Fatty Acid Profiling and Physiochemical Analysis of *Citrus Sinensis* (L) Oil

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ABSTRACT

The study of the physical and chemical properties of oils plays a very vital role in determining their importance and the fields they can be judiciously applied. Many valuable materials are found in oil from orange fruits peels and so oil extracted from the peel of *Citrus sinensis* (sweet orange) using Soxhlet extraction method was assayed for physicochemical properties and also characterized using standard method. The physicochemical analysis was carried out according to American Organization of Analytical Chemists (AOAC)/IUPAC standard methods. The result of the analysis showed that the oil sample contains an acid value of 16.32 mg/g, iodine value (30.46 g/100g), saponification value (805.35mg/g), peroxide value (5.77mg/g), density (0.85 g) and viscosity (0.168 Pa.s.) The obtained lipid was trans esterified by a base catalyst. The methyl esters were analyzed using GC-MS. The lipid profile showed that Elaidic Acid was the most predominant fatty acid in the oil samplehaving the highest value (14.26%), followed by Heptadecanoic Acid (6.36%), while the least was Undecanoic Acid (0.03%. The oil sample contains predominantly monounsaturated fatty acids. However, 61.95% of the entire constituents of the oil sample could not be identified. The peels werefound to be potential source of valuable oil, suitable for numerous industrial applications such as soap, candles and cosmetic industries.

Keywords: Fatty Acid; *Citrus sinensis*; GC-MS; Physicochemical Properties; Trans esterification

INTRODUCTION

Pulp containing fruits are mainly rich in oil and seeds at various ratios. Extraction of oil from such fruits may be from either the fruit or from the pulp as well as from the seeds, depending on the type of fruit.

The chemical structure of fatty acids play a prominent role in the physical and chemical composition of such vegetable oils. Also the glyceride distribution in the oil and their linkage with glycerol are true reflections of the chemical structure of the fatty acids. The biochemical diversity of these oils pose great challenges to both chemists and other scientists alike, hence the need for extensive studies on the fatty acid compositions of these oils as true reflection of the properties of these oils. One of the most important components of plant is the vegetable oil which serves as food in diverse ways, as well as energy source and renewable alternative of fossil fuel.

Citrus as one of the world's major fruit crops is produced in many tropical and subtropical climate Countries. Citrus trees bear some of the most popular fruits and are grown globally for food, medicinal and other industrial applications as oils extracted from fruits and seeds may serve nutritional purposes or as part of raw materials for the chemical production of drying oils, cosmetic emulsifiers, soap and detergents, surface coatings, pharmaceuticals, lubricants, surfactants and fat-liquoring of leather in tanneries (Favela-Hernández *et al*., 2016; Ordoudi *et al*., 2018; Wilczynska & Modrzewski, 2018 and Suri*et al*., 2022).

With a total annual production of nearly 85 million tons, the major citrus producing countries in the world include the following:- Brazil, USA, Japan, China, Mexico, Pakistan as well as countries of the Mediterranean region (i.e., Syria), Africa and South Asia (Khaled *et al*., 2018 and Paw *et al*., 2020).

Orange trees were mainly found in the forests and was planted for the first time in Southern China around 2500BC, before it found its way to the northeast India and Burma (Khaled *et al*., 2018).

Valuable species of Citrus genus abound such as C. medica (citron), C. limon (lemon), C. aurantium (sour orange), C. reticulata (mandarin, tangerine), C. paradise (grapefruit), C. clementina (clementine) and C. sinensis (sweet orange) (Khaled *et al*., 2018). These species are either processed to juices, beverage products, and jams or consumed as fresh fruits with the peel being the main by-product of processing. Two parts of the fruit can be identified anatomically, including the outer peel and the pulp with juice sac glands (Favela-Hernández *et al*., 2016; Salonia *et al*., 2020 and Suri *et al*., 2022). From the peel, the following main parts can be identified: -the outer pigmented flavedo with parenchymatous cells and cuticle, and the white albedo part lying under the flavedo (Suri *et al*., 2022).

The presence of bioactive coumarins (i.e., bergamottin, scopoletin, and umbelliferone), flavonoids (i.e., rutin, quercetin, and kaempferol), limonoids (i.e., limonin and nomilin), and acridone alkaloids (i.e., citruscridoneand citrusinine-I) have been revealed in Citrus plants through phytochemical investigations (Tesfamicael & Ele, 2016; Samuel *et al*., 2017; Mohanan *et al*., 2018; Al juhaimi *et al*., 2018; Mahawar *et al.*, 2020; Wang *et al*; 2020 and Zayed *et al*., 2021). Equally detected in Orange peels were carotenoids, and phenolicphytoconstituents, i.e., phenolic acids, Dietary fiber, minerals, flavanones, and polymethoxylated flavones (Kukeera *et al*., 2015; Ledesma-Escobar *et al*., 2015; Shafiya *et al*., 2018; Mahato *et al*., 2019; Paw *et al*., 2020; Kim *et al.,* 2021; Chew & Ali, 2021; Gedefaw *et al*., 2021 and Afifi *et al*., 2023).

Wide spread application of different plant parts has been reported in manycountries in different recipes to treat various ailments including but not limited to skin inflammation, cough, stomach disorders muscular pain and nausea, as well as being used as a slimming agent (Atolani *et al*., 2020).

Due to the presence of many chemical elements, orange fruit was considered as one of the most important food materials. Orange peels contain Essential oil which is one of the active substances against fungi and bacteria (Boudkhili *et al.,* 2015; Hasija*et al*., 2015; Mahawar *et al*., 2020; Wang *et al*., 2020; Rousseaux *et al.,* 2020 and Li *et al.,* 2021).

Phenolic compounds especially flavonoids, which abound in citrus peel, have acquired practical importance in human health due to their free radicals scavenging (anti-oxidants activity) (Benalia *et al.*, 2015; Xu *et al.*, 2015; Boudkhili *et al*., 2015; Xiaoman *et al*., 2015; Jorge *et al*., 2016; Xi *et al*., 2017; Zema *et al*., 2018; Mohanan *et al*., 2018; Zhao *et al;* 2019; Singh *et al*., 2020; Redrouthu *et al.*, 2020; Rohman & Irnawati, 2020; Boudkhili *et al.*, 2021; Kim *et al.*, 2021 and Wei *et al*., 2022).

The present work aims at profiling the fatty acid and chemical compositions of Sweet Orange Peel Oil (SOPO), with a view to exploring its potential to be used as food/feed and source of raw materials for our industries.

