



Identification of Nutritional Components of Different Varieties of Peanut Oil by Chromatographic Analysis

Long D. Thanh¹, Thuy N.T. Thu², Thuyet C. Thi², Long N. Tien^{1*}¹Institute of Biotechnology, Hue University, Hue city, Thua Thien Hue 49000, Viet Nam²University of Agriculture and Forestry, Hue University, Hue city, Thua Thien Hue 49000, Vietnam

ARTICLE INFO

Article history:

Received 21 August 2023

Revised 05 October 2023

Accepted 19 October 2023

Published online 01 November 2023

Copyright: © 2023 Thanh *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

ABSTRACT

Peanut (*Arachis hypogaea*) is an energy-rich food containing a substantial amount of fats, proteins, carbohydrates, vitamins, minerals, and phytochemicals. In this study, Gas Chromatography-Mass spectroscopy (GC-MS) and High-Performance Liquid Chromatography (HPLC) were applied to identify various phytochemical and nutritional ingredients in the oil of ten peanut varieties (TL03, LDH09, CNC, L14, TL04, SENNA, LDH01, L27, L19, and LACDOBG) collected from different provinces in Vietnam. The chromatographic analysis identified twenty-five (25) secondary metabolites in the oil of the peanut varieties. Vitamins E and B1 contents ranged from 0.195 to 0.230 mg/mL and 0.003 to 0.005 µg/mL, respectively. Whereas, Vitamins A and B3 were absent in the peanut oil. There were high levels of omega 6, omega 9, saturated fatty acids, monounsaturated fatty acids (MUFAs), and polyunsaturated fatty acids (PUFAs) in the oil of all the peanut varieties, while omega 3 was present in the oil of some peanut varieties but was not detected in the oil of CNC, L14, TL04, LACDOBG, L27, and SENNA. A total of 22 fatty acids were found in the oil of the peanut varieties, with the highest concentration being C18:1 n-7 - Oleic acid (43.13 to 50.71 g/100 g), followed by C18:2 n-6 - Linoleic acid (26.01 to 33.63 g/100 g) and C16:0 - Palmitic acid (11.24 to 12.19 g/100 g). The present study demonstrated that peanut oil is a rich source of energy, vitamins, and essential fatty acids that benefit human health.

Keywords: *Arachis hypogaea*, Fatty acids, Peanut oil, Vitamins.

Introduction

Cultivated peanuts (*Arachis hypogaea* L.) have an aneuploid chromosome set with the genome formula AABB (2n = 4 × = 40).¹ *Arachis* is a genus of 81 species that grows widely in South America.² Peanut is one of the world's most important sources of oil and cash crops.³ They are grown in more than 100 countries spanning many tropical and subtropical regions. In 2019, the total production of peanuts was about 48.8 million tons.⁴ Peanuts are a food source with nutritional functions and health benefits. *In vivo* and *in vitro* studies have shown that peanuts have important biological effects such as anti-cancer, anti-inflammatory, cardioprotective, and antibacterial effects, and also help in the regulation of intestinal flora.⁵ In the world, peanut is ranked 5th among the trees used in oil production with an output of more than 5 million tons in 2012.⁴ Peanut oil is one of the main oils in human diet. They are rich in proteins, lipids, more than 30 essential vitamins (E, K, and B complex) and many other nutrients and are part of a healthy balanced diet for human health in developed as well as developing countries.^{3, 6, 7} The many biological activities of peanuts suggest their potential for disease prevention and treatment.⁸ Peanut oil is mainly composed of triglycerides of eight fatty acids. About 80% of these fatty acids are oleic acid (monounsaturated, C18:1) or linoleic acid (polyunsaturated, C18:2).⁹

Previous peanut nutritive assessments have substantially concentrated on allergens and/or peanut-specific phytonutrients.¹⁰ All foods are chemical composites, which may be defined as macro or micronutrients like proteins, carbohydrates, fats, vitamins, minerals, and phytonutrients. The present study aims to carryout a nutritional evaluation of different varieties of peanut oil with a view to assessing their health benefits.

Materials and Methods

Plant material

Ten peanut varieties from different provinces/cities in Vietnam (Table 1) were collected and grown in the net house at the Institute of Biotechnology, Hue University. After 4 months, peanut seeds were harvested and washed with tap water. The peanuts were then dried in the oven at 50°C (MOV- 212) until the moisture content was below 10% and used to extract oil for further analysis.

Extraction of oil

The oil content in the peanut seeds was determined based on standard procedure (TCVN 8951-1:2011; ISO 734-1:2006).¹¹ Finely ground peanut seeds (10 g) was accurately weighed using a precision balance (KERN PCB 250-3, China), and dried to less than 10% moisture content. The ground peanut seeds was wrapped in Whatman paper (0.45 mm) and transferred into an extraction tube with a globe flask containing 5.1 L of n-Hexane. The parts of the extraction system were connected and placed on the electric stove. Heating was carried out under reflux and extraction was achieved at a rate of 3 drops per second. After extraction for 6 hours, the extract in the globe flask was removed and allowed to cool in a desiccator for at least 1 hour to ambient temperature and then weigh to the nearest 1 mg so that the difference between two successive weighings does not exceed 10 mg. The final weight was recorded and the percentage oil content (w) was calculated from the formula as shown in equation 1 below.

$$w = \frac{m_1}{m_0} \times 100 \quad (1)$$

*Corresponding author. E mail: ntienlong@hueuni.edu.vn
Tel: +840914415198

Citation: Thanh LD, Thu TNT, Thi TC, Tien LN. Identification of Nutritional Components of Different Varieties of Peanut Oil by Chromatographic Analysis. Trop J Nat Prod Res. 2023; 7(10):4894-4901. <http://www.doi.org/10.26538/tjnpr/v7i10.26>.

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Where; m_0 is the mass in grams (g) of the ground peanut seeds, m_1 is the mass in grams (g) of the dried extract.

Determination of total protein content

The total protein content of the peanuts seeds was determined according to the Bradford method.¹²

Finely ground peanuts seeds (100 mg) was accurately weighed into a 10 mL test tube and suspend in 1 mL Arakawa extraction buffer (1 mM phenylmethylsulfonyl fluoride; 200 mM Tris/HCl, pH 8; 100 mM NaCl; 400 mM sucrose; 10 mM EDTA; 14 mM 2-mercaptoethanol; 0.05% Tween-20). The mixture was vortexed (Vortex 3 Genius, IKA) for 20 minutes and then left overnight. The supernatant was aspirated into a new tube and centrifuged (Centrifuge 5417 R, Eppendorf) at 15000 rpm for 20 min at 4°C. The total protein content was determined using the Bradford Reagents with the aid of a spectrophotometer (SmartSpec™Plus, BIO-RAD, USA) at wavelength of 595 nm. The protein content was calculated based on bovine serum albumin (BSA-Promega) standard calibration curve.

