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Estimation of Physico-Chemical properties of different peanut species (*Arachis hypogaea*) grown in India

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Abstract

In the present study four different groundnut varieties such as TMV-2(Tmv-2) ,G2-52,Dharni (Dn) and Kardi-9 (Kd-9) were grown under different geographical places in India. Physicochemical properties of collected groundnut seeds and their extracted oils were determined for their nutritional assay. It was found that the seeds contained in the range of extracted oil, the specific gravity, viscosities, impurities, refractive index and colour of the extracted groundnut seed oil were in the range of 38.98-40.48 (± 0.6671), 0.915-0.916 (± 0.0004), 91.55-91.66 (± 0.0513), 0.035-0.06 (± 0.0104), 1.4628-1.4638 (± 0.0004) and 1.12-2.1 (± 0.4414) yellow units respectively. AV (mg KOH/g oil), IV (g I₂/100 g oil), SV (mg KOH/ g oil), ester value and unsaponifiable matter content (%) of the extracted oil from groundnut seeds were in the range of 1.81-2.43 (± 0.5861), 88.47-92.8 (± 1.9975), 190.08-192.47 (± 1.006), 187.7-190.04 (± 1.0903) and 0.52-0.63 (± 0.0535) % respectively. This study is empirical and on the basis of finding it revealed that groundnut seed oil can be a valuable source of edible oil.

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Introduction

Nuts are a good source of oil containing higher unsaturated fatty acids (UFAs) to saturated fatty acids (SFAs) ratio (Sabate, 2003). The pea nut, often called as "The King of Oilseeds", is botanically known as *Arachis hypogaea* and belongs to family Leguminosae, which is also called Fabaceae. The pea nuts differ in the quantity as well the quality of oil. These differences in the pea nut oil may be due to several factors *i.e.* genotype, the level of maturity of the seed, season and geographical area of production (Brown *et al.*, 1975). About 80% of the total fatty acid content of pea nut oil constitutes unsaturated fatty acids mainly oleic acid and linoleic acid Thus the chemistry and quality of pea nut oil mostly depend on the oleic to linoleic ratio (Ahmed & Young,

1982). Groundnut is now grown worldwide in the tropical and temperate zones primarily as an oil seed crop (Bansal *et al.*, 1993). The nutritive value of food is high as the groundnut is affordable and serves as good source of oil and protein (Atasie *et al.*, 2009). India is the largest producer of groundnut in the world. Around 88% of the groundnut area and production in India is concentrated in five states: Andhra Pradesh, Gujarat, Karnataka, Tamil Nadu, and Maharashtra. Nearly 83% of the total area is under rainy-season groundnut and the other 17% is cultivated during the post rainy season. During 1995-98, groundnut was grown in India over 7.47 Mha with a total production of 8.02 Mt (CMIE, 2000). The fat content in groundnut has been largely studied. In general, Groundnut seed contain 44-56% oil and 22-30% protein on a dry seed basis and is a rich source of minerals such

as phosphorus, calcium, magnesium and potassium and vitamins like E, K and B group (Savage *et al.*, 1994).

India possesses varying climatic conditions results in cultivation of a wide range oil bearing crops trees and nuts. Peanuts make an important contribution to the diet in many countries. Peanut seeds are a good source of protein, lipid and fatty acids for human nutrition (Young *et al.*, 1975). The oil content of groundnut differs in quantity, the relative proportion of fatty acids, geographical location, seasons and growing conditions (Adeyeye *et al.*, 1992). Barku *et al.*, 2012 have reported changes on the chemical composition as a result of processing. However, little information on the effect of traditional processing on peanuts quality was reported. The chemical properties of oils are amongst the most important properties that determine the quality and help to describe the present condition of oils. Its constitute one of the essential components of balanced diet as good source of energy. The study indicated that Peanut oil, may have a higher shelf life, nutritional value and industrial applications. Vegetable oil had made an important contribution to the diet in many countries.