MATERIALS AND METHODS

Materials

Sample collection and preparation

Fresh *Citrus sinensis*fruits (Sweet orange) were obtained from Umungasi Market in Aba North LGA of Abia State, Nigeria. The collected samples were washed and the fruits squeezed and the peel was cut into 1-2 mm^2 pieces,dried in hot air incubator at 50°C for 3 days, then the peel was furtherdried in an oven at a temperature of 65^0C for 2 hourstill a constant dry weight was obtained. Dried orange peel was thereafter ground into powder using a grinder. The powder was subsequently stored in dried glass bottle until analysis.

Extraction of Oil

This was done according to Uba & Muhammad (2019) and Schiavon (2016). The Soxhlet extraction was utilized to extract the oil from *Citrus sinensis* peel. The processed *Citrus sinensis* peel (50 g) were appropriately packed into the thimble of the Soxhlet extractor, and petroleum ether (300 mL) was put into the Soxhlet extractor's 500 mL round bottom flask. The oil from *Citrus sinensis* peel was extracted using the Soxhlet extraction system with petroleum ether (boiling point 60-80 °C). The extraction process took 6 hours. The extract was filtered using filter paper before being dried in a rotary evaporator at 40 °C. The recovered oils were stored in a vial at 4 °C until further analyses were carried out. The oil yield from *Citrus sinensis* peel was calculated using equation (1)

$$Oil\ Content(\%) \quad = \frac{W_3 - W_2}{W_1} \quad X \quad 100 \qquad ....(1)$$

Where, W_3 = weight of extraction cup + oil, W_2 = weight of extraction cup, W_1 = is weight of original sample.

Physical and Chemical parameters of the oil

The state, odour and colour of the oil was noted using visual inspection at room temperature (Magu *et al.*, 2017).

Determination of saponification value.A mixture 5 g of oil and 25 mL of 0.5 M ethanolic potassium hydroxide solution were heated under reflux for 60 minute in a conical flask. Thereafter, the saponified mixture was titrated with 0.5 M HCl after the addition of 2.0 mL of 0.05% phenolphthalein. The end point was achieved as the pink colour became colourless (Magu*et al*., 2017 ; Timilsena *et al*., 2017 andYusuf *et al*., 2021). The titration was also performed on the blank solution as the saponification value was calculated using equation (2)

$$\text{Saponification value}\ \left(\text{mg}\frac{\text{KOH}}{\text{g}}\right) \quad = \quad \frac{VHCl(mL) \quad X \quad MHCl \quad X}{Mass\ of\ the\ sample\ in\ g} \quad 56.1 \qquad ...(2)$$

Where, V is the volume of HCl used for titration and calculated by subtracting the total volume needed for titration of the blank solution and the sample (V HCl for blank - sample).

M is concentration in molarity of HCl.

Determination of iodine value. This was done using the Shimamoto *et al (2016).* Method, with slight modifications (reduction in sample weight and volume of distilled water). (0.5 g) aliquot of the oil sample was weighed and transferred to a 250 mL Erlenmeyer flask, and was dissolved in 15 mL 99% ethanol for 5 min with magnetic swirling vigorously. 20 mL of the 0.1 M ethanolic iodine solution after the stirring. After additional 5 minutes of stirring, 100 mL of cold distilled water was thereafter added as the stirring was slowed down.

The flask was kept under cover for 5 min and gently swirled with a magnetic bar, consciously without losing any Iodine. The solution was titrated with a standard 0.1 M sodium thiosulfate solution until a pale yellow colour was observed. After that, 3 mL of a 1% starch solution was added. The titration was continued until the blue color disappeared, leaving a milky solution. A blank was examined in the same way as the sample. The Iodine content was measured in milligrams per 100 grams of oil using equation (3)

$$Iodine\ value\ = \frac{VNa_2S_2O_3(ml)\ x\ MNa_2S_2O_3\ \ x\ 12.691}{Mas\ of\ the\ sample\ (g)} \qquad ... \qquad (3)$$

where, $VNa_2S_2O_3$ is a volume needed for titration, M is concentration of $Na_2S_2O_3$ in molarity and calculated by subtracting the total volume needed for titration, the blank solution and the sample (V $Na_2S_2O_3$ for blank - sample).

Determination of peroxide value. This was achieved by transferring 5 g of the oil sample into a 250 mL conical flask. This was dissolved in 30 mL of chloroform and a (2:3) ratio of glacial acetic acid. The mixture was shaken vigorously for exactly one minute. After that, 30 mL of distilled water was added. The solution was then titrated with sodium thiosulfate 0.1 M until it turned into a pale yellow solution. 1 mL 1% starch solution was added to this solution and titration continued until the blue color disappeared (Saeed & Shola, 2015; Magu *et al*., 2017; Shao *et al.*, 2019 and Paw *et al.*, 2020). The peroxide number was calculated using equation (4).

Peroxide number (meq/kg) = (V $Na_2S_2O_3$ (mL) × M $Na_2S_2O_3$ × 1000)/(Mass of the sample, g)…(4)

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$$Peroxide\ number\ \left(\frac{meq}{kg}\right) = \frac{VNa_2S_2O_3(ml)\ \ x\ MNa_2S_2O_3\ \ \ x\ 1000}{Mas\ of\ the\ sample\ (g)} \quad ...(4)$$

Determination of acid value. 5 g of oil sample was transferred into a 250 mL conical flask, then 100mL of neutralized ethanol (warmed to 60–65 ^{o}C) was added, along with 2 mL of 1% phenolphthalein, and titrated with 0.1 M ethanolic KOH till light pink color was observed.

Equation (5) was used to calculate the sample's acid value:

$$\text{Acid value}\ \left(\frac{\text{mg}}{\text{g}}\right) = \frac{VKOH(mL)\ \ X\ \ MKOH\ \ \ X\ \ 56.1}{Mass\ of\ the\ sample\ (g)} \quad ...\ \ (5)$$

Where, V = volume of KOH, and M (molarity) = concentration of KOH solution.

Polyunsaturated/ Saturated fatty acids (P/S Index).

Saturated fatty acids (SFAs) have negative impacts on blood lipoprotein profiles and coronary heart disease (CHD), according to WHO/FAO report. The ratio of polyunsaturated to saturated fatty acids (P/S index values) can be used to calculate the nutritional value of oils and any associated health hazards. Values higher than one P/S index for oils and fats are said to have a positive nutritional impact. Less lipid is deposited in the body when the P/S index is much higher than one (Gedefaw *et al.*,2021). The P/S index was measured by taking the ratio of total polyunsaturated fatty acid to the total saturated fatty acid.

Determination of density. Density of a substance speaks volume about its purity, concentration of components, and composition. The density (and concentration) of liquid products also greatly impact their quality, behavior, and use. This was measured using the density meter.