Equation 2 below show the formula for calculating the protein content in the sample.

$$X = \frac{C \times V \times a}{1000} \quad (2)$$

Where; X = Protein content in 100 g of sample (g/100g); C = Protein content according to the standard curve (mg/mL); V = sample solution volume (mL); a = solution dilution factor.

Determination of total sugar content

The total sugar content in the peanut seeds was determined using the method described by AOAC 923.09 (AOAC 923.09)¹³ and Lindsay (1973).¹⁴

Accurately weighed peanut powder (1 g) was placed into a 250 mL conical flask. Forty-five millilitres (45 mL) of distilled water and 5 mL of hydrochloric acid (HCl) were added, and the solution was boiled on a water bath for 3 h, then allowed to cool under cold running water. To the solution was added 3-5 drops of phenolphthalein, and then neutralized with NaOH (50%) until the colour of the solution turns pink. The entire solution was transferred into a 100 mL volumetric flask, 15 mL of potassium ferrocyanide [$K_4Fe(CN)_6$] (15%) and 15 mL of zinc acetate [$Zn(O_2CCH_3)_2$] (30%) were added and mixed thoroughly. The solution was made up to the 100 mL mark with distilled water, and then filtered on filter paper to collect sugar solution. Glucose at concentrations of 0.2 - 1 mg/mL was used to prepare a calibration curve. The sugar solution (test sample or standard sample) (250 μ L) was reacted with 250 μ L DNS and 50 μ L of saturated sodium carbonate (Na_2CO_3). The reaction mixture was boiled in a water bath for 5 minutes, and then allowed to cool to room temperature. A blank was prepared using distilled water in place of the test sample. The Optical

Density of the resulting solution was measured at 570 nm (OD_{570nm}) on a spectrophotometer (SmartSpec™Plus, BIO-RAD, USA). The total sugar content was calculated according to the formula in equation 3 below:

$$X \text{ (g/100g)} = \frac{y \times n \times V}{V_1 \times m \times 1000} \quad (3)$$

Where; y = Sugar content according to the standard curve (mg/mL); V = Sugar solution volume (mL); m = weight of sample (g); n: solution dilution; V_1 : Reaction sample volume (μ L).

GC-MS/MS Analysis

GC-MS/MS analysis of the peanut oil was carried out according to the method described by QuOil (2019).¹⁵ Briefly, 2 g \pm 0.02 g of sample homogenate was added 10 mL of acetonitrile, and placed on a mechanical shaker for 45 minutes to allow for oil extraction. To the extract was added QuEChERS buffer-partitioning salts mixture and shaken for 1-2 minutes. The extract was centrifuged at >3000 \times g and then subjected to dispersive solid phase extraction (SPE) cleanup with C18/PSA/MgSO₄ (25/25/150 mg/mL extract) to remove lipids and fatty acids. The extract was filled into GC-vials for GC-MS/MS analysis on the TG-5MS (size \times I.D. 30 m \times 0.25 mm, df 0.25 μ m) column and the GC-MS/MS thermo TSQ 9610 system.

The determination of vitamins contents

Vitamin A (Retinol), vitamin E (α -tocopherol), Vitamin B1 (Thiamine), and Vitamin B3 (Niacin) contents were analyzed by HPLC on HPLC-FLD-DAD thermo system with UV or FLD detector according to standard methods; AOAC 992.04 (Vitamin A), AOAC 992.03 (Vitamin E), EN 15652 (Vitamin B3) and EN 14122:2003 (Vitamin B1). Peanut oil (5 mL) was used for each analysis. The extract (10 μ L) after passing through the 0.45 μ m filter was loaded into the column (C18-MS 2.1 \times 150 mm, 5 μ m).

Vitamins A and E: The Sample was dissolved in water, followed by addition of pyrogallol (antioxidant) and potassium hydroxide. Then the sample was digested by shaking in water bath (Model: 3032, GFL) at 70°C for 25 min. The digested sample solution was extracted with hexane:methylene chloride (3:1), the solvent was evaporated to obtain a dry residue. The residue was then reconstituted in the mobile phase [Hexane:isopropyl alcohol (100:0.25) for vitamin A analysis, and Hexane:isopropyl alcohol (99.92:0.08) for vitamin E analysis].

Vitamins B1 and B3: Homogenized test sample was digested in hydrochloric acid or sulfuric acid solution (pH < 2.0) for 30 min at 121°C. Extract solution was adjusted to optimal pH with sodium acetate buffer, after which the enzyme - takadiastase was added, and then incubated at 37°C for 2 h.

Table 1: List of peanut varieties used in the study

No.	Variety	Sampling location	GPS
1	TL03	Institute of Biotechnology, Hue University	16.49507376314785, 107.60579659125655
2	LDH09	Agricultural Science Institute for Southern Coastal Central of Vietnam	13.770231991283724, 109.19083471824304
3	CNC	Agricultural Science Institute for Southern Coastal Central of Vietnam	13.770231991283724, 109.19083471824304
4	L14	Thua Thien Hue	16.52902808085653, 107.46565463851884
5	TL04	Institute of Biotechnology, Hue University	16.49507376314785, 107.60579659125655
6	SENNA	Nghe An	18.715240751877314, 105.64247729807703
7	LDH01	Agricultural Science Institute for Southern Coastal Central of Vietnam	13.770231991283724, 109.19083471824304
8	L27	Gia Lai	13.979714296304923, 108.00422333848425
9	L19	Quang Binh	17.486561209262288, 106.6022187827131
10	LACDOBG	Bac Giang Agriculture and Forestry University	21.284028021456862, 106.09247769627501

The sample extract was then diluted with distilled water prior to HPLC analysis (for Vitamin B1) and subjected to post-column derivatization with UV irradiation, followed by fluorimetric detection to determine Niacin (Vitamin B3) content.