Aim of this study is to investigate and evaluate physical and chemical composition of Groundnut seed varieties of TMV-2(Tmv-2), G2-52, Dharni (Dn) and Kardi-9 (Kd-9) which are having different geographical places. In this study the groundnut seeds of different places were assessed and analyzed for yield of extracted oil. The extracted oils were analyzed for physical and chemical parameters, such as refractive index, specific gravity, viscosity, impurities, colour, iodine value (IV), acid value (AV), saponification value(SV) and unsaponifiable matter etc.

Materials and Methods

Procurement of Materials

Proximate, physical and chemical analysis of seeds and extracted oil were based on four peanut varieties namely TMV-2(Tmv-2), G2-52, Dharni (Dn) and Kardi-9 (Kd-9) have been used for cultivation in arid zone as well as irrigated regions of Rajasthan, Madhya Pradesh and Gujarat then used for extraction of oil which was provided by oil mills association, Jalgaon (Maharashtra). The seeds and pod shells were separated manually. For analysis, mature and healthy seeds were stored in grinded form in glass containers. The chemical reagents and glassware are used in this research work are analytical grade.

Methods

Extraction of oil of collected seeds

The groundnut oil seed were purchased from local market. The groundnut seeds were separated from shaft by hand picking method. The seeds were freed of the dirt were collected into a separate pre cleaned beaker. From each sample 500 g were crushed and weighed using commercial grinder and fed to a Soxhlet extractor and hexane was used as the extraction solvent, equipped with thimble and fitted with a 2 L round bottomed flask. The extraction was carried out for a period of 8 hours. At the end of the extraction period, the solvent was recovered by using a rotary evaporator and residual oil was dried at 75⁰ C for one hour. The extract was transferred to desiccators and then stored in air tight container until needed for further analysis (Pearson, 1981).

The amount of oil extracted was determined using the following equation

$$\text{Oil content (\%)} = \frac{\text{weight of oil extracted}}{\text{weight of seed}} \times 100$$

Determination of physical and chemical properties of extracted oil

The extracted oil was immediately analyzed for chemical properties, such as iodine, acid and saponification value, ester value and unsaponifiable matter while specific gravity, viscosity, refractive index, impurities and colour were examined for physical properties. The refractive indices of the oil at room temperature were determined with Abbe/ Butyro Refractometer and the specific gravity measurement (also carried out at room temperature), using specific gravity bottle. The state and colour of the oil were noted, using Lovibond tintometer at room temperature. Viscosity measurement with Hakke viscometer (rheoVT550) at room temperature and yield were determined, using the method described by the association of official chemists (AOAC). Results are expressed as the means of three separate determinations.

Determination of physical properties of extracted oil

Results of the physical properties of the selected oils examined are shown in Table 1.

Determination of colour

The method determines the colour of oils by comparison with Lovibond glasses of known colour characteristics. The colour is expressed as the sum of the yellow and red

slides used to match the colour of the oil in a cell of the specified size in the Lovibond Tintometer. Clean the glass cell of desired size with carbon tetrachloride and allow it to dry. Fill it with oil and place the cell in position in the tintometer. Match the colour with sliding red, yellow and blue colours. Reports the colour of the oil in terms Lovibond units as follows (ISI, 1984).

$$\text{Colour reading} = (a Y + 5 b R) \text{----- (i)}$$

Where, a= sum total of the various yellow slides (Y) used, b = sum total of the various red (R) slides used

Determination of the Refractive index at 40 °C

The refractive index was measured, as this figure can help determine the level of unsaturated of the fatty acids in oils, a nutritive quality of interest for this study (Nagre *et al.*, 2011). Measurement of the refractive index of the sample is done by means of a suitable Butyro refractometer at 40°C, a refractometer was used to measure the refractive index of extracted oils. Distilled water which has refractive index 1.3330 at 20°C and 1.3306 at 40°C, the usual temperature of taking readings. Make sure sample is completely dry, circulate stream of water through the instrument. Adjust the temperature of the refractometer to the desired temperature. Ensure that the prisms are clean and dry. Place a few drops of the sample on the prism. Close the prisms and allow standing for 1-2 min. Adjust the instrument and lighting to obtain the most distinct reading possible and determining the refractive index or butyro-refractometer number. (ISI, 1984).

