

Variation in Physico-Chemical Constituents and Lipid Composition Among *Arachis Hypogea* Cultivars

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Summary: Four cultivars of ground nut (*Arachis hypogea*) were collected from different areas of Pakistan (Tillagang, Gujar Khan). The oils were extracted for the evaluation of lipid. The physico-chemical values of oils were determined like refractive index, peroxide value, unsaponifiable matter, acid value, iodine value, free fatty acid and ester value. The lipid profile as indicated by GLC showed that palmitic acid ranged from 8.2-8.8%, proportions of oleic acid and linoleic acid varied from 57.8-59.87% and 22.5-24.1% respectively. Gujar Khan variety of peanut has higher % age of oil (56.79). The higher yield of oil in this variety is not at the cost of any nutritional quality of peanut oil.

Introduction

Arachis hypogea belongs to the family Leguminosae. About 16400 species in over 657 genera are of cosmopolitan distribution living in every soil and climate. Trees, shrubs, water plants, xerophytes and climbers are their representatives [1]. Leguminosae is the most important family in the Dicotyledonae [2]. *Arachis hypogea* is the second largest oilseed crop after rape/mustard seeds in the country. The nut is normally composed of 25-35% shell and 65-70% kernel that in turn contained 45-55% oil and 25-30% protein [3]. Oil derived from peanut is pale gold and subtly flavored with the richness of peanut [4]. Oil is used in cooking and in the preparation of shortening, margarines and mayonnaise. It is also used as salad oil and the meal is used as food and also for feed purpose [5-7]. Peanut oil has health promoting chemistry. It has more than 80 percent oleic fatty acid, therefore, affects HDL and LDL ratios positively, reduces platelet stickiness, increase circulation, reduces blood pressure, slows cholesterol production, prevents inflammation, reduces joint tenderness, stimulates the immune system, assists the balancing of hormones especially as related to PMS and menopause and can lower blood pressure and alleviate other cardiovascular disorder [8]. Due to the importance of the oil of peanut better planning and efforts are needed for increasing the production of groundnut by introducing high yielding and high quality varieties. The groundnut crop has not yet been considered for increasing the potential of edible oil in the country. The present work is therefore a step in this direction.

Edible oil shortage has been a chronic problem in Pakistan for the past three decades and consequently imports have been increasing to meet the local market demands. The import of edible oils during some last years has been high, 1.6 to 2.0 million tons at heavy cost of foreign exchange equivalent to Rs 35-40 billions. Pakistan's per capita consumption of about 14 kg. per annum is one of the highest in the world against the world average of 3 kg. per annum. The local production of edible oils in the country caters about 30 percent of the local demand and the remaining 70 is met by imports. [9, 10]. Therefore, it is essential to increase the local oil production by utilizing all available agriculture resources including the groundnut. Four peanut varieties cultivated in Gujar Khan, Pipplan, Hanoi and Tillagang have now been studied to evaluate their oil in the country. Specific techniques have been used for extraction of oils; their purification, identification and their characterization. The oils were hydrolyzed, methylated and the fatty acid composition of these oils were determined by means of GLC.

Results and Discussion

By using methanol and chloroform mixture, the oils of the peanuts varieties were extracted (Table-1). The solvent was then removed at reduced pressure to avoid changes like polymerization and decomposition of oils. The % age yield of four varieties is given in Table-1. All the varieties showed almost the same range of yield as indicated in the

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Table-1: % age of oils of four cultivars of peanut.

S. No.	Sample	Solvent used in extraction	Weight of peanuts in grams	Yield of oil in grams	% Yield of oil
1	Peanut oil of Pipplan variety	Chloroform: Methanol	59.018 gm	23.503 gm	39.83 %
2	Peanut oil of Hanoi variety	Chloroform: Methanol	55.620 gm	21.620 gm	38.42 %
3	Peanut oil of Gujar Khan variety	Chloroform: Methanol	32.827 gm	18.645 g	56.79 %
4	Peanut oil of Tillagang variety	Chloroform: Methanol	46.899 gm	20.566 g	43.83 %

earlier studies [11] except the Gujar Khan variety which showed the higher % age of yield (56.79 %) and Hanoi variety has minimum oil % age (38.42 %). The differences in the oil % age of the four varieties may be due to different soil properties and climatic conditions of different areas.