Viscosity.

The viscosity of the oil sample was determined using the Ostwald-U-tube viscometer (Bwade & Aliyu,2020). The viscometer was suspended at room-temperature, as the instrument was filled with oil to the mark at the top of the lower reservoir using a pipette as the tube was left above the dry mark. The oil was then moved into the other arm by means of the pressure on the respective arm of the tube, so that the meniscus was 1 cm above the mark at the top of the upper reservoir. The liquid was then allowed to flow freely through the tube, and the time it took for the meniscus to pass from the mark above the upper reservoir to the mark at the bottom of the upper reservoir was recorded. The viscosity of oil was calculated using equation (6)

$$Viscosity\ of\ the\ Oil\ = \frac{T - T_0}{T_0} \quad ...\ \ (6)$$

Where: T = flow time of the oil, T_0: flow time of distilled water.

Determination of fatty acid using gas chromatography-mass spectrometry (GC-MS)

Preparation of fatty acid methyl esters (FAME)

For fatty acids to be effectively measured bygas chromatography-mass spectrometer (GC-MS) analysis, it has to be converted into its derivative such as fatty acid methyl ester. According to Uba and Muhammad (2019), 1 g of the extracted oil and 2% methanolic KOH (6 mL) were added to a 50 mL round bottom flask. The condenser was connected to the round bottom flask, and the mixture was heated in a water bath for 1 hour at 60-70 °C with constant stirring. The reaction mixture was allowed to cool to room temperature. The solution was transferred to separatory funnel after adding 40 mL of n-hexane to it. The organic layer (upper layer) was separated and filtered through Whatman filter paper (110 mm) after drying over anhydrous sodium sulphate.

The esterified sample was concentrated as the solvent was removed through rotary evaporation to reduce the volume. However the solvent (n-hexane) was selectively removed from the esterified samples because it is more volatile than the ester (esterified sample). The methyl ester was kept at 4 ^{o}C until the GC-MS analysis. For methylation of the standards of free fatty acids, a similar procedure was used, and for GC-MS analysis, a trans esterified sample was produced at a concentration of 10mg/L.

Fatty Acid Profile

The fatty acid compositions were determined using an Agilent Technology 5977E MSD with an auto-sampler and an Agilent 7820A GC system (USA). The chromatographic separation was performed on a DB-1701 micro-column (30 m length, 0.25 mm internal diameter, 0.25 μm film thickness) at an 8 psi pressure and a 1 mL/min flow rate. At constantflow mode. Helium was used as the carrier gas and the total run time was 34.77 minutes. The injection of 1.0 μL of the sample into the inlet was done with anAgilent G4567A auto sampler and heated to 275 °C using a split less injection mode. The oven temperature was set to 100 °C for 2 min, with the beginning column temperature set to 100 °C. The temperature of the column was raised at a rate of 15 °C/min to 220 °C, then increased at a rate of 3 °C/min until it reached 240 °C. The temperatures of the transfer line and the ion source were 280 °C and 230 °C, respectively. The electron energy was set to 70 eV as ions with mass to charge ratio of 40 to 650 were collected. The identification of the fatty acids were done by comparing the retention times of a standard mixture to the retention times of the fatty acids, and by comparing with NIST spectral library

RESULTS AND DISCUSSION

Oil Yield

The oil content of *Citrus sinensis* peel (6.95%) as displayed in table 1indicates that sweet orange peel can actually serve as a source of oil in this regard. Khaled *et al. (*2018) had reported a lower oil yield of 3.62%. This could be attributed to the fact that higher temperature and yield of oil have a positive relation as shown in previous study, which stated that warmer climate increases oil content (Onukwube *et al*., 2024) .

However, Aneke *et al.* (2023) and Aydeniz-Guneser, (2020) reported higher oil yields of 53.33% and 51.80% respectively from sweet orange seeds, thus confirming the abundance of oil in the seed than the peel of sweet orange.

Physicochemical properties of *Citrus sinensis* peeloil.

Tables 1 and 2 show the physicochemical properties of the analyzed *Citrus sinensis*,Sweet orange peel oil (SOPO).

Acid Value

The acid value is employed as a quality control parameter in the analysis of fat and oil. The higher the acid number and free fatty acid content, the lower the quality of the oil, even as the acid number increases further with the age of an oil, because of transformation of triglycerides into glycerol and fatty acids with time (Onukwube *et al.,* 2024).

The acid value is usually a measure of how much potassium hydroxide (mg) is needed to neutralize the free acids in one gram of fat (Shimamoto *et al*., 2016). The result obtained for acid value was 16.32 mg KOH/g for (SOPO) as against 24mg/g ASTM value (Table 2). Thus the (SOPO) sample is said to be good quality oil.

An oil sample with a low acid value exudes fewer free acids and minimizes the risk of rancidity (Mulu *et al*., 2023).

Saponification value

As a quality control parameter for the characterization of fats and oil, the saponification value carries information on the average molecular weight of all fatty acids that are present in a sample. The higher the saponification number, the lower the molecular weight of all fatty acids, and vice versa (Yusuf *et al.*, 2021). The saponification value is actually a measure in milligrams of potassium hydroxide (KOH) necessary to saponify one gram of fat. The saponification value was found to be 805.04 mg KOH/g for the (SOPO) sample as against 122.74 – 158.42

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mg KOH/g for ASTM value (Table 2). This study's saponification value was higher than those reported for various seed oil samples, which were 210.345, 210.779, and 208.411 mg KOH/g for Soyabean, Peanut and cotton seed oils respectively (Table 5). The saponification value of (SOPO) was also higher than those of Pumpkin seed oils (175, 189, and 199 mg KOH/g) reported by Jiao *et al.* (2014); Kouba *et al. (*2016) and Bwade & Aliyu(2020) respectively. Furthermore, the saponification value in this study was higher than that for Moringa seed oil (171.9 mg KOH/g) and cashew seed oil (161 mg KOH/g) (Saeed & Shola, 2015 and Sicari *et al*., 2017). A high saponification value in oil generally indicates a better quality oil that is suitable for soap-making and other applications. It signifies that the oil has a higher proportion of fatty acids that are easily saponified, resulting in a harder, lathering soap (Joseph & Abdullahi, 2016).

Iodine Value

The iodine number as a quality control parameter is used for the quantification of the amount of unsaturated fatty acids present in fats and oils. The higher the degree of unsaturation, the more the number of iodine needed to react with the double bonds, resulting in a higher iodine value. Thus, Iodine value is a measure of the degree of unsaturation of fats and oil (Yusuf *et al.*, 2021). It is simply defined as the amount of iodine that can be added to 100 g of oil (Kukeera *et al*., 2015).