Determination of specific gravity

The specific gravity of extracted oil was recorded as a general measure of oil density compared to the density of water (Nagre *et al.* 2011). This is useful for physically comparing and identifying oils. The specific gravity was determined using the specific gravity bottle method. The following formula was used to calculate the specific gravity of extracted oils (ISI, 1984).

$$\text{Specific gravity} = (\text{weight of bottle} + \text{oil}) - (\text{weight of bottle}) / (\text{weight of water}) \text{----(ii)}$$

Impurities

The level of impurities (mesocarp fibers, insoluble materials, phosphatides, trace metals, and oxidation

products) was measured in each oil, as high levels of these substances are typically prohibited in the regulated production of edible oils (Watanapoon, 2004). Two grams (2 g) of oil was weighed into a 500 ml flask and mixed with 20 ml of a 1:1 solvent (petroleum ether and diethyl ether). The contents were vigorously shaken, covered, and allowed to stand for 24 hours. The mixture was filtered through a weighed 11 cm qualitative filter paper. The paper was then washed with 10 ml of the 1:1 solvent and placed in an oven at 103 °C for one hour. The dried paper was then weighed. The impurity (%) of oil was calculated with the following formula (DGHS, 2012).

$$\text{Impurities (\%)} = (w_2 - w_1) / w_3 \text{----- (iii)}$$

w_2 = Weight of paper before filtering, w_1 = Weight of paper after filtering, w_3 = Weight of initial sample.

Determination of viscosity

The viscosity of extracted oil was measured as an additional proxy for fat unsaturation, as prior studies have described an inverse relationship between viscosity and fatty acid unsaturation in oils (Abramovic, 2012). Viscosity was determined at room temperature 25°C, using a Hakke viscometer (rheo VT550).

Determination of chemical properties of extracted oil

Results of the chemical properties of the selected oils examined are shown in Table 2.

Determination of acid value

The acid value, an indirect measurement of free fatty acid levels, was recorded to test the oils' freshness and likeliness to develop taste and odor defects (Akinoye *et al.*, 2011, Nagre *et al.*, 2011). The acid value is determined by directly the oil in an alcoholic medium against standard potassium hydroxide/sodium hydroxide solution. Mix the oil or melted fat thoroughly before weighing. Weigh accurately about 5 to 10 g of cooled oil sample in a 250 ml conical flask and 50 ml to 100 ml of freshly neutralised hot ethyl alcohol and about one ml of phenolphthalein indicator solution. Boil the mixture for about five minutes and titrate while hot against standard alkali solution shaking vigorously during the titration (DGHS, 2012).

$$\text{Acid value} = (56.1) (V) (N) / W \text{----- (iv)}$$

Where V = Volume in ml of standard sodium hydroxide solution used, N = Normality of the standard sodium hydroxide solution, and W = Weight in g of the sample.

Determination of iodine value (Wij's method)

The iodine value of such oil was measured, as this value is also useful for determining the unsaturation level of the fatty acids in oil (Akinyeye *et al.*, 2011). The iodine value of an oil / Fat is the number of grams of iodine absorbed by 100 g of the oil /fat, when determined by using Wijs solution. The oil / fat sample taken in carbon tetrachloride is treated with a known excess of iodine monochloride is treated with glacial acetic acid (Wijs solution) The excess of iodine monochloride is treated with potassium iodide and the liberated iodine estimated by titration with sodium thiosulphate solution. The iodine value is a measure of the amount of unsaturation (number of double bonds) in a fat (AOAC, 2000).

$$\text{Iodine value} = 12.69 (B - S) N / W \text{ ----- (v)}$$

Where, B = volume in ml of standard sodium thiosulphate solution required for the blank,

S = volume in ml of standard sodium thiosulphate solution required for the sample, N = normality of standard sodium thiosulphate solution, W = weight in g of the sample.