Chemical constituents are valuable to evaluate oils with respect to its utilization. Some important physico-chemical properties of *Arachis hypogea* oils of all four varieties were determined to evaluate the quality of the oil. These are given in Table-2. Oils can be classified on the basis of iodine value. The iodine values of all the varieties were 95.76, 95.68, 101.38, 98.73 for which the oil is considered to be semi drying. Gujar Khan variety peanut is highly monounsaturated and considered to be high oleic acid oil. The saponification values 167.32 of Hanoi, 186.23 of Pipplan, 212.11 of Gujar Khan and 186.67 of Tillagang show the presence of fatty acids having high molecular weight. Low free fatty acids present in the oils of all peanut varieties indicate that the oils of all varieties are fit for human consumption. The refractive indexes of all varieties are slightly different from each other. Its mean density, molecular weight and internal arrangement of fatty acids, related to refractive index, are slightly different from each other. This is due to cultivation of crops in different regions of country which have slightly different climatic conditions. High peroxide values of the oils indicated formation of epoxides. The high values are due to the crude form of oils. The oils of this range of peroxide value in crude form is considered to be fit for edible purpose. The other physicochemical values of the four varieties are more

Table-2: Physico-chemical values of lipids of four cultivars of peanut.

S. No.	Physico Chemical Property	Peanut oil of Hanoi Variety	Peanut oil of Pipplan Variety	Peanut oil of Gujar Khan Variety	Peanut oil of Tillagang variety
1	Refractive index	1.4633	1.4631	1.4637	1.4635
2	Peroxide Value	3.74	10.13	6.49	5.60
3	Iodine value	95.68	95.76	98.73	101.38
4	Saponification value	187.32	186.23	212.11	186.67
5	Unsaponifiable matter	0.018	0.080	0.318	0.204
6	Acid value	0.34	0.55	0.45	0.52
7	Free fatty acid	0.174	0.275	0.227	0.262
8	Ester value	186.97	185.68	211.66	186.15

or less the same with only slight variation in them. The variation may be due to different climatic condition and soil properties. The pattern of variation is more or less similar as indicated in earlier studies [12, 13]. J. E. Eheart (1955) [14] also confirmed this.

The fatty acid profile of the peanuts oil of all varieties (Table-3) show higher percentage of unsaturated fatty acid as compared to saturated fatty acids which is according to the earlier investigating by Anderson and H. E Pattee (1967) [15]. Among saturated fatty acid palmitic acid C16:0 present in high concentration among all the varieties, stearic acid C 18:0 also present but not in huge amount. Arachidic acid present in very low concentration in all the four peanut varieties and the characteristics of this oil. High % age of unsaturated acid is the characteristics of vegetable oil. Oleic acid is the major fatty acid in all the four varieties of peanut, this high concentration C 18:1 is very useful for human because it lower HDL, which decreases CVS risk [16].

Table-3: Fatty acid profile of four cultivars of peanut.

S. No.	Name of Fatty Acid	Short hand name	Peanut oil of Hanoi	Peanut oil of Pipplan	Peanut oil of Gujar Khan	Peanut oil of Tillagang
1	Capric acid	C 10:0	T	T	0.09	T
2	Lauric Acid	C 12:0	T	0.05	0.02	0.2
3	Myristic acid	C 14:0	T	0.03	0.02	0.2
4	Palmitic acid	C 16:0	8.2	8.43	8.20	8.82
5	Palmitoleic acid	C 16:1	T	T	T	T
6	Stearic Acid	C 18:0	3.9	4.3	2.5	3.
7	Oleic acid	C 18:1	59.87	57.8	58.7	59.1
8	Linoleic acid	C 18:2	22.9	22.5	24.1	22.7
9	Linolenic acid	C 18:3	1.3	1.38	1.43	1.48
10	Arachidic acid	C 20+higher or above	3.7	4.8	3.9	3.6

Table-4 showed more than 90 % yield of neutral lipids and lesser concentration of polar lipids in all peanut varieties.