Determination of fatty acids

Analysis of fatty acid composition in the peanut oil was done by gas chromatography-mass spectrometry (GC-MS) according to standard procedure (TCVN 9675-2:2013 - ISO 12966-2:2011).¹⁶ The sample was saponified and converted to a methyl ester by reaction with BF₃-MeOH (boron trifluoride-methanol) according to the method previously described (COI/T.20/Doc. No. 24:2001 (COI/T.20/Doc., 2001)).¹⁷ The fatty acid ester composition (FAME) was analyzed on a gas chromatograph with flame ionization detector (GC-FID), mobile phase: Helium gas, high polarity column HP-88, 100 m x 0.25 mm ID, 0.2 μm (JW112-88A7) (GC Agilent system with FID (20:46) detector, Case Laboratory Company, Ho Chi Minh City, Vietnam). Fatty acids were identified by comparing their retention times and mass spectra data with that found in a standard library.

Statistical Analysis

Data was subjected to one-way analysis of variance (ANOVA), and differences between means were compared using Duncan's test. The values were regarded as statistically significant at $p < 0.05$. All statistical analyses were performed using SPSS software (Version 20).

Results and Discussion

Peanut (*Arachis hypogaea*) also known as groundnut belongs to the legume family and is an important crop in Vietnam and around the world. Asia is the major producer of peanuts, accounting for 60% of the world's production.¹⁸ There are many different varieties of peanuts, however, four main varieties (Runner, Virginia, Spanish, and Valencia) have gained market acceptance due to their taste, oil content, size, shape, and disease resistance.¹⁹ Peanuts are rich in protein, vitamins, oil, fiber, and carbohydrates and are consumed worldwide due to their availability and affordable price.^{6,7} Peanut oil is mainly composed of triglycerides of eight fatty acids. About 80% of these fatty acids are oleic acid (monounsaturated, C18:1) or linoleic acid (polyunsaturated, C18:2).⁹

In Vietnam, the majority of this crop is processed into cooking oil, peanut butter, salted peanuts, and snack bars for consumption. The by-product of oil extraction is a powder rich in protein, fiber, antioxidants, vitamins, and minerals, and is utilized as animal feed or further processed for human consumption.²⁰ Other beneficial effects of using peanuts with limited evidence are reduced rates of obesity, cancer, hypertension, and inflammation.^{5,21}

In addition to macronutrients, peanuts are an excellent source of several important vitamins including B vitamins and vitamin E.

Nutritional composition of peanut seeds

There were differences in the nutritional composition of the seeds of peanut varieties collected in different localities. The most significant variation occur in the oil content which ranged from 45.731 to 63.557%. The peanut variety L14 collected in Thua Thien Hue showed the highest oil content (63.557%). The total protein and total sugar contents in the different peanut varieties ranged from 24.660 to 26.632 g/100 g sample (total protein), and from 4.403 to 5.002 g/100 g sample (sugar total). Of all the studied varieties, the Peanut variety with the code - CNC collected at the Agricultural Science Institute for Southern Coastal Central of Vietnam had the highest total protein and total sugar contents (Table 2).

Composition of peanut oil

Gas Chromatography-Mass spectrometric (GC-MS) analysis of the different peanut oil identified a total of twenty-five secondary metabolites (Figure 1). The chemical names, retention times (RT), molecular formulae, molecular weights (MW), and peak areas (%) of the compounds are presented in Table 3.

In the present study, secondary metabolites with a percentage peak area of less than 5% were considered insignificant. The secondary metabolites with significant percentage peak area were n-Hexadecanoic acid (ranging from 9.23 to 16.59%), cis-13-Octadecenoic acid (ranging from 53.62 to 63.85%), and 1,2-Cyclohexanedicarboxylic acid, bis(2-ethylhexyl) ester (ranging from 2.79 to 13.64%). Two compounds n-Hexadecanoic acid (ranging from 9.23 to 16.59%) and cis-13-Octadecenoic acid (ranging from 53.62 to 63.85%), were the most abundant in the oil extracted from the peanut variety - CNC with a proportion of 16.59% and 63.85%, respectively, while Cyclohexanedicarboxylic acid, bis(2-ethylhexyl) ester in the oil extracted from this peanut variety had the smallest percentage peak area compared to other peanut varieties used in this study. The composition of the peanut oil varies among the different peanut varieties. The peanut variety TL03 gave the highest number of compounds (21 out of 25 compounds), followed by peanut variety LDH09 (19 out of 25 compounds), while the peanut varieties TL04, LACDOBG, L27, and SENNA had the lowest compound composition (15 out of 25 compounds) (Table 3 and Figure 1).

Vitamins composition of Peanut oil

The results of the analysis of the composition of four different vitamins in peanut oil extracted from different peanut varieties are presented in Table 4. The results showed that only 2 out of 4 vitamins are present in the oil of the studied peanut varieties. Vitamin E accounted for the highest content, ranging from 0.195 to 0.230 mg/mL, in which the oil extracted from peanut variety L14 collected in Thua Thien Hue province, Vietnam gave the highest vitamin E content (0.230 mg/mL).

Table 2: Some nutritional components of different varieties of peanut seeds

No.	Sample	Oil content (%)	Protein total (g/100 g)	Sugar total (g/100 g)
1	TL03	57.344 ^{ab} ± 1.731	25.820 ^b ± 0.556	4.602 ± 1.032
2	LDH09	55.874 ^b ± 0.633	25.704 ^b ± 1.001	4.669 ^b ± 1.100
3	CNC	52.929 ^b ± 1.190	26.632 ^a ± 0.620	5.002 ^a ± 0.302
4	L14	63.557 ^a ± 0.677	26.012 ^a ± 1.301	4.592 ^{bc} ± 0.655
5	L19	50.196 ^{bc} ± 2.147	25.507 ^b ± 0.664	4.796 ^b ± 0.027
6	TL04	52.085 ^b ± 0.899	25.902 ^b ± 0.872	4.705 ^b ± 1.226
7	LDH01	48.267 ^{cd} ± 2.794	24.660 ^c ± 2.061	4.801 ^b ± 0.730
8	LACDOBG	49.006 ^c ± 0.912	25.011 ^b ± 0.822	4.403 ^c ± 0.856
9	L27	45.836 ^d ± 1.465	25.206 ^b ± 0.368	4.526 ^{bc} ± 0.902
10	SENNA	45.731 ^d ± 1.050	25.100 ^b ± 1.201	4.720 ^b ± 0.056

Note. Different letters in the same column indicate a statistically significant difference of the sample mean at $p < 0.05$ (Duncan's test).