The iodine content of the (SOPO) was found to be 30.46g I_2/100 g as against 75.68 -77.85 I_2/100 g for ASTM value which implies a lower degree of unsaturation.

The present study shows that iodine value of (SOPO) was lower than those of other seed oil samples (136.740, 97.536, 101.112 g I_2/100 g) for Soyabean, Peanut and Cotton seed oils respectively. It was also found to be lower than that of pumpkin seed oil (Jiao *et al.*, 2014 and Yusuf *et al.*, 2021), which was 183 and 153 g I_2/100 g, respectively.

The degree of unsaturation of an oil sample is a function of the amount of iodine therein. Thus, the amount of iodine in an oil is directly proportional to the degree of unsaturation in that oil.

Oil is more likely to oxidize and its stability compromised at an elevated iodine level (Uba & Muhammad, 2019). SOPO with low iodine value from the study tends towards more stability.

Peroxide Value

The peroxide number talks about the amount of peroxide compounds present in the oil and so gives information about the quality and age of the edible oil. The lower the peroxidenumber,the better and/or fresher the oil. In other words, a higher peroxide value indicates a greater degree of oxidation, which can lead to rancidity and flavor deterioration in the oil.

The extent of rancidity of oils during storage is a function of the peroxide value and therefore the peroxide value can serve as an indicator for the determination of the stability of fats and oils (Magu *et al*., 2017) . The degree to which a fat has been oxidized is represented by milliequivalent (meq) of oxygen per 1000 g of fat, which is the peroxide value (Samuel *et al.*, 2017 and Xu *et al*., 2015).

Table 2 shows that the peroxide value of (SOPO) is 5.77meq O_2/kg, which is greater than those of other different seed oil samples (1.299, 2.398 and 3.396 meq O_2/kg) for soyabean, Peanut and Cottonseed oils respectively.

The value is equally greater than those of pumpkin seed oil peroxide values of 2.3, 2.5, and 2.5 meq O_2/kg published by Kukeera *et al.* (2015); Odoom *et al.*,(2015); and Bwade & Aliyu(2020), respectively. Thus, SOPO may not be suitable for use in the food industries as it is susceptible to deterioration in terms of rancidity.

Viscosity

The viscosity was found to be 0.17 Pa.s, which is lower than that previously reported by Ogbuagu & Okoye (2020). .

P/S Index

For edible oils, a P/S index of greater than one had been recommended by the World Health Organization (Tesfamicael & Ele, 2016). Higher values of P/S index are associated with oils that have more poly unsaturated compounds compared to saturated ones (Gedefaw *et al.*, 2021).

There exists a direct relationship between the higher value of the P/S index and the extent of oil solubility in the blood stream. Saturated fatty acids (SFAs) have the ability to tightly stick to cell membranes in sharp contrast to poly unsaturated fatty acids (PUFAs) (Onukwube *et al.*, 2024). Those SFAs with carbon number between 12 and 16 have the propensity to increase low-density lipoprotein (LDL) cholesterol (Gedefaw *et al*., 2021). The P/S index in this work was found to be 0.048

This is however below the acceptable range that was recommended by the WHO and thus SOPO can be said to have a negative nutritional impact. The low P/S index could be attributed to the high percentage of unidentified acids in the oil sample as various factors such as climatic conditions, variety, stage of maturity andexposure to sun can affect the FA compositions of fruit crops (Uba & Muhammad, 2019).

Fatty acid compositions of SOPO.

SOPO is made up of three groups of acids namely Monounsaturated Fatty Acid (MUFA) which is the most predominant, followed by Saturated Fatty Acid (SAFA) and lastly Polyunsaturated Fatty Acid (PUFA) with some unidentified as shown in Tables 3 and 4 as well Fig 2. The lipid profile in Table 3 showed that Elaidic acid was the most predominant fatty acid in the SOPO sample having the highest value (14.26%), followed by Heptadecanoic Acid (6.36%), while the least was Caprylic Acid(0.019%). Polyunsaturated fatty acids which are generally referred to as essential fatty acids are known to perform essential functions in the human body, including the development and maintenance of cell membranes and control of gene activity within the cell (Saini & Keum, 2018) . Thus eating a diet rich in polyunsaturated fatty acids and low in saturated fatty acids is encouraged as it is helpful to one's health.

However, since SOPO is predominantly composed of Monounsaturated fatty acids (MUFAs), it is therefore associated with negative health impact and its usage as major edible oil is discouraged.

Table 1: Physical Characterization of Sweet orange peel oil (SOPO) from Soxhlet Extraction (SE) method

Parameters	**SoxhletExtraction (SE)**
Yield (%)	6.95
Density g/cm^3	0.85
Melting Point (^{0}C)	-0.50
Viscosity (Pa.s)	0.17
Moisture (%)	65.00
pH	6.30
Colour	Brown
P/S Index	0.048

Table 2: Chemical Characterization of (SOPO)from Soxhlet Extraction (SE)

Properties	**Soxhlet Extraction (SE)**	**ASTM Standard value**
Acid Value(mg/g)	16.32	24

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Iodine Value(g I_2/100 g)	30.46	75.68 -77.85
Peroxide Value(meq/kg)	5.77	
SaponificationValue(mgKOH/g)	805.04	122.74 – 158.42
Flash point (0C)	85	95

Table 3. FAs analysis, with their chromatographic and mass spectrometry parameters.

