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Comparative Analysis of Biochemical Parameters of Seeds from New Accessions in the Groundnut (*Arachis hypogaea L.*) Collection

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Abstract This research article reveals findings from a comparative study that assessed both protein and oil levels in groundnut seeds belonging to foreign and local *Arachis hypogaea L.* collections. Research revealed several accessions including PL 1 from India, Hippagi 2-20 from India, Var Cuba from Japan, Virginia Improved from Israel, Zac Trang from Vietnam, and Philippine Pink from Portugal which show significant seed protein and oil content levels. Researchers regard these accessions as essential genetic resources to create future groundnut cultivars with greater yields and improved biochemical properties.

Keywords Groundnut (*Arachis hypogaea L.*), Protein content, Oil content, Genetic resources, International accessions, Seed quality, Plant breeding, Nutritional value

1. Introduction

Expanding populations worldwide coupled with increased food product requirements present multiple serious challenges to the agricultural industry. The biggest challenge currently faced by the agricultural sector is how to maintain a supply of ecologically clean agricultural products. Industrial-scale production now more frequently utilizes leguminous and oilseed crops along with nutritionally dense plants. The solution to current protein deficiency problems requires us to choose plant species and their varieties which produce plentiful yields and high-quality proteins. Oilseed crops including sunflower, flax, sesame, soybean, and groundnut hold significant value because their products produce edible oils containing essential nutrients. The worldwide market shows a strong preference for natural food products that support human health and contain essential minerals, macro- and microelements, vitamins, and organic compounds. These crops serve a vital function within both the food sector and the larger agricultural economic framework [1].

Groundnut (*Arachis hypogaea L.*) plays a vital role as a strategic crop to maintain food security and enhance agricultural sector strength. Groundnut farming occurs in multiple countries worldwide such as India, China, the United States, Senegal, Indonesia, Nigeria, Myanmar, Brazil, and Argentina. The cultivation area of groundnuts is quickly growing in Ghana, Mali, Somalia, Sudan, Thailand, Vietnam, Uganda, Mozambique and numerous regions throughout Africa. The majority of groundnut cultivation area (97%) and production output (94%) occur within developing nations. Groundnut production in Uzbekistan now functions as both a primary and secondary crop while achieving yields between 2.0 to 4.0 tons per hectare through the application of advanced technologies [2].

The increasing demand for food products requires us to improve oilseed crop yields and seed quality with a specific focus on groundnut (*Arachis hypogaea L.*). Groundnut is a vital oil- and protein-rich plant, with its seeds, pods, and stems serving various purposes: Groundnut products serve multiple purposes including direct human consumption and animal feed while naturally enriching the soil with nitrogen. The potassium found in groundnut shells helps to deliver vital organic and mineral substances into the soil. Groundnut shells play an industrial role in making construction materials like chipboard and soundproofing insulators. Groundnut serves as an oil crop but its high antioxidant and vitamin content benefits skin and hair health. Consuming groundnut milk benefits gastrointestinal health and the paste serves as a dense energy source that enhances physical endurance [3].

Groundnut seeds exhibit high oil content which is especially prominent in cultivated varieties developed for oil extraction. These plants have large light-green leaves and stand apart from types developed mainly to supply the food industry [4].

Groundnut kernels provide 26–28% high-quality protein that digests easily and also contain a variety of essential minerals and vitamins. The simple absorption of essential

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amino acids that facilitate protein synthesis boosts the nutritional benefits of groundnut. Groundnut seeds serve as a popular food choice because they can be eaten raw or roasted. The seeds average 18% carbohydrate content and a high mineral density with elements like potassium (K), calcium (Ca), magnesium (Mg), phosphorus (P), and sulfur (S). The oil delivers multiple essential vitamins including A, B and E and contains eight essential fatty acids that elevate its dietary value [5].

Groundnut serves as an essential dietary component because it provides both high-quality protein and abundant energy [6]. The twenty nutrients this substance contains play crucial roles in both growth and metabolism and feature nine essential amino acids that humans cannot produce independently [7].

Groundnut oil contains about 50% monounsaturated fatty acids while its polyunsaturated fatty acids make up 33% and saturated fatty acids account for 4%. The presence of bioactive compounds like phytosterols, flavonoids, and tocopherols accounts for the high nutritional value of groundnut oil. The oil provides high levels of vitamins A and B complex including vitamin B9 which supports nervous system function alongside DNA and RNA synthesis and brain health. The substance contains crucial mineral elements potassium (K) and magnesium (Mg) [8].