Table 3: Chemical composition of different varieties of peanut oil

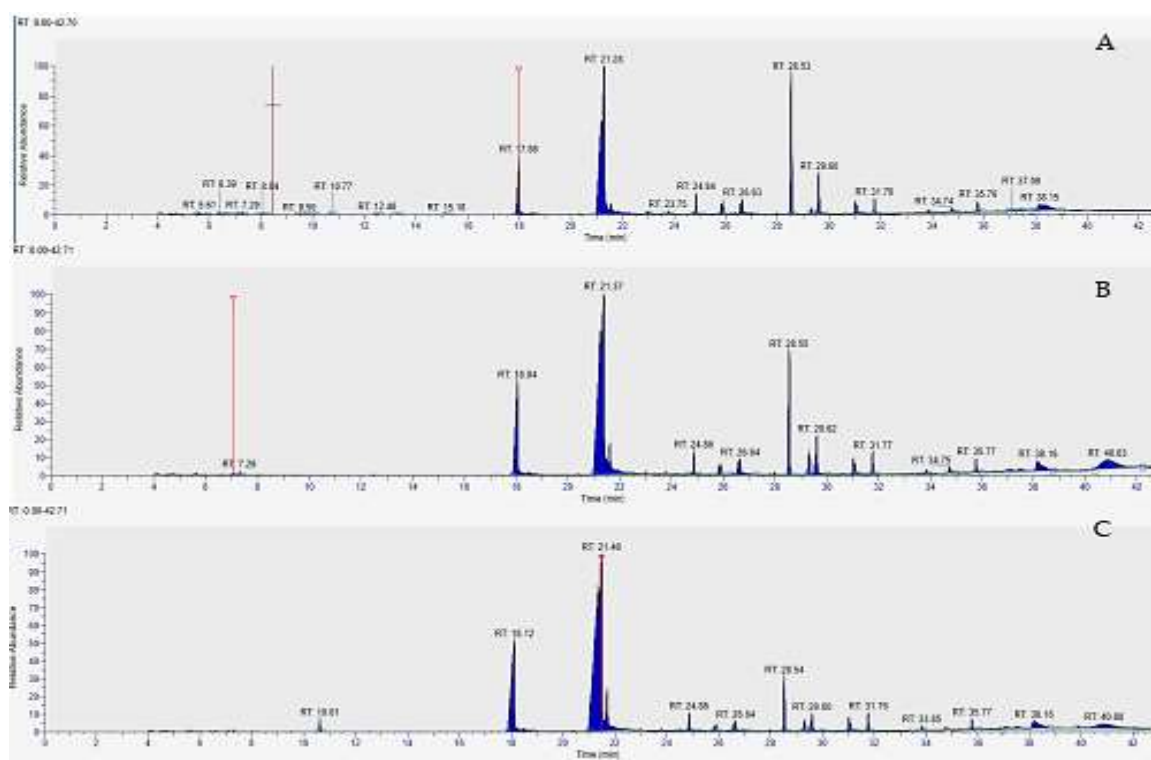
RT (Min)	MW (g/mol)	Formula	Compounds	% Peak Area										
				TL03	LDH09	CNC	L14	L19	TL04	LDH01	LACDOB G	L27	SENNA	
5.61	282.48	C ₁₃ H ₂₂ O ₃ Si ₂	2,5-Dihydroxybenzaldehyde, 2TMS derivative	0.33	-	-	-	-	-	-	-	-	-	-
6.39	354.4	C ₁₆ H ₂₂ N ₂ O ₅ S	Benzofuran, 2,3-dihydro-2,2-dimethyl-7-(N-(N-methyl-N-ethoxycarbonylaminothio)-N-methylcarbamoyloxy)-	0.08	-	-	-	-	-	-	-	-	-	-
7.29	152.23	C ₁₀ H ₁₆ O	2,4-Decadienal	0.16	0.12	0.03	0.12	0.03	0.20	0.15	0.03	0.06	0.12	
8.04	458.6	C ₂₅ H ₃₄ O ₆ Si	2'-(Tert.-butyldimethylsilyloxy)-2,4,4',6'-tetramethoxychalcone	0.02	-	-	-	-	-	-	-	-	-	-
9.50	579.2	C ₁₆ H ₅₀ O ₇ Si ₈	Octasiloxane, 1,1,3,3,5,5,7,7,9,9,11,11,13,13,15,15-hexadecamethyl-	0.02	-	-	-	-	-	-	-	-	-	-
10.77	523.94	C ₃₁ H ₅₆ O ₃ Si ₂	19-Hydroxytestosterone, O,O'-bis(tert.-butyldimethylsilyl)-	0.01	0.01	0.01	-	-	-	-	-	-	-	-
12.48	240.38	C ₁₅ H ₂₈ O ₂	Cyclohexanecarboxylic acid, 2-ethylhexyl ester	0.05	0.05	-	-	0.04	-	0.05	-	-	-	-
17.22	307.27	C ₁₄ H ₂₇ BrO ₂	2-Bromotetradecanoic acid	-	0.05	-	-	-	-	-	-	-	-	-
18.14	256.42	C ₁₆ H ₃₂ O ₂	n-Hexadecanoic acid	9.23	11.24	16.59	12.44	11.25	13.56	11.28	16.39	12.54	11.15	
20.30	294.5	C ₁₉ H ₃₄ O ₂	7,10-Octadecadienoic acid, methyl ester	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04	
20.4	296.5	C ₁₉ H ₃₆ O ₂	Methyl (9E)-9-octadecenoate is a fatty acid methyl ester	-	0.06	-	-	-	-	-	-	-	-	-
21.30	282.5	C ₁₈ H ₃₄ O ₂	cis-13-Octadecenoic acid	59.04	59.51	63.85	54.64	53.62	59.61	56.41	60.85	58.24	55.43	
23.76	312.5	C ₁₉ H ₃₆ O ₃	Glycidyl palmitate	0.23	0.23	0.23	0.23	0.23	0.23	0.23	0.23	0.23	0.23	
24.84	281.5	C ₁₈ H ₃₅ NO	9-Octadecenamide, (Z)-	2.06	2.18	1.27	1.42	1.25	1.87	1.84	1.34	1.76	1.54	
25.88	356.54	C ₂₁ H ₄₀ O ₄	9-Octadecenoic acid (Z)-,2,3-dihydroxypropyl ester	1.90	1.54	0.53	0.93	1.30	1.24	1.09	1.72	1.64	1.66	
26.63	338.5	C ₂₁ H ₃₈ O ₃	9-Octadecenoic acid (Z)-, oxiranylmethyl ester	3.37	3.13	1.27	2.24	3.02	2.72	2.85	2.76	3.11	2.94	
27.95	310.5	C ₂₀ H ₃₈ O ₂	Meadowlactone	0.15	0.17	0.18	0.16	0.15	0.15	0.17	0.18	0.16	0.15	
28.53	396.60	C ₂₄ H ₄₄ O ₄	1,2-Cyclohexanedicarboxylic acid, bis(2-ethylhexyl)ester	13.64	10.37	2.79	6.87	8.02	7.46	13.64	6.37	4.79	6.87	
31.02	338.57	C ₂₂ H ₄₂ O ₂	Erucic acid	2.11	2.22	1.22	1.53	2.03	1.94	1.76	2.02	1.24	1.57	
31.76	410.7	C ₃₀ H ₅₀	Squalene	1.27	1.56	0.94	1.15	1.37	1.47	1.26	1.48	1.04	1.53	
34.74	416.7	C ₂₈ H ₄₈ O ₂	γ-Tocopherol	0.75	0.39	0.41	0.56	0.73	0.58	0.72	0.49	0.41	0.66	
35.76	430.7	C ₂₉ H ₅₀ O ₂	α-Tocopherol	1.12	1.06	0.64	0.85	0.83	1.07	1.06	0.64	0.85	0.92	
38.14	414.7	C ₂₉ H ₅₀ O	γ-Sitosterol	3.63	3.63	3.63	4.3	4.01	5.55	3.52	3.70	4.3	.368	
40.83	428.6	C ₂₇ H ₄₀ O ₄	3-Hydroxyspirost-8-en-11-one	-	-	3.96	7.04	3.96	-	-	-	-	-	