SOPO SAMPLE						
RetTime [min]		**TypeArea**	**Amt/Area [pA*s]**	**Norm %**	**Grp**	**Name (FAME)**
3.875	VV T	96.92098	$1.93724e^{-1}$	0.500517	1	Butyric Acid Methyl Ester
4.166	VV T	2581.55811	$9.00248e^{-1}$	61.952824		ND
7.202	VV	123.98971	$2.95759e^{-1}$	0.977553	1	Caproic AcidMethyl Ester
11.896	BV	4.46307	$1.61805e^{-1}$	0.019250	1	Caprylic AcidMethyl Ester
13.804	V V	93.35983	$3.66571e^{-1}$	0.912295	1	Capric AcidMethyl Ester
15.498	V V	13.93383	$7.07380e^{-2}$	0.026275	1	Undecanoic AcidMethyl Ester
17.309	B V	4.84686	$2.24512e^{-1}$	0.029008	1	Lauric AcidMethyl Ester
18.579	V B	66.44740	1.70558e-^{1}	0.302112	1	Tridecanoic AcidMethyl Ester
18.884	B V	120.38950	7.57185e-^{2}	0.243001	1	Myristic AcidMethyl Ester
20.023	V V	72.19380	1.52330e-^{1}	0.293159	2	Myristoleic AcidMethyl Ester
20.200	VV	338.70093	6.93435e-^{2}	0.626093	1	Pentadecanoic AcidMethyl Ester
21.364	VV	52.19682	1.11761e-^{1}	0.155507	1	Palmitic AcidMethyl Ester
21.681	VV	591.22571	4.56118e-^{2}	0.718865	2	Palmitoleic AcidMethyl Ester
22.781	VV	1097.03992	2.17631e-^{1}	6.364438	1	Heptadecanoic AcidMethyl Ester
22.979	VV	1212.14551	1.52307e-^{1}	4.921436	1	Stearic AcidMethyl Ester
24.030	VV	1.0793344	4.95533e-^{2}	14.257507	2	Elaidic AcidMethyl Ester
24.112	VV	1133.15967	1.37798e-^{1}	4.162475	2	Linolelaidic AcidMethyl Ester
24.363	VV	210.30989	3.33347e-^{2}	0.186884	2	Oleic AcidMethyl Ester
27.804	VV	92.46461	$9.29057e^{-2}$	0.229000	3	Linoleic AcidMethyl Ester
28.305	BV	48.36203	$6.79066e^{-2}$	0.087545	1	Arachidic AcidMethyl Ester
30.635	VB	297.49188	$7.73864e^{-2}$	0.613701	3	Linolenic AcidMethyl Ester

34.037	VV	326.07114	$2.16612e^{-1}$	1.882835	1	Heneicosanoic AcidMethyl Ester
34.765	VV	258.67279	$7.79807e\text{-}^{2}$	0.537718	1	Behenic AcidMethyl Ester
40.024				- - -	2	Erucic AcidMethyl Ester
44.907				- - -	3	Arachidonic AcidMethyl Ester
46.788				- - -	1	Tridecanoic Acid Methyl Ester
Groups	**(1= SAFA, 2= MUFA, 3= PUFA, ND = NON DETECTED)**					

Table 4: Percentage composition of various groups of Fatty acids

	SAFA	**MUFA**	**PUFA**	**ND**
SOPO (%)	17.59	19.62	0.84	61.95

Table 5: Comparison of Physicochemical properties of various oil fractions

	SOPO	**Avocado pear**	**Soybean**	**Peanut**	**Cottonseed**
Moisture Content (%)	65.00	0.9	0.210	0.219	0.220
Peroxide Value(meq/kg)	5.77	2.100	1.299	2.398	3.396
Iodine Value(g I_2/100 g)	30.46	25.810	136.740	97.536	101.112
Saponification value(mg KOH/g)	805.04	230.010	210.345	210.779	208.411
Acid value(mg/g)	16.32	0.7000	0.420	0.630	0.561

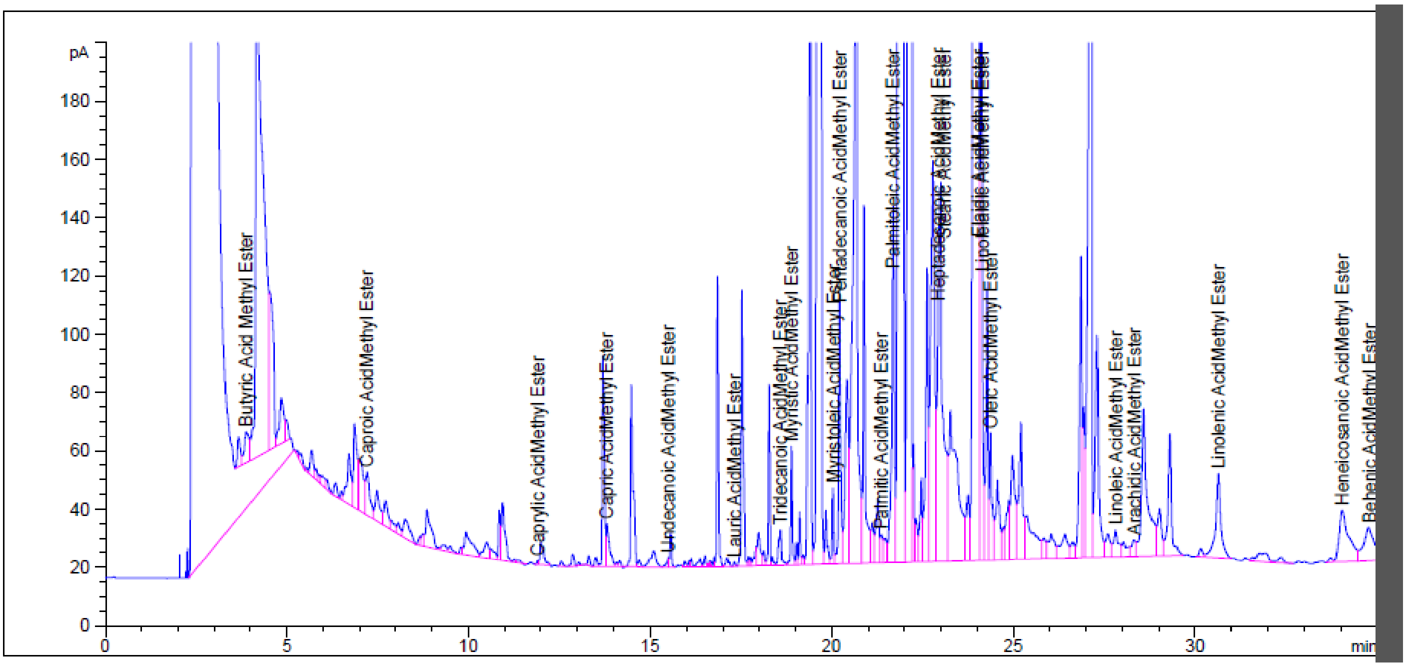

Fig.1: GC–MS total ion chromatogram of Fatty acid composition of SOPO sample from Soxhlet extraction (SE).