Table-4: Yields of neutral and polar lipids of four cultivars of peanut.

Cultivars	Neutral Lipids	Polar Lipids
Tillagang	93.8 %	6.2 %
Gujar Khan	94.2 %	5.8 %
Hanoi	96.3 %	3.7 %
Pipplan	96.8 %	3.2 %

Similarly both neutral and polar lipids of all the four cultivars exhibited very high concentration of oleic acid. Proportions of fatty acids of polar and neutral lipids of all the four cultivars are shown in Table-5.

Table-5: Proportions of fatty acids in neutral and polar lipids of four cultivars of peanut.

Cultivars	Proportions of fatty acids				
	C 16:0	C 18:0	C 18:1	C 18:2	C 18:3
	Neutral Lipids				
Tillagang	9.6	3.4	60.3	21.5	1.6
Hanoi	9.3	4.1	59.94	23.4	1.47
Pipplan	8.8	4.6	58.37	20.93	1.05
Gujar Khan	8.75	2.73	59.08	25.3	1.7
	Polar Lipids				
Tillagang	15.7	3.08	57.4	20.03	2.1
Hanoi	14.6	4.5	57.09	22.7	1.8
Pipplan	9.1	3.9	59.79	23.8	1.84
Gujar Khan	10.8	4.75	60.8	19.9	1.73

Overall the present study indicates that Gujar Khan variety produces significantly higher yield of peanut oil, which also contained higher % age of linoleic acid. This is may be due to the reason that vegetable oils form temperature region contained more linoleic acid as compared to other regions. It is noteworthy that the higher yield obtained in Gujar Khan variety is not at the cost of other positive values linked to peanut qualities, It is therefore imperative that Gujar Khan variety of peanut be promoted by the Agricultural Department to enhance the availability of edible oil in the country.

Experimental

Extraction of Oil

The seeds of peanuts of four varieties were procured from four different areas of Pakistan like Gujar Khan, Pipplan, Hanoi and Tillagang. The seeds were dried in oven at 1.50 °C and crushed into fine powder. The lipids were extracted with 500ml chloroform: methanol (2:1 v/v) [17] mixture at room temperature by shaking on magnetic stirrer for 2

hours. After filtration the residual material was treated three times with 100 ml of same mixture. All the extracts were combined and three consecutive washings with Folch solution [18] were given to remove the non-lipid impurities. After removal of solvent under reduced pressure, the oil was stored in an inert atmosphere.

Physico Chemical Values of the Oil

The physico-chemical values like saponification value, iodine value, ester value, free fatty acid were determined according to British standard specification and [19] procedure. Refractive index was determined with Abbe's refractometer.

The oil was refluxed with 0.5 N alcoholic potassium hydroxide solutions for 3 hours. The soap solution reacted with 2 N H₂SO₄ to liberate fatty acids after separation of unsaponifiable matter by diethyl ether [20]. The methyl ester of fatty acids was prepared with BF₃- Methanol reagent [21].

Separation of Lipid Classes from Oil

The lipid classes were fractionated quantitatively by thin layer chromatography. Known weight of lipid (10 % solution in chloroform) was applied on plate 3 cm above the lower edge of the plate coated with 0.5 mm silica gel. The developing media [22] for neutral and polar lipids were hexane: diethylether:aceticacid (80:20:2 v/v) and chloroform:methanol:30 % ammonium hydroxide:water (60:35:5:2.5 v/v). The developing bands were identified by comparing their R_f values with standards and also by applying their specific color tests.

Identification of Fatty Acids by GLC

The methyl ester of lipid fractions were analyzed on Shimadzu GC-4A gas chromatograph equipped with flame ionization detector and polar (PEG) capillary column (25mx 0.2mm i.d). The temperature programming of column oven was 180 °C-3 °C/min-220 °C. Nitrogen was used as carrier gas with a flow rate of 2 ml/min. The temperature of injector and detector was 230 °C and 250 °C respectively. The peaks were temperature of injector and detector was 230 °C and 250 °C respectively. The peaks were recorded on Shimadzu CR-4A chromatopac and identified by comparing their retention time with those of standards run under the same parameters.

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