Note: MW: Molecular Weight; RT: retention time; -: Not detected

Table 4: Composition of vitamins in different varieties of peanut oil

No.	Sample	Vitamin E (α -tocopherol) (mg/mL)	Vitamin A (mg/mL)	Vitamin B1 (thiamine) (μ g/mL)	Vitamin B3 (niacin) (μ g/mL)
1	TL03	0.204	-	0.003	-
2	LDH09	0.214	-	0.005	-
3	CNC	0.195	-	0.004	-
4	L14	0.230	-	0.004	-
5	L19	0.226	-	0.003	-
6	TL04	0.203	-	0.003	-
7	LDH01	0.220	-	0.004	-
8	LACDOBG	0.201	-	0.003	-
9	L27	0.197	-	0.004	-
10	SENNA	0.202	-	0.003	-

Note. -: Not detected

**Figure 1:** GC-MS Chromatogram of peanut oil. A: peanut variety TL03, B: peanut variety CNC, and C: peanut variety L14

The content of vitamin B1 in the oil obtained from the peanut varieties did not differ significantly, ranging from 0.003 to 0.005 μ g/mL. Oil of peanut variety LDH09 collected at the Agricultural Science Institute for Southern Coastal Central of Vietnam showed the highest vitamin B1 content (0.005 μ g/mL). Vitamins A and B3 were not detected in the oil obtained from the peanut varieties used in the study (Table 4).

Vitamins are essential micronutrients needed by the body to perform many functions such as strengthening the immune system, energy production and food digestion. Among the 14 vitamins, the human body can synthesize vitamin D3 and niacin. The remaining vitamins must be obtained from the diet for the normal biochemical and physiological functioning of the body.²² Vitamin overload occurs with an overdose of fat-soluble vitamins because they can be stored in the liver and body's fatty tissues unlike water-soluble vitamins.²³

Thiamine (Vitamin B1) is a part of enzymes necessary for energy metabolism, and it is important for brain and nerve function.²⁴ Niacin (Vitamin B3) and thiamine are responsible for the normal functioning of the digestive system, skin, as well as for the functioning of the heart, muscles and nervous system.²⁵ Vitamin A promotes good vision,

vitamin E functions as an antioxidant to protect cell membranes.²⁶ Vitamin E is not a single compound but a group of eight compounds that act as antioxidants maintaining the stability of cell membranes against oxidative stress.²⁷ The foods with the highest vitamin E content are oilseeds. Peanuts have been documented by many studies to be an excellent source of thiamine, niacin, and vitamin E. The literature on B vitamins in peanuts is very limited because the analysis of B vitamins in oilseeds is difficult in terms of technique: the process of extracting and purifying vitamin B from cereal grains and oilseeds is time-consuming, and different types of B vitamins need to be analyzed by different methods.²⁸ The main challenge is the loss of vitamins at the end of the extraction due to exposure to light and air. Research by Nadathur *et al.* showed that peanuts in the United States are rich in niacin but contain moderate amounts of thiamine. Peanuts grown in other locations have B vitamin compositions similar to peanuts grown in the United States: 0.6 mg/100 g thiamine, and 12.1 mg/100 g niacin.²⁹ It has been reported that people who consume peanuts have adequate amounts of B vitamins such as niacin, and thiamine.³⁰ In this study, we analyzed some vitamins in oil extracted from different peanut varieties.

The result showed that vitamin E content accounted for the highest and the content of vitamin B1 accounted for the lowest. The variation in the content of these vitamins among the peanut varieties were considered insignificant.

Fatty acid composition

The fatty acid composition in peanut oil is shown in Table 5. The results showed that the fatty acid composition in the oil obtained from the peanut varieties are different, with the percentage content ranging from 98.14 to 100%. In all the peanut varieties, the oil contained high levels of Omega 6, Omega 9, Saturated fatty acids, Monounsaturated fatty acids (MUFA), and Polyunsaturated fatty acids (PUFA), ranging from 26.01 to 33.19 g/100 g sample, 44.03 to 55.59 g/100 g sample, 19.53 to 21.67 g/100 g sample, 44.12 to 50.71 g/100 g, and 26.66 to 33.92 g/100 g sample, respectively. However, Omega 3 and trans fats were present only in the oil of some peanut varieties such as TL03, LDH09, L19, and LDH01, but not found in the oil of the remaining peanut varieties including CNC, L14, TL04, LACDOBG, L27, and SENNA (Table 5 and Figure 2).