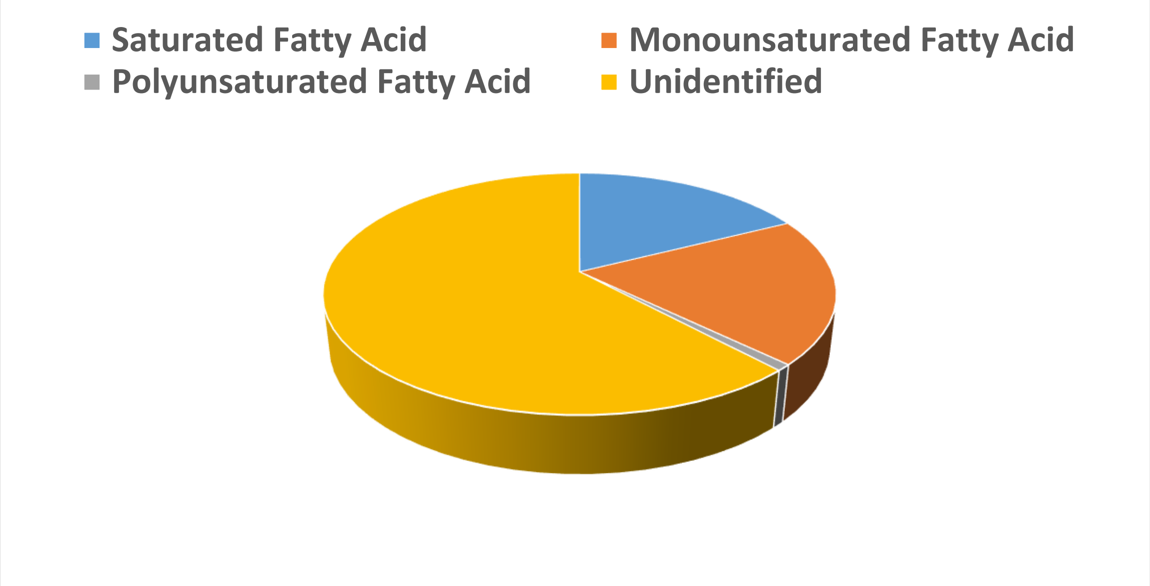

Fig. 2: Fatty Acid Group in SOPO Sample

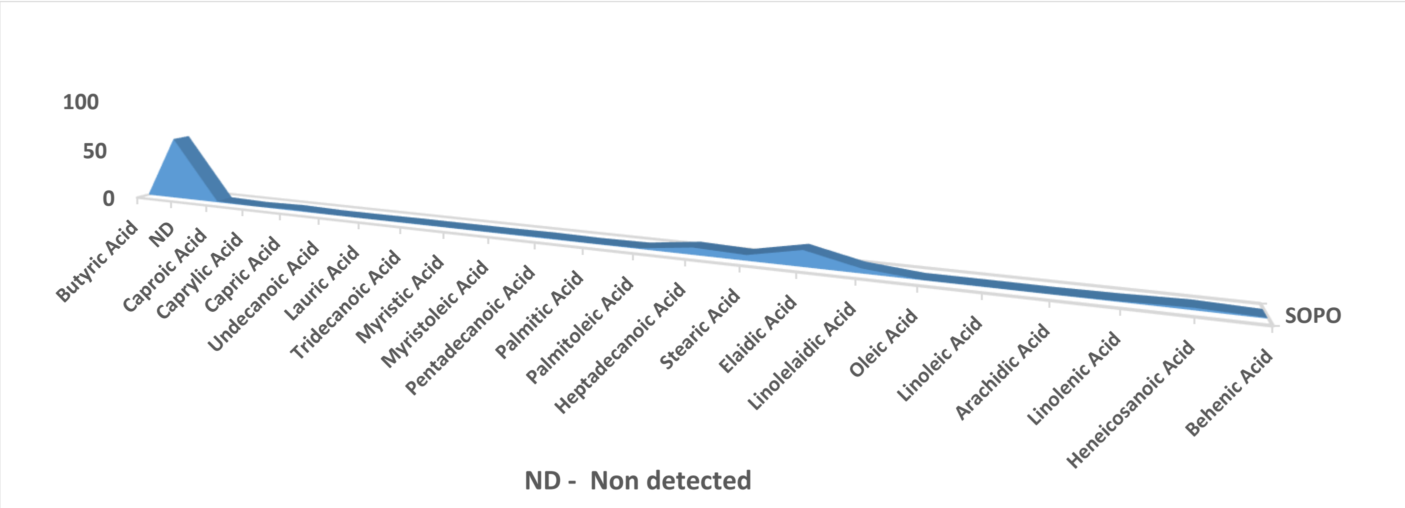

Fig. 3: FA profile of SOPO in percentage extracted through Soxhlet Method

CONCLUSION

The origin, weather conditions, svariety, and extraction methods all influence the compositions and quality of oils including sweet orange peel oil. The present study characterized *Citrus cinensis* (Sweet orange peel oil) as a mainly saturated oil, with a minute proportion of polyunsaturated fatty acids, with large percentage of the oil sample unidentified, which may include other bioactivecompounds, present in the unsaponifiable fraction, such as tocopherols and polyphenols.

With Elaidic acid, a trans-fatty acid, which is primarily known for its potential health implications, particularly its association with increased LDL cholesterol and reduced HDL cholesterol as the most predominant acid in SOPO, its application in human diet has been rendered insignificant.

Therefore,Fatty acids compositions of SOPO revealed that the oil sample is a potential source of valuable oil, suitable for numerous industrial applications such as soap, candles cosmetic industries (Khaled *et al*., 2018 and Suri, *et al*., 2022).

The use of these peels will greatly abate global warming by mitigating environmental pollution associated with their decomposition to a minimal and acceptable level.

Furthermore, extra research is recommended on *Citrus sinensis* peels, employing specific enzymes in the determination of their phytochemical compositions.