Groundnut (*Arachis hypogaea L.*) is part of the Fabaceae family which encompasses over 70 species. The only species that has undergone domestication and receives extensive agricultural use is *Arachis hypogaea L*. Cultivated groundnut is classified into two subspecies: The subspecies Hypogaea and Fastigiata split into botanical types like hypogaea hirsuta, fastigiata, vulgaris, peruviana, and aequatoriana. Historians trace the origins of groundnut to South America with a particular focus on Brazil. The plant requires warm temperatures while preferring moist conditions and depending on light exposure for growth during shorter daylight periods [2,6].

2. Materials and Methods

The research took place at the Durmon scientific station field experimental site which belongs to the Institute of genetics and experimental plant biology of the Academy of Sciences of Uzbekistan. The research was conducted at the institute's Laboratory of genetics, breeding and seed production of leguminous, oilseed and medicinal plants utilizing ten international accessions. The evaluated accessions included: The accessions studied included Uganda Erect SB 33 from Uganda, Virginia Improved from Israel, Zac Trang from Vietnam, Philippin Pink and Namuno from Portugal, R 30 from Israel, Var Cuba from Japan, Dessertny from Russia, PL 1 and Hippagi 2-20 from India together with a local variety called Polvon. Researchers chose these seed samples to analyze and contrast their total protein and oil levels.

Researchers employed the Kjeldahl method to measure total protein content. This technique calculates protein levels by measuring nitrogen as an indirect gauge. The procedure uses concentrated sulfuric acid to hydrolyze the organic matter in the sample while transforming protein amino groups into ammonium sulfate salts.

Nitrogenous organic matter + $H_2SO_4 \rightarrow (NH_4)_2SO_4$ + $CO_2\uparrow + H_2O$

After hydrolysis is complete, sodium hydroxide is added to convert the resulting ammonium sulfate into ammonia.

$$(NH_4)_2SO_4 + 2NaOH \longrightarrow Na_2SO_4 + 2NH_4OH \begin{matrix} \nearrow 2H_2O \\ \searrow 2NH_3 \end{matrix}$$

Through a neutralization process sulfuric acid solution absorbs the ammonia or ammonium hydroxide which has been released. An alkali solution titrates the remaining acid. The nitrogen content determination comes from the calculated quantity of ammonia. A precisely weighed portion of the finely ground seed sample is transferred into a test tube for analysis while maintaining a measurement error below 0.1%. The quantitative processing of the sample takes place in a Kjeldahl flask. The established protocol [9] guided all subsequent steps of the procedure.

The analysis of the sample's nitrogen mass fraction (X) involved a standard formula which utilized the volume of alkali necessary to titrate the excess acid following ammonia absorption from diluted sulfuric acid.

$$X = \frac{(\mathbf{V_1} - \mathbf{V_0}) * \mathbf{K} * \mathbf{0.0014}}{m} * 100\%$$

The volume V_0 represents the amount (in milliliters) of 0.1 mol/L sodium hydroxide solution required for titration of the remaining 0.1 mol/L sulfuric acid solution during sample analysis.

The oil content determination followed the standard requirements set by the Interstate Council for Standardization, Metrology, and Certification (ISJ) using the methodology described in "Oils and Fats of Animal and Plant Origin. Sampling" (GOST ISO 5555—2016). Sampling" [10].

3. Results and Discussion

The scientific study assessed the total protein levels present within the seeds of both foreign and local groundnut (*Arachis hypogaea L.*) accessions.

The collection displays extensive differences among accessions regarding their total protein content according to the data. Foreign and local groundnut accessions show substantial differences in seed protein content which demonstrates the genetic variation present in the germplasm under study.

The total protein content showed significant variation across the examined groundnut accessions with levels between 19.50% and 30.38%. The groundnut accession PL#1 from India showed the highest protein content at 30.38% followed by Var Cuba from Japan at 28.13% and Virginia Improved from Israel at 26.88%. The accessions R 30 (Israel) showed 22.44%, Polvon (Uzbekistan) showed 20.75%, and Philippine Pink (Portugal) showed 19.50% protein values which are detailed in Tables 1 and 2.

Table 1. Total protein content in seeds of foreign and local accessions of the groundnut (*Arachis hypogaea L.*) collection

№	Accession name	Total protein content (%)	Difference from control Polvon, ±%
1.	Uganda Erect Sb 33 (Uganda)	22,63%	+1,88
2.	Virginia Improved (Israel)	26,88%	**+6,13**
3.	Zac Trang (Vietnam)	25,13%	**+4,38**
4.	Philippine Pink (Portugal)	19,50%	-1,25
5.	Namuno (Portugal)	26,06%	**+5,31**
6.	R 30 (Israel)	22,44%	+1,69
7.	Var Cuba (Japan)	28,13%	**+7,38**
8.	Dessertny (Russia)	22,56%	**+1,81**
9.	PL#1 (India)	30,38%	**9,63**
10.	Hippagi 2-20 (India)	23,44%	**2,69**
11.	Nazorat Polvon (Uzbekistan)	20,75%	-
12.	***LSD$_{0.05}$**	**2.54%**	-

Note: *$LSD_{0.05}$ (Least Significant Difference at 5% level)* is a statistical criterion used to determine the minimum difference required between variants to consider the variation statistically significant. If the observed difference between variants exceeds $LSD_{0.05}$ or $LSD_{0.01}$, it is regarded as a true difference.