As a result of this difference, some fatty acids such as C14:0 (Myristic acid), C16:1 n-7 (Palmitoleic acid), C16:1 (Hexadecenoic acid and other isomers), C17:0 (Margaric acid), C17:1

n-8 (9-cis-heptadecenoic acid), C18:1 tr6-8 (Trans-(6) -8)-octadecenoic acid), C18:1 tr9 (Elaidic acid), C18:1 tr10 (Trans-10-octadecenoic acid), C18:1 c11 (Cis-vaccenic acid), C18:2 n-6 trans (Linolelaidic acid), C18:3 n-3 (α -linolenic acid), C19:1 (Nonadecenoic acid), C20:1 n-9 (Gondoic acid), C20:3 n-9 (Cis-5,8,11-eicosatrienoic acid) and C22:1 n-9 (Erucic acid) were not found in the oil of the studied peanut varieties. The results of analysis of fatty acids composition in the oil of different peanut varieties showed that 22 out of 91 fatty acids analyzed were found and the fatty acids composition were different among the studied peanut varieties. The fatty acids with high content in the peanut oil of all the peanut varieties are C16:0 (Palmitic acid), C18:1 c9 (Oleic acid), C18:2 n-6 (Linoleic acid), C20:0 (Arachidic acid), C22:0 (Behenic acid), and C24:0 (Lignoceric acid), with the fatty acid C18:1 c9 (Oleic acid) accounting for the highest content, ranging from 43.13 to 50.71 g/100 g sample, followed by C18:2 n-6 (Linoleic acid), ranging from 26.01 to 33.63 g/100 g sample, C16:0 (Palmitic acid), ranging from 11.24 to 12.19 g/100 g sample. The fatty acids with low content in the peanut oil are C22:0 (Behenic acid), ranging from 1.42 to 1.87 g/100 g sample, and C24:0 (Lignoceric acid), ranging from 1.07 to 1.29 g/100 g sample (Table 5 and Figure 2).

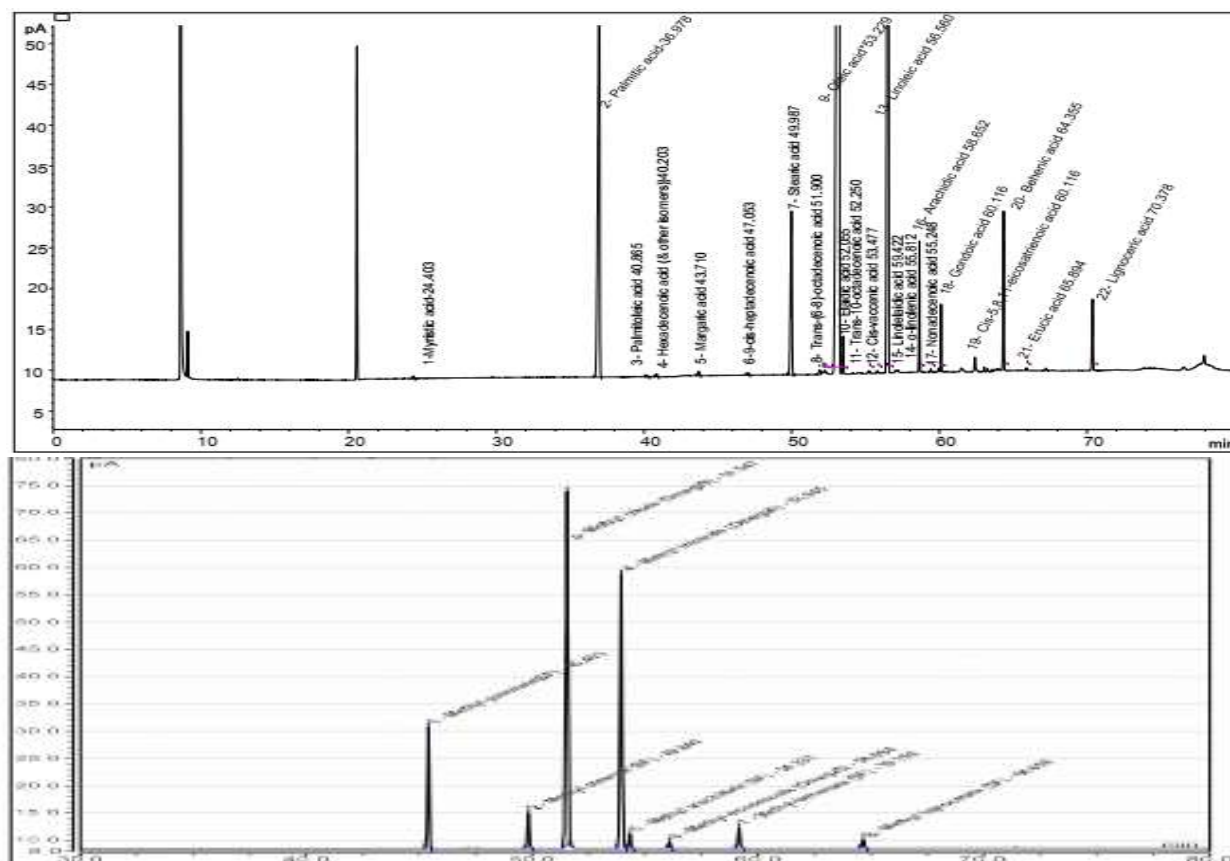


Figure 2: UHPLC chromatogram of fatty acids composition of peanut oil. A: peanut varieties TL03 and B: peanut varieties CNC

Although peanuts and other tree nuts are high in lipids, peanut oil is rich in unsaturated fats, mainly monounsaturated fats (MUFAs) which are associated with lower cardiovascular risk.³¹ Peanut oil collected in the United States has a typical MUFA content ranging from 49 - 57%, and the medium and high oleic acid content of monounsaturated fat (MUFA) in the oil is 66 - 69% and 78 - 80%, respectively.³² Consumption of monounsaturated fats promotes arterial cleansing, which helps keep blood flowing and reduces the risk of atherosclerosis, heart attack, or stroke.³³ Clinical studies have demonstrated that MUFA and PUFA intake is associated with a lower risk of cardiovascular disease (CVD) and death, while the saturated and trans fat intake is associated with a higher risk of CVD.³¹

In this study, the contents of Monounsaturated fatty acids (MUFA) and Polyunsaturated fatty acids (PUFA) in the peanut oil were high accounting for 44.12 - 50.71% (MUFA) and 26.66 - 33.92% (PUFA). The contents of C18:1 c9 (Oleic acid) and C18:2 n-6 (Linoleic acid) accounted for the highest proportion, with their percentage composition ranging from 43.13 - 50.71% and 26.01 - 33.63%, respectively, followed by C16:0 (Palmitic acid) with the content of 12.19 - 12.21%. This result is similar to that obtained from the study of Shin *et al.* on oil of peanut varieties in the US. Therefore, regular peanut consumption may provide sufficient PUFA and MUFA to protect against CVD, certain types of cancer, and age-related cognitive decline.³²

Table 5: Fatty acids composition of different varieties of peanut oil (g/100 g)