REFERENCES

1. Afifi, S.M., Kabbash, E.M.,Berger, R.G., Krings, U. & Esatbeyoglu,T. (2023). Comparative Untargeted Metabolic Profiling of Different Parts of Citrus sinensis Fruits via Liquid Chromatography–Mass Spectrometry Coupled with Multivariate Data Analyses to Unravel Authenticity. Foods. 12, 579 - 586.
2. Al Juhaimi, F., Ozcan, M.M., Uslu, N. & Ghafoor, K. (2018). The effect of drying temperatures on antioxidant activity, phenolic compounds, fatty acid composition and tocopherol contents in citrus seed and oils. Journal of Food Science and Technology, 55, 190–197.
3. Atolani, O., Adamu, N. & Oguntoye, O.S.(2020). Chemical characterization, antioxidant, cytotoxicity, anti-toxoplasma gondii and antimicrobial potentials of the Citrus sinensis seed oil for sustainable cosmeceutical production. Heliyon, 6, 33- 43.
4. Aydeniz Gunes € er, B. & Yilmaz, E. (2019). Comparing the effects of conventional and microwave roasting methods for bioactive composition and the sensory quality of cold-pressed orange seed oil. Journal of Food Science and Technology, 56, 634–642.
5. Benalia, M., Djeridane, A., Gourine, N., Nia, S. Ajandouz, E. & Yousfia, M. (2015). Fatty acid profile, tocopherols content and antioxidant activity of Algerian pumpkin seeds oil (Cucurbita pepo L.). Mediter. J. Nutr. Metab. 8, 9–25.
6. Boudkhili, M. (2015). Isolation and antioxidant activity of flavonoids from Coriaria myrtifolia methanolic extract. Chemistry of Natural Compounds. 51(1):141-142.
7. Boudkhili, M., Rharad, Z., Elguedarri, L and Misbahi, H. (2021). Phytochemical Analysis, Antioxidant Activity of Citrus sinensis l. and Punica granatum l. Peels Extracts on Stabilization of Linseed oil. Plant Cell Biotechnology and Molecular Biology 22(15&16):117-123.
8. Bwade, K.E. & Aliyu, B. (2020). Physicochemical properties of pumpkin seed oil relevant to bio-diesel production and other industrial applications. Int. J. Eng. Bus. Enterp. Appl. 4, 72–78.
9. Chew, S.C. & Ali, M.A. (2021). Recent advances in ultrasound technology applications of vegetable oil refining. Trends in Food Science & Technology, 116, 468–479.
10. Favela-Hernández, J.M.J., González-Santiago, O., Ramírez-Cabrera, M.A., Esquivel-Ferriño, P.C. & Camacho-Corona, M.D.R. (2016). Chemistry and pharmacology of Citrus sinensis. Molecules. PubMed 21, 247-252.
11. Gedefaw, Z., Menon, M., Ele, E. & Genet, S. (2021). Assessment of fatty acid content in various brands of imported oils in Ethiopia by gas chromatography. Int. J. Biochem. Mol. Biol. 6, 18–25.
12. Hasija, S.,Ibrahim, G & Wadia, A. (2015). Antimicrobial activity of Citrus sinensis (Orange), Citrus limetta (sweet lime) andCitrus limon (lemon) peel oil on selected food borne pathogens. International Journal of Life Sciences Research.3 (3): 35-39.
13. Jiao, J., Li, Z.G., Gai, Q.Y., Li, X.J., Wei, F.Y., Fu, Y.J. & Ma, W. (2014). Microwave assisted aqueous enzymatic extraction of oil from pumpkin seeds and evaluation of its physicochemical properties, fatty acid compositions and antioxidant activities. Food Chem. 147, 17–24.
14. Jorge, N., Da Silva, A.C. & Aranha, C.P.M. (2016). Antioxidant activity of oils extracted from orange (Citrus sinensis) seeds. Anais da Academia Brasileira de Ciencias , 88, 951–958.
15. Joseph, G. and Abdullahi, S. (2016) Physicochemical and Proximate Analysis of Extracts from Citrus sinensis of Dutsinma, Katsina State, Nigeria. Open Access Library Journal, 3: e2495.
16. Khaled, R., Souhail, N. & Amina, I. (2018). Orange peel fixed oil (citrus sinensis "valencia"), physiochemical properties, fatty acids profile, potential uses and the effect of environmental factors on it**.** Bulgarian Journal of Agricultural Science, 24 (1): 91–98.
17. Kim, D. S., Lee, S., Park, S.M., Yun, S.H., Gab, H. S., Kim, S.S. & Kim, H.J. (2021). Comparative metabolomics analysis of Citrus varieties. Foods. 10, 2826 - 2835.
18. Kouba, M., Mhemdi, H., Barba, F.J., Roohinejad, S., Greiner, R. & Vorobiev, E. (2016). Oil seed treatment by ultrasounds and microwaves to improve oil yield and quality. Food Res. Int. 85, 59–66.
19. Kukeera, T., Banadda, N., Tumutegyereize, P., Kiggundu, N., & Asuman, R. (2015). Extraction, quantification and characterization of oil from pumpkin seeds. Int. J. Agric. Biol. Eng. 8, 98–102.

20. Ledesma-Escobar, C., Priego-Capote, F. and Luque de Castro, M. (2015). Characterization of lemon (Citrus limon) polar extract by liquid chromatography–tandem mass spectrometry in high resolution mode. J. Mass Spectrom. 50, 1196–1205.
21. Li, Z., Jin, R., Yang, Z. et al. (2021). Comparative study on physicochemical, nutritional and enzymatic properties of two Satsuma mandarin (Citrus unshiu Marc.) varieties from different regions. Journal of Food Composition and Analysis, 95, 103- 114.
22. Magu, O.T., Sunday, A.E., Louis, H., Upla, U.P., Ukpanah, A.M. & Nyong, E.B. (2017). Physico-chemical properties and fatty acid composition of Lagneraria siceraria seed oil. Wnofns .10, 70–79.
23. Mahato, N., Sinha, M., Sharma, K., Koteswararao, R. & Cho, M.H. (2019). Modern extraction and purification techniques for obtaining high purity food-grade bioactive compounds and value added co-products from citrus wastes. Food, 8, 523 -530.
24. Mahawar, M.K., Jalgaonkar, K., Bibwe, B., Bhushan, B., Meena, V.S. & Sonkar, R.K. (2020). Post-harvest processing and valorization of Kinnow mandarin (citrus reticulate L.): a review. Journal of Food Science and Technology, 573, 799–815.
25. Mohanan, M., Nickerson, T. and Ghosh, S. (2018). Oxidative stability of flaxseed oil: Effect of hydrophilic, hydrophobic and intermediate polarity antioxidants. Food Chemistry. 66, 524-533.
26. Mulu, H., Estifanos, E. Y., Bhagwan, S. C. & Mesfin, R. (2023).Determination of fatty acids composition by gc-ms and physicochemical parameters of pumpkin (cucurbita maxima) seed oil cultivated in Ethiopia. Bull. Chem. Soc. Ethiop. 37(3): 565-577.
27. Odoom, W. & Edusei, V.O. (2015). Evaluation of saponification value, iodine value and insoluble impurities in coconut oils from Jomoro District of the Western region of Ghana. Asian Journal of Agriculture and Food Sciences, 3, 494–499.
28. Onukwube N. D.1, Amadi, E. G., Anarado I. L. & Ehiemere, A. C. (2024).Comparative Fatty Acid Profiling and Physiochemical Analysis of Persea Americana Mill. Oil Extracted from Two Different Methods. Intl. J. of research and inno. in applied sci. 9(5): 48 - 61
29. Ordoudi, S.A., Bakirtzi, C. & Tsimidou, M.Z. (2018). The potential of tree fruit stone and seed wastes in Greece as sources of bioactive ingredients. Recycling, 3, 9 - 21.
30. Paw, M., Begum, T., Gogoi, R., Pandey, S.K. & Lal, M.(2020). Chemical Composition of Citrus limon L. Burmf Peel Essential Oil from North East India. J. Essent. Oil Bear Plants. 23, 337–344.
31. Redrouthu, R., Sundramurthy, P.V. & Zergu, B. (2020). Extraction of essential oils from pumpkin seeds: RSM based process modeling, optimization and oil characterization. Int. J. Eng. Adv. Technol. 9, 2249–8958.
32. Rohman, A. & Irnawati, I. (2020). Pumpkin (Cucurbita maxima) seed oil: Chemical composition, antioxidant activities and its authentication analysis. Food Res. 4, 578–584.
33. Rousseaux, M.C., Cherbiy-Hoffmann, S.U., Hall, A.J. & Searles, P.S. (2020). Fatty acid composition of olive oil in response to fruit canopy position and artificial shading. Sci. Hortic., 271, 109477 - 109489.
34. Saeed, M.D. & Shola, E.A. (2015). Extraction and physicochemical properties of some edible seed oils sampled in kano metropolis, kano state. J. Pure App. Sci. 8, 239–244.
35. Saini, R.K. & Keum, Y.S. (2018). Omega-3 and omega-6 polyunsaturated fatty acids: Dietary sources, metabolism, and significance. Life. Sci. 203, 255–267.
36. Salonia, F.; Ciacciulli, A.; Poles, L.; Pappalardo, H.D.; La Malfa, S.; Licciardello, C. (2020).New plant breeding techniques in citrus for the improvement of important agronomic traits. A Review. Front Plant Sci. 11, 1234 - 1240.
37. Samuel, B.C., Barine, D.K. & JoY, E.E. (2017). Physicochemical properties and fatty acid profile of shea butter and fluted pumpkin seed oil, a suitable blend in bakery fat production. Int. J. Nut. Food Sci. 6, 122–128.
38. Schiavon, S., Pellattiero, E., Cecchinato, A., Tagliapietra, F., Dannenberger, D., Nuernberg, K., Nuernberg, G. & Bittante, G. (2016). The influence of different sample preparation procedures on the determination of fatty acid profiles of beef subcutaneous fat. liver and muscle by gas chromatography. J. Food Compos. Anal., 50, 10–18.
39. Shafiya, R., Rajkumari, K., Nadia, A.S.B., Fiza, A.N. and Gulzar, N. (2018). Citrus peel as a source of functional ingredient: A review. Journal of the Saudi Society of Agricultural Sciences.17, 351–358.
40. Shao, J., Liu, X., Zhou, H. & Liu, J. (2019). Analysis of extraction of grapefruit seed oil, fatty acids composition and physical and chemical properties. Fresenius Environmental Bulletin, 28, 4238–4246.