Determination of Saponification value

The saponification value is the number of mg of potassium hydroxide required to saponify 1 gram of oil /fat. The saponification value of such oil was measured to explore the potential industrial uses for the oils, as this parameter reveals oil's suitability to be made into soap (Cuppert, 2001). Two grams (2 g) of oil was dissolved in 25 ml of alcoholic potassium hydroxide. The mixture was refluxed for 45 minutes and then cooled. 1 ml of phenolphthalein indicator was added. The solution was titrated using 0.5 M HCL. A blank determination was conducted. The oil sample is saponified by refluxing with a known excess of alcoholic potassium hydroxide solution. The alkali required for saponification is determined by titration of the excess potassium hydroxide with standard hydrochloric acid (AOAC, 2000).

$$\text{Saponification value} = 56.1 (B - S) N / W \text{ ----- (vi)}$$

Where, B= Volume in ml of standard hydrochloric acid required for the blank, S = Volume in ml of standard hydrochloric acid required for the sample, N = Normality of the standard hydrochloric acid, W = Weight in gm of the oil .fat taken for the test.

Determination of ester value

The ester value is the 'mg' of KOH required to react with glycerol/ glycerin after saponify 1 g of oil sample. Ester value is calculated by the following relation

$$\text{Ester Value} = \text{Saponification Value} - \text{Acid Value}$$

Determination of Unsaponifiable matter

The Unsaponifiable matter was determined using the neutralized liquid after titration for the determination of saponification value. The neutralized liquid was transferred quantitatively into a separating funnel using 50ml of water for washing the flask. Add to the flask 50ml of petroleum ether, shake vigorously, and allow the layers to separate. Transfer the lower soap layer into another separating funnel and repeat the ether extraction for another 3 times using 50 ml portions of petroleum ether. Wash the combined ether extract three times with 25 ml portions of aqueous alcohol followed by washing with 25 ml portions of distilled water to ensure ether extract is free of alkali (washing are no longer alkaline to phenolphthalein) Transfer ether solution to 250 ml beaker, rinse separator with ether, add rinsing to main solution. Evaporate to about 5 ml and transfer quantitatively using several portions of ether to Erlenmeyer flask previously dried and weighed. Evaporate ether. When all ether has been removed add 2-3 ml acetone and while heating on steam or water bath completely remove solvent under a gentle air. To remove last traces of ether, dry at 100 OC for 30 minutes till constant weight is obtained dissolve residue in 50 ml of warm ethanol which has been neutralised to a phenolphthalein end point. Titrate with 0.02 N NaOH (AOAC, 2000).

$$\text{Weight in g of the free fatty acids in the extract as oleic acid} = (0.282) (V) (N)$$

Where, V = Volume in ml of standard sodium hydroxide solution, N = Normality of standard sodium hydroxide solution

$$\text{Unsaponifiable matter} = 100 (A - B) / W \text{ ----- (vii)}$$

Where, A = Weight in g of the residue, B = Weight in g of the free fatty acids in the extract

W = Weight in g of the sample.

Statistical analysis

The statistical parameter like standard deviation, coefficient of variance and standard mean error were calculated for physical and chemical parameters. All the experiment was carried out in triplicate and the results are presented as the mean \pm SD, CV, \pm SEM. Descriptive statistics of different groundnut varieties from different parts of India as shown in figure 1.

Results and Discussions

Results of the physical and chemical characteristics of the different varieties of groundnut seed extracted oil were presented in (Table 1 and 2). Results obtained from this work indicate that the oil yield ranges from 38.98-40.48 and the colour in yellow units ranges from 1.12 to 2.1 on Lovibond tintometer. Specific gravity ranges from 0.915 to 0.916 for all the varieties of groundnut. The value of the viscosity of the various extracted oils ranges from 91.55 to 91.66. The refractive index analysis shows that the values between 1.4628 and 1.4638, the refractive index increases as the double bond increases. Others could be attributed to the presence of some impurities and other components of the crude oil mixture ranges from 0.035-0.06%. The acid value of the oils as determined ranges from 1.81-2.43 mg KOH/g oil. The saponification values of the various oils were found to be

in ranges from 190.08 to 192.47. The iodine values show increase in the average degree of un-saturation of the oil, ranges from 88.47 to 92.8. Unsaponifiable matter in the range from 0.52-0.63, ester value ranges from 187.7-190.04. As a result of their agreement with standard, all the oils could be classified as non-drying oils; since their iodine values are lower than 100 (gI₂/100 g sample) .