No.	Parameters	TL03	LDH09	CNC	L14	L19	TL04	LDH01	LACDOBG	L27	SENNA
1	C 14:0 (Myristic acid)	0.05	-	-	-	0.05	-	-	-	-	-
2	C 16:0 (Palmitic acid)	11.24	11.87	11.34	12.01	11.29	12.11	11.81	11.30	12.21	12.19
3	C 16:1 n-7 (Palmitoleic acid)	0.07	0.07	-	-	0.07	-	0.07	-	-	-
4	C 16:1 (Hexadecenoic acid & other isomers)	0.05	-	-	-	0.04	-	-	-	-	-
5	C 17:0 (Margaric acid)	0.10	0.09	-	-	0.10	-	0.09	-	-	-
6	C 17:1 n-8 (9-cis-heptadecenoic acid)	0.05	0.05	-	-	0.05	-	0.05	-	-	-
7	C 18:0 (Stearic acid)	3.60	3.52	4.85	-	3.63	3.81	3.55	4.90	3.61	3.70
8	C 18:1 tr6-8 (Trans-(6-8)-octadecenoic acid)	0.13	0.06	-	-	0.12	-	0.06	-	-	-
9	C 18:1 tr9 (Elaidic acid)	0.05	0.03	-	-	0.05	-	0.03	-	-	-
10	C 18:1 tr10 (Trans-10-octadecenoic acid)	0.14	0.09	-	-	0.14	-	0.09	-	-	-
11	C 18:1 c9 (Oleic acid*)	47.33	43.13	49.81	45.44	48.12	44.97	43.80	50.71	44.82	44.12
12	C 18:1 c11 (Cis-vaccenic acid)	0.57	0.52	-	-	0.58	-	0.53	-	-	-
13	C 18:2 n-6 trans (Linolelaidic acid)	0.06	0.05	-	-	0.06	-	0.05	-	-	-
14	C 18:2 n-6 (Linoleic acid)	29.50	32.88	26.01	32.62	30.21	33.19	33.63	26.75	32.01	32.35
15	C 18:3 n-3 (α -linolenic acid)	0.04	0.05	-	-	0.04	-	0.05	-	-	-
16	C 19:1 (Nonadecenoic acid)	0.04	0.04	-	-	0.04	-	0.04	-	-	-
17	C 20:0 (Arachidic acid)	1.42	1.47	1.80	1.59	1.45	1.56	1.50	1.87	1.56	1.54
18	C 20:1 n-9 (Gondoic acid)	0.79	0.83	-	-	0.81	-	0.85	-	-	-
19	C 20:3 n-9 (Cis-5,8,11-eicosatrienoic acid)	-	-	0.65	0.79	-	0.73	-	0.66	0.77	0.72
20	C 22:0 (Behenic acid)	1.99	2.45	2.39	2.66	2.05	2.54	2.52	2.48	2.59	2.50
21	C 22:1 n-9 (Erucic acid)	-	0.04	-	-	-	-	0.04	-	-	-
22	C 24:0 (Lignoceric acid)	1.08	1.20	1.29	1.24	1.12	1.09	1.24	1.33	1.23	1.07
SUMMARY											
1	n-3 (Sum of omega-3)	0.04	0.05	-	-	0.04	-	0.05	-	-	-
2	n-6 (Sum of omega-6)	29.55	32.93	26.01	32.62	30.27	33.19	33.68	26.75	32.01	32.35
3	n-9 (Sum of omega-9)	48.16	44.03	50.46	46.23	48.98	45.70	44.72	51.37	55.59	44.84
4	Sum of Trans fat	0.37	0.23	-	-	0.38	-	0.23	-	-	-
5	Saturated fatty acids	19.53	20.60	21.67	21.15	19.65	21.11	20.71	21.88	21.20	21.00
6	Monounsaturated fatty acids (MUFA)	49.21	44.86	49.81	45.44	50.05	44.97	45.56	50.71	44.82	44.12
7	Polyunsaturated fatty acids (PUFA)	29.59	32.97	26.66	33.41	30.31	33.92	33.73	27.41	32.78	33.07
	Total fatty acids	98.33	98.44	98.14	100	100	100	100	100	98.80	98.19

Note: -: peak not present on the chromatogram; 0: Value less than 0.05% relative or 0.005 g/100 g; *: May contain others isomers

The tree showing the genetic relationships of 10 peanut varieties collected from different localities was built based on the presence and absence of vitamins, fatty acids, and some other compounds in peanut oil. The results as presented in Figure 3 show that ten peanut varieties was classified into two main groups; Group I includes four peanut varieties with codes TL03, LDH09, LDH01, and L19. This group is further divided into two subgroups, the first subgroup has only one peanut variety provided by the Institute of Biotechnology, Hue University, and the second subgroup includes two peanut varieties offered by the Agricultural Science Institute for Southern Coastal Central of Vietnam and located in the same branch with cultivar L19 collected from growers in Quang Binh province, Vietnam. Group II includes the remaining six peanut varieties (CNC, L14, TL04, SENNA, L27, and LACDOBG) with similar genetic distances but in different branches.

Conclusion

The study has shown that peanut seeds and peanut oil are rich sources of phytonutrients containing a large number of secondary metabolites, vitamins, fatty acids, and proteins. Different varieties of peanuts collected from different localities in Vietnam showed variations in the composition of these metabolites. Vitamin E, some fatty acids such as C18:1 c9 (Oleic acid), C18:2 n-6 (Linoleic acid), and C16:0 (Palmitic acid) have the highest concentration, with differences among the peanut varieties. Therefore, the multiple nutritional ingredients of peanut oil indicates its potential for use in nutritional products and supplements that can help to improve human health, in disease prevention and treatment.

Conflict of Interest

The authors declare no conflict of interest.

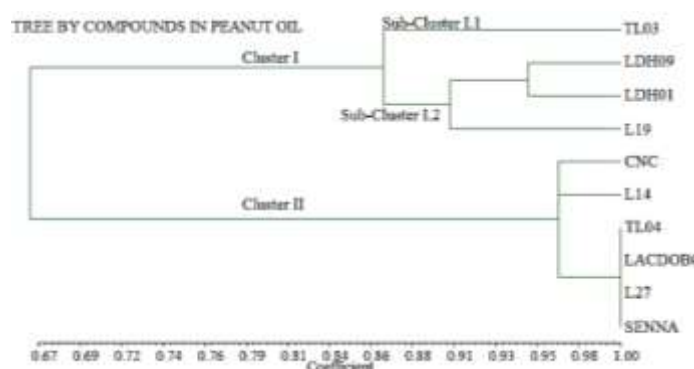


Figure 3: The tree showing the relationship among 10 cultivars of peanut (*Arachis hypogaea*) based on the compounds in the peanut oil

