It was reported that almost all the aldehydes present in pork such as heptanal and nonanal are oxidation products of oleic acid and linoleic acids which were the most abundant unsaturated fatty acids of pork. (Meinert, et al., 2007). The result obtained also showed that the volatile profile of pork contained a higher proportion of heptanal, which correlated with the experiment done by Shahidi (1997) that aldehydes are the major components identified in the volatiles of cooked pork.

Xu et al. (2012) identified a total of 44 volatile compounds in oxidised lard which 13 comprised from aldehydes, 6 acids, 9 alcohols, 6 ketones, 6 hydrocarbons, 3 esters and 1 furan. Moreover, they found that aldehydes were the largest group of volatile compounds based on peak areas. Among these aldehydes, (E)- 2-decenal, 2 - undecenal, (E) - ocetenal, nonanal, octanal, (E, E)- 2, 4- decadienal and (E)- 2-heptenal were predominant in oxidised lard. However, Um et al. (1992) identified that (E)- 2-nonenal, (E)-2-decenal and 2- undecenal were present in the heated beef at high concentrations. When they compare between two breeds of lard (ENL and TCL), hexanal was one of the main volatile oxidation products. Hexanal is a typical oxidation volatile from linoleic acid which acts as source of fatty aroma. (Stahnke, 1994). Moreover, it is also commonly been monitored as a measure for lipid oxidation in foods.

Meanwhile, decomposition of linoleic acid hydroperoxide will produce (E, E)-2, 4- decadienal (Torres et al., 2005) and volatile oxidation of linoleic acid is (E)- 2-heptenal (Lee et al., 2007). Nonanal, (E)- 2- decenal and 2- undecenal has positive relation with oleic acid (Torres et al., 2005). Heptanal and octanal are also originated from oleic acid (Machiel, 2004). At low concentration, these alkanals are important since they contribute to pleasant fruity aroma. However, at high concentration, they will produce sharp and pungent attributes (Paleari et al., 2006). Jelen at al. (2006)

reported that the fatty acid composition in oil will greatly influenced the production of volatile composition compounds.

Xu et al. (2012) detected two most abundant alcohols in lard which are 1-octanol and 1-octen- 3- ol. 1-octen- 3- ol is generated from linoleic acid and the contributor to the off- flavour. 2- pentyl furan was the only furan detected in the two lard sources and is a product from linoleic acid.

Fatty acid methyl esters (FAME) profiles may be used to discriminate lard from other animal fats. In the study done Indrasti et al. (2010), when gas chromatography hyphenated with time- of- flight mass spectrometry (GC-TOF-MS) combine with two different microbore columns (SLB-5 ms and DB-wax), the differentiation of lard from other animal fats by three FAMEs constituents are methyl trans-9,12,15-octadecatrienoate (C18:3 n3t), methyl 11,14,17- eicosatrienoate (C20:3 n3t) and methyl 11,14-eicosadienoate (C20:2n6) which are not present in other animal or plant fats.

2.4 PRINCIPLE COMPONENTS ANALYSIS (PCA)

2.4.1 Introduction

Principle components analysis used a statistical technique to transform an original data set of variables into a smaller set on uncorrelated variables. It has the ability to represent most of the information in the original data set. The technique has been used to wide area such as biology, medicine, chemistry, meteorology geology and social science. Principal components analysis is similar to other multivariate procedures like discriminant analysis and canonical correlation analysis. Both of them involve linear combinations of correlated variables which variables weights in the linear combination that derived on the basis of maximizing some statistical property.

2.4.2 Mathematical Description of PCA

PCA is a chemometrics method for the decomposition of two-way matrices. The variance in the data matrix X, with m observations and n variables, is decomposed by successively estimating principal components (PCs) that capture the variance in the data in scores and loadings. Firstly the X-space is given a coordinate system where each variable gets an axis whose length corresponds to its scaling. Each observation in this space is represented by a point. The average of each variable is then calculated and subtracted (mean centering). This is equivalent to moving the swarm of points to the centre of the coordinate system. Thereafter a function is fitted to the data that describes as closely as possible the variance of the observations in the X-space where it is call first PC.

The influence of each variable (*loading value)* is determine by the angle between the line and each variable axis. One loading value is given for each variable in the data set ($\mathbf{p}_1$). When the first PC has been calculated, the remaining unexplained variance is left in the *residual matrix,* **E**:

$$X = TP' + E$$

After the first PC has been calculated, the next is calculated on the residual matrix $\mathbf{E}_1$, which contains the variance not explained by the first PC.

$$X = t_1p_1' + t_2p_2' + \ldots + t_ap_a' + E$$

The second PC is orthogonal to the first. More PCs can be calculated as long as unexplained information is left. The significant number of principal components can be estimated by different methods, of which cross validation is an often-used method. The variance of a principal component is described by the *eigenvalue*, which is proportional to the variance explained by a PC.

The eigenvalue (λ) can be described as the length of the PC and estimated as the sum of squares of the scores:

$$\lambda = \sum_{m=1}^{M} t_{ma}^{2}$$

where t_{ma} is the score of object m for component a. Hence the length of the score vector is proportional to the importance of the particular PC describing how much of the variance in **X** that is explained (Wiberg, 2004).