Conclusion

The physicochemical properties of different varieties of groundnut seed oils have been analyzed. Though the saponification value is high, a property adequate for soap making industry, a low iodine value in the nut is the suitability for cooking. All the oils have very low degree of unsaturation and could be classified as non-drying oils. The percentage oil content of most of the seeds selected from different varieties of groundnut, show them as high oil yielding. Groundnut characteristically contained high level of oil and this makes it a potential source of edible oil. The most of the values obtained complied with the standard specified as per the food product and standards regulations 2011 of Food safety and standard act 2006, under the food safety and standards authority of India (FSSAI). The oil is of good quality and could be recommended as suitable for cooking usage especially, all the oils show good tendency to be used as unrefined. It is concluded that all the groundnut varieties are revealed that groundnut seed oil can be a valuable source of edible oil.

Table.1 Physical properties of different groundnut extracted seed oils.

Sr. No	Oils / Name of varieties	Yield (%)	Specific gravity	Viscosity at 25 ⁰ C/CP	Impurities (%)	RI at 40 ⁰ C	Colour (Yellow unit)
1	Tmv-2	40.29	0.9160	91.66	0.035	1.4638	1.22
2	G2-52	38.98	0.9158	91.58	0.05	1.4628	1.12
3	Dn	40.48	0.9150	91.55	0.06	1.4630	2.10
4	Kd-11	39.95	0.9154	91.64	0.045	1.4634	1.42
5	Total	159.7	3.6622	366.43	0.19	5.853	5.86
6	Mean	39.93	0.9156	91.61	0.048	1.4633	1.47
7	SD	0.6671	0.0004	0.0513	0.0104	0.0004	0.4414
8	CV	1.6706	0.0488	0.056	21.7506	0.0306	30.0246
9	SEM	0.3335	0.0002	0.0257	0.0052	0.0002	0.2207

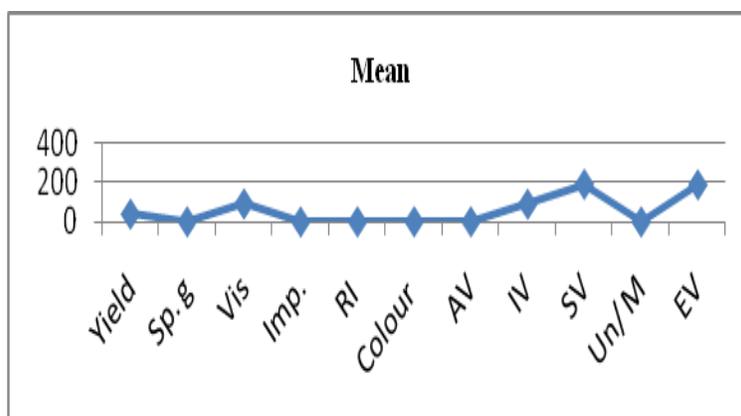
Values are mean \pm SD, SEM (standard mean error), RI-Refractive index, SD-Standard deviation, CV-Coefficient of variance

Table.2 Chemical properties of different groundnut extracted seed oils.

Sr. No	Oils /Name of varieties	AV* (mg KOH/g)	IV (Wijs)	SV (mg KOH/g)	Unsaponifiable matter (g/kg)	EV
1	Tmv-2	2.29	89.22	191.26	0.63	188.97
2	G2-52	1.81	92.8	191.76	0.54	189.95
3	Dn	2.38	88.47	190.08	0.52	187.7
4	Kd-11	2.43	91.46	192.47	0.61	190.04
5	Total	6.53	361.95	765.57	2.3	756.66
6	Mean	1.63	90.49	191.39	0.575	189.17
7	SD	0.5861	1.9975	1.006	0.054	1.0903
8	CV	31.9592	2.2075	0.5256	9.2318	0.5763
9	SEM	0.2931	0.9988	0.503	0.0268	0.5451

AV-Acid value, IV-Iodine value, SV-Saponification value, EV-Ester value, SD-Standard deviation, CV-Coefficient of variance, values are mean ±SD, SEM (standard mean error)

Figure.1 Accuracy of descriptive statistics of different varieties of groundnut grown in different parts of India



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