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgments

This research work was supported by a grant from the Ministry of Education and Training, Viet Nam (Grant Number CT-2021-01-DHH-05)

References

- Toomer OT. Nutritional chemistry of the peanut (*Arachis hypogaea*). *Crit Rev Food Sci*. 2018; 58(17):3042-3053.
- Burow MD, Simpson CE, Faries MW, Starr JL, Paterson AH. Molecular biogeographic study of recently described B- and A-genome *Arachis* species, also providing new insights into the origins of cultivated peanut. *Genome*. 2009; 52:107-119.
- Zhao H, Tian R, Xia H, Li C, Li G, Li A, Zhang X, Zhou X, Ma J, Huang H, Zhang K, Thudi M, Ma C, Wang X and Zhao C. High-Density Genetic Variation Map Reveals Key Candidate Loci and Genes Associated With Important Agronomic Traits in Peanut. *Front Genet* 2022; 13:845-602.
- FAOSTAT. Food and Agriculture Organization of the United Nations Statistics Division. Available at [<http://faostat3.fao.org/browse/Q/QD/E>]. Accessed March 18; 2015.
- Mingrou L, Guo S, Ho CT, Bai N. Review on chemical compositions and biological activities of peanut (*Arachis hypogaea* L.). *J Food Biochem*. 2022; 46(7):e14119.
- Belay F, Meresa H, Syum S. Variation and association for kernel yield and yield related traits of released groundnut (*Arachis hypogaea* L.) varieties in abergelle district, northern Ethiopia. *J Med Plants*. 2018; 6:265-271.
- Janila P, Nigam SN, Pandey MK, Nagesh P, Varshney RK. Groundnut improvement: Use of genetic and genomic tools. *Front Plant Sci*. 2013; 4:23.
- Liu C, Hao L, Chen F, Yang C. Study on Extraction of Peanut Protein and Oil Bodies by Aqueous Enzymatic Extraction and Characterization of Protein. *Hindawi J Chem*. 2020; 1-11.
- Rachaputi RCN and Wright G. Peanuts, Overview. *Ref Module Food Sci*. 2016. <https://doi.org/10.1016/B978-0-08-100596-5.00038-X>
- Sáiz J, Montealegre C, Marina ML, Garcia-Ruiz C. Peanut allergens: An overview. *Crit Rev Food Sci Nutr*. 2013; 53(7):722-737.
- Ministry of Science and Technology of Vietnam. TCVN 8951-1:2011 (Oilseeds meals - Determination of oil content - Part 1: Extraction method with hexane (or light petroleum). 2011.
- Bradford MM. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem*. 1976; 72:248-254.
- AOAC Official Method 923.09. Invert Sugar in Sugar and Syrups. Lane-Eynon General Volumetric Method.
- Nguyen Van Mui. Practice biochemistry. Hanoi National University Publishing House. 2001.
- SIST-TS CEN/TS 17062:2019 - Foods of plant origin - Multimethod for the determination of pesticide residues in vegetable oils by LC-MS/MS (QuOil). 2019.
- Ministry of Science and Technology of Vietnam. TCVN 9675-2:2013 (ISO 12966-2:2011). Animal and vegetable fats and oils - Gas chromatography of fatty acid methyl esters - Part 2: Preparation of methyl esters of fatty acids. 2013.
- COI/T.20/Doc. No. 24:2001. Preparation of the fatty acid methyl esters from olive oil and olive-pomace oil. 2001.
- Nautiyal PC. GROUNDNUT - Food and Agriculture Organization. Available from http://www.fao.org/fileadmin/user_upload/inpho/docs/Post_Harvest_Compendium_-_Groundnut. 2019. Accessed August 6, 2002.
- Li Y, Qian H, Sun X, Cui Y, Wang H, Du C, Xia X. The effects of germination on chemical composition of peanut seed. *Food Sci Technol Res*. 2014; 20:883-889.
- Zhao X, Chen J, Du F. Potential use of peanut by-products in food processing: A review. *J Food Sci Technol*. 2011; 49:521-529.
- Lutz M and Luna L. Nuts and body weight - an overview. *J Nutr Health Sci*. 2016; 3:1-7.
- Glinko A, Bozym MJ, Owens ML, Usher KM. Reversed-phase HPLC separation of water-soluble vitamins on agilent ZORBAX eclipse plus columns. *Agilent Technol*. 2008; 1:1-7.
- Bonku R and Yu J. Health aspects of peanuts as an outcome of its chemical composition. *Food Sci Hum Well*. 2020; 9:21-30.
- Sarwar F. The role of oilseeds nutrition in human health: a critical review. *J Cereals Oilseeds*. 2013; 4:97-100.
- Zhang Y, Zhou WE, Yan JQ, Liu M, Zhou Y, Shen X, Ma YL, Feng XS, Yang J, Li GH. A review of the extraction and determination methods of thirteen essential vitamins to the human body: an update from 2010. *Molecules*. 2018; 23(6):1484.
- Settaluri VK, Kandala N, Puppala JS. Peanuts and their nutritional aspects-a review. *Food Nutr Sci*. 2012; 3:1644-1650.
- Whitney EN and Rolfes SR. Understanding Nutrition, 15th ed. Boston, MA, USA: Cengage. 2019.
- Fernández-Muino MA, Sancho-Ortiz MT, Valls-García F. Water-soluble vitamins. In: W.J. Hurst (Ed.), *Methods of Analysis for Functional Foods and Nutraceuticals*. CRC Press, Boca Raton. 2002. 275-287p.
- Nadathur SR, Wanasundara JP, Scanlin L. Sustainable protein sources, Academic Press in an imprint of Elsevier. Amsterdam. 2017.
- Griel AE, Eissenstat B, Juturu V, Hsieh G, Kris-Etherton PM. Improved diet quality with peanut consumption. *J Am Coll Nutr*. 2004; 23(6):660-668.
- Sacks FM, Lichtenstein AH, Wu JHY, Appel LJ, Creager MA, Kris-Etherton PM, Miller M, Rimm EB, Rudel LL, Robinson JG, Stone NJ, Van Horn LV. Dietary fats and cardiovascular disease: a presidential advisory from the American Heart Association. *Circul*. 2017; 136(3):e1-e23.
- Shin EC, Pegg RB, Phillips RD, Eitenmiller RR. Commercial Runner peanut cultivars in the USA: fatty acid composition. *Eur J Lipid Sci Technol*. 2010; 112:195-207.
- National Heart, Lung, and Blood Institute. Atherosclerosis. Available from <https://www.nhlbi.nih.gov/health-topics/atherosclerosis>. 2019.