41. Shimamoto, G.G., Aricetti, A.J. & Tubino, M. (2016). A simple, fast, and green titrimetric method for the determination of the iodine value of vegetable oils without wijs solution (ICl). Food Anal. Methods. 9, 016-040.
42. Sicari, V., Messina, F. & Pellicano, T.M. (2017). Comparison of physicochemical characteristics and composition of bergamot oil seed extracted from three different cultivars. Emirates Journal of Food and Agriculture, 29, 470–475.
43. Singh, B., Singh, J.P., Kaur, A. & Singh, N. (2020). Phenolic composition, antioxidant potential and health benefits of citrus peel. Food Res. Int. 132, 109 - 114.
44. Suri, S., Singh, A. & Nema, P.K. (2022). Current applications of citrus fruit processing waste: A scientific outlook. Appl. Food Res. 2, 100050 - 100061.
45. Tesfamicael, F. & Ele, E.(2016). Assessment of fatty acids composition in commercially available and widely consumed edible oils in Ethiopia: their relevance and health implications. Sinet: Ethiop. J. Sci. 39, 21–33.
46. Timilsena, Y.P., Vongsvivut, J., Adhikari, R. & Adhikari, B. (2017). Physicochemical and thermal characteristics of Australian chia seed oil. Food Chem. 228, 394–402.
47. Uba, B. & Muhammad, C. (2019). Determination of fatty acid composition and physicochemical properties of Cucurbita maxima (pumpkin) seed oil cultivated in Northeast Nigeria. East Afr. Sch. Mult. Bull. 2, 231–234.
48. Wang, F., Huang, Y., Wu, W., Zhu, C., Zhang, R., Chen, J. & Zeng, J. (2020). Metabolomics analysis of the peels of different coloured citrus fruits (Citrus reticulata cv.'Shatangju') during the maturation period based on UHPLC-QQQ-MS. Molecules. 25, 396 - 412
49. Wei, L., Xiaoxue, M. & Zhiqin, Z. (2022). Analysis of physicochemical properties, fatty acid composition and antioxidant activity of seed oil extracted from 12 citrus materials. International Journal of Food Science and Technology. 57, 6480–6491.
50. Wilczynska, A. & Modrzewski, A.F. (2018). Chapter 15 – Fatty acids in human diet and their impact on cognitive and emotional functioning. The Role of Functional Food Security in Global Health. 261–270
51. Xi, W., Lu, J., Qun, J. & Jiao, B. (2017). Characterization of phenolic profile and antioxidant capacity of different fruit part from lemon (Citrus Limon Burm.) cultivars. Journal of Food Science and Technology, 545, 1108–1118.
52. Xiaoman, S., Liangxiao, Z., Peiwu, L., Baocheng, X., Fei, M., Qi, Z. and Wen, Z. (2015). Fatty acid profiles based adulteration detection for flaxseed oil by gas chromatography mass spectrometry. WT - Food Science and Technology. 430–436.
53. Xu, T.T., Li, J., Fan, W.Y., Zheng, W.T. & Deng, W.Z. (2015). Comparison of oxidative stability among edible oils under continuous frying conditions. Int. J. Food Prop. 18, 1478–1490.
54. Yusuf, J., Kantoma, D. & Mosunmola, O.R. (2021). Studies on Cucurbita maxima seed oil for its potentials as feedstock for biodiesel production in Nigeria. Nigerian Res. J. Chem. Sci. 9, 215–233.
55. Zayed, A., Badawy, M.T. & Farag, M.A. (2021). Valorization and extraction optimization of citrus seeds for food and functional food applications. Food Chemistry, 355, 129 - 138.
56. Zema, D.A., Calabro, P.S., Folino, A., Tamburino, V., Zappia, G. & Zimbone, S.M. (2018). Valorisation of citrus processing waste: a review. Waste Management, 80, 252–273.
57. Zhao, X.J., Xing, T.T., Li, Y.F. & Jiao, B.N. (2019). Analysis of phytochemical contributors to antioxidant capacity of the peel of Chinese mandarin and orange varieties. Int. J. Food Sci. Nutr. 70, 825–833.