Table 2. Relative total protein content of foreign and local (*Arachis hypogaea L.*) accessions compared to the control variety

Accession name	Protein content (%)	Difference from control (d)	Squared difference (d^2)	Relative index to control (%)
Uganda Erect Sb 33	22,63	1,88	3,5344	109,1
Virginia Improved	26,88	**6,13**	37,5769	129,5
Zac Trang	25,13	**4,38**	19,1844	121,1
Philippine Pink	19,5	-1,25	1,5625	94,0
Namuno	26,06	**5,31**	28,1961	125,6
R 30	22,44	1,69	2,8561	108,1
Var Cuba	28,13	**7,38**	54,4644	135,6
Dessertny	22,56	**1,81**	3,2761	108,7
PL#1	30,38	**9,63**	92,7369	146,4
Hippagi 2-20	23,44	**2,69**	7,2361	113,0
Polvon Nazorat	20,75	0	0	**100,0**
Total	**267,9**	**39,65**	**250,62**	
Average	**24,35**	**3,61**		

The groundnut accessions demonstrate four distinct categories when classified by their protein content. The lowest protein content group contains varieties from 19.50% to 20.75% protein including Philippine Pink (Portugal) and Polvon (Uzbekistan). Groundnut breeders find the Polvon variety useful for breeding because its large seed size offers potential for improved yield development.

The standard deviation of the differences (S_d) was calculated using the following formula:

$$S_d = \sqrt{\frac{\sum d^2 - (\sum d)^2 : n}{n(n-1)}} = \frac{250.62 - 39.65^2 : 4}{11(11-1)} = 1.139$$

*The calculated **t-value** is:*

$$t = \frac{d}{S_d} = \frac{3.61}{1.139} = 3.16$$

Critical values for statistical significance were as follows:

- **t_{0.05} = 2.23**
- **t_{0.01} = 3.17**

*The **Least Significant Differences (LSD)** at the 5% and 1% levels were calculated as:*

- ***LSD_{0.05} = t_{0.05}** × **S_d = 2.23** × **1.139 = 2.54%***
- ***LSD_{0.01} = t_{0.01}** × **S_d = 3.17** × **1.139 = 3.61%***

*These results indicate that any difference in protein content between accessions exceeding **2.54%** is statistically significant at the **p < 0.05** level, and any difference exceeding **3.61%** is significant at the **p < 0.01** level.*

Accessions within the second group show seed protein content between 22.44% and 23.44% and include R 30

(Israel), Dessertny (Russia), Uganda Erect Sb 33 (Uganda), and Hippagi 2-20 (India). These accessions display their distinguishing growth pattern through spreading near the ground surface. Research shows that groundnut plants with this type of growth pattern develop more underground pods which results in higher yields. The demonstrated characteristics of these accessions make them ideal candidates for selection programs that target high-yield groundnut cultivar development.

Analysis of the protein content data allows accessions to be sorted into third and fourth protein-rich categories which include Zac Trang from Vietnam at 25.13%, Namuno from Portugal at 26.06%, Virginia Improved from Israel at 26.88%, Var Cuba from Japan at 28.13%, and PL#1 from India at 30.38%. Protein levels in these accessions far exceeded the values documented in previous research studies. These varieties demonstrate significant potential for use in breeding programs aimed at producing groundnut strains with better grain quality and higher protein content.

Groundnut serves as an important crop for both oil production and food consumption. The seeds of this plant store easily digestible oil which manufacturers refine to produce high-quality edible peanut oil. Peanut oil carries a flavor profile that resembles olive oil. The product finds extensive application in premium vegetable preserves and margarine manufacturing while also serving as a fundamental component in the confectionery and perfumery sectors. Groundnut serves as the main ingredient for over 60 confectionery products such as cooking oil, chocolate, cookies, halva, coffee, candies, ice cream among others.

Groundnut kernels offer essential nutrients because they contain high-quality digestible protein levels between 26–28% along with vitamins and minerals. Nutrient-rich stems of the plant match alfalfa in nutritional value. The leftover press cake from oil extraction retains 48% protein content and 8% residual oil which establishes its importance as livestock and poultry feed. The production of bread now often incorporates groundnut cake to improve both taste and nutrient content. Cookies and bread made with groundnut cake additives maintain premium status because their high demand persists even though they come at a higher market price [11].

The research team pursued additional studies to evaluate the oil content in seeds from foreign accessions of Groundnut (*Arachis hypogaea L.*) The research focused on measuring and contrasting the oil content between local and foreign groundnut accessions. Figure 1 below illustrates the findings from this analysis.

Foreign groundnut (*Arachis hypogaea L.*) accessions demonstrate greater oil content in their seeds when compared to the local variety Polvon. The study showed that oil content levels in the collection varied between 36.1% and 49.2%. Two distinct groups emerge when the accessions are sorted according to their oil content. Accessions with low oil content between 36.1% to 40.3% make up the first class including PL#1 from India, Polvon from Uzbekistan, Var Cuba from Japan, Dessertny from Russia, and Namuno from Portugal. R-30 from Israel, Hippagi 2-20 from India, Virginia Improved from Israel, Zac Trang from Vietnam, and Philippine Pink from Portugal represent the second class which features high oil content (42.6% to 49.2%). The accessions from Israel, Vietnam, and Portugal recorded the highest values among all. Research identifies high-oil accessions as essential resources for developing new groundnut varieties that produce more oil. Current scientific studies continue to progress actively within this area of research.

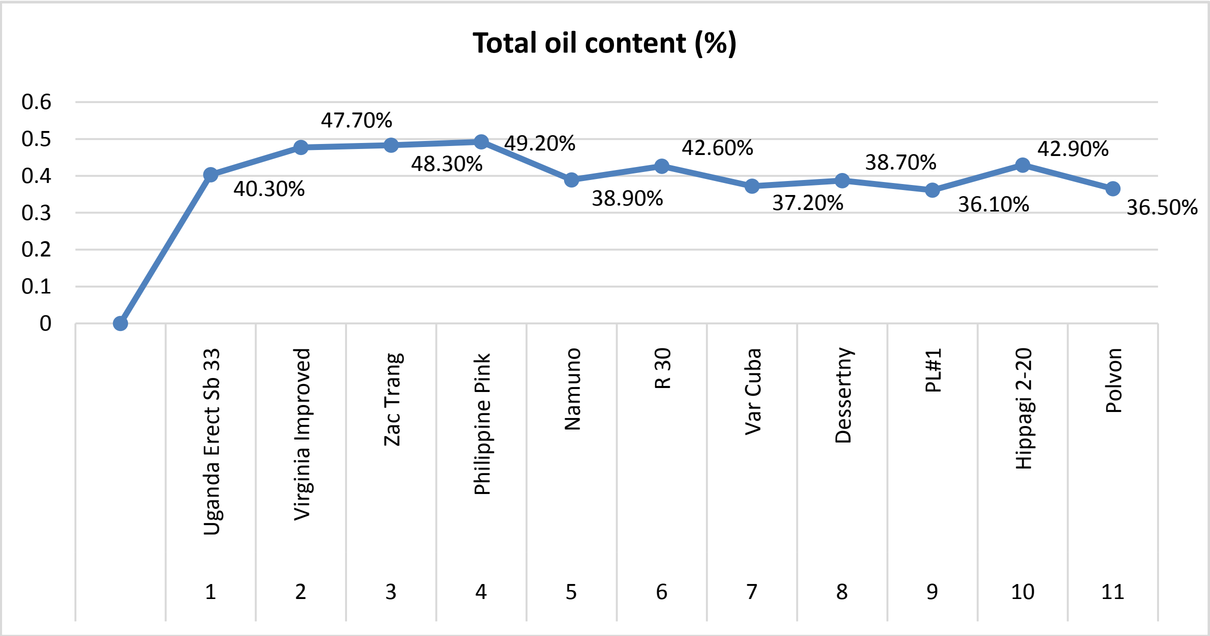

Figure 1. Oil content (%) in seeds of foreign and local accessions of groundnut (*Arachis hypogaea L.*)

4. Conclusions

All groundnut collection accessions except the **Portuguese Philippine Pink** demonstrated higher total protein content than the control variety **Polvon** according to statistical analysis. The accessions Virginia Improved, Var Cuba and PL#1 demonstrated protein levels that were between 6.13% and 9.63% superior to the control. The study established significant differences in protein levels between local and foreign *Arachis hypogaea L*. accessions and identified multiple high-protein accessions from these findings. The oil content analysis showed that the highest oil percentages were found in accessions from Israel named Virginia Improved along with Zac Trang from Vietnam and Philippine Pink from Portugal. The discovered accessions serve as crucial genetic resources for developing new groundnut breeds with improved protein and oil levels. Persistent investigation along this route offers significant potential to create crop cultivars that produce more yield and have better nutritional properties. The groundnut accessions PL#1 from India, Hippagi 2-20 from India, Var Cuba from Japan, Virginia Improved from Israel, Zac Trang from Vietnam and Philippine Pink from Portugal demonstrate excellent protein and oil content which makes them prime candidates for breeding programs focused on enhancing yield and nutritional quality in groundnut crops.

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