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CHARACTERIZATION OF GROUNDNUT (*Arachis hypogaea* L.) GENOTYPES THROUGH SEED BIOCHEMICAL ATTRIBUTES

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ABSTRACT

The investigation aimed to evaluate biochemical variability parameters among groundnut genotypes to assess their phylogenetic relationships. A total of twenty genotypes were selected for detailed biochemical characterization during 2024-25. The results of biochemical analysis indicated the existence of considerable variability among the studied genotypes for all traits, suggesting a broad genetic base. Moisture content showed substantial variation, ranging from 5.90% to 8.97%, with the highest value recorded in TCGS-894 and the lowest in JB-1625. Protein content varied moderately between 20.41% and 23.12%, with GG-40 exhibiting the minimum and JB-1677 the maximum values. Oil content, an economically important trait in groundnut, ranged from 47.94% in JL-1085 to 52.42% in JB-1632. Total carbohydrate content also differed significantly among genotypes, varying from 16.00% (J-118) to 21.43% (JB-1642). In addition, total soluble sugars ranged from 8.35% to 11.03%, while reducing sugars and non-reducing sugars varied from 1.97% to 2.94% and 5.87% to 8.65%, respectively. The observed biochemical variability reflects underlying genetic differences and highlights the potential of these genotypes for further breeding and improvement programs. Similarity analysis grouped the groundnut genotypes into two major clusters, Cluster I and Cluster II, each with further sub-grouping. Cluster I was divided into Groups A (A1: JB-1632, GG-39; A2: DH-256, GG-40) and B (B1: JB-1625; B2: JB-1642). Cluster II was subdivided into Group A (A1: JB-1628, J-120, JB-1647, J-116; A2: TCGS-1157, JL-1085, K-1812, JB-1613, TCGS-894, JB-1677, KDG-160, JB-1621, VRI-11) and Group B (J-118). Overall, the clustering pattern reflects the genetic relationships and diversity among the studied genotypes.

Keywords **:** Groundnut, Biochemical, Protein, Oil, Clustering.

Introduction

Groundnut (*Arachis hypogaea* L.) is an important oilseed crop widely cultivated in tropical and subtropical regions due to its high nutritional and economic value. It belongs to the family *Leguminosae* and is commonly known by various names such as peanut, earthnut and monkey nut. The crop is valued for its rich composition of proteins, healthy fats, carbohydrates, vitamins and essential minerals, making it a key component of human diet as well as animal feed. Owing to its versatility and wide range of uses, groundnut is often referred to as the "King of Oilseeds" and the "Poor Man's Cashew" (Kalyani and Sasidharan, 2021).

Taxonomically, groundnut is divided into two subspecies, *Arachis hypogaea* ssp. *hypogaea* and *Arachis hypogaea* ssp. *fastigiata*, based on morphological characteristics. Cytogenetically, it is an allotetraploid species (2n = 4x = 40) with an AABB genome, believed to have originated from hybridization between two diploid ancestors, *Arachis duranensis* (A genome) and *Arachis ipaensis* (B genome), followed by chromosome doubling. The genus *Arachis* comprises around 80 species, most of which are diploid. Based on growth habit, groundnut

varieties are classified into spreading, semi-spreading and bunch types, reflecting their adaptability and agronomic diversity.

India is the second-largest producer of groundnut after China and ranks first among oilseed crops in the country. Major producing countries include China, India, USA, Senegal and Brazil. In India, Gujarat, Andhra Pradesh, Karnataka, Maharashtra, Rajasthan and Tamil Nadu together contribute about 90% of the total cultivation area. During 2024-25, groundnut was grown over 48.80 lakh hectares with a production of 103.60 lakh tonnes and productivity of 2122 kg/ha (Anon., 2024a).

Nutritionally, groundnut seeds are rich in protein, oil and dietary fibre, along with essential vitamins such as B-complex and minerals like calcium, phosphorus, potassium and iron. Groundnut oil is widely used for cooking and industrial purposes, while kernels are consumed in various forms including roasted nuts, peanut butter and confectionery products. The oilcake serves as a valuable livestock feed and organic manure. The fatty acid composition of groundnut oil, particularly the ratio of oleic to linoleic acid, is an important determinant of oil quality and human health benefits.

Biochemical profiling of groundnut is essential for evaluating its nutritional quality, varietal improvement and suitability for different industrial and other food applications. The estimation of total protein helps in assessing its dietary significance, while oil content is crucial for determining its commercial value. Total carbohydrate content, including reducing and non-reducing sugars, reflects the energy-yielding potential of the seeds. Moisture content plays a key role in storage and post-harvest shelf life, whereas total soluble sugars contribute to taste and processing quality (Dwivedi *et al.*, 1996).

Materials and Methods

The investigation was carried out with 20 groundnut (*Arachis hypogaea* L.) varieties, which were procured from ICAR-Indian Institute of Groundnut Research, Junagadh and Main Oilseeds Research Station, Junagadh Agricultural University, Junagadh. The biochemical parameters of mature groundnut seeds were analysed using a Completely Randomized Design (CRD) with three replications.

Moisture content was determined following the standard procedure of the Association of Official Analytical Chemists (AOAC, 2005a). Five grams of seed sample were weighed in Petri dishes and dried in a hot air oven at 90°C for 5 hours. After drying, the samples were cooled in a desiccator and weighed again. The percentage moisture content was calculated based on the difference between fresh and oven-dried sample weights. Total protein content was estimated by the micro-Kjeldahl method as described by AOAC (2005b). Finely ground seed samples were digested with concentrated sulphuric acid in the presence of a copper digestion mixture, followed by distillation of liberated ammonia into boric acid solution. The distillate was titrated against standard sulphuric acid and nitrogen percentage was calculated, which was further multiplied by a conversion factor of 6.25 to obtain crude protein content.

Oil content of groundnut seeds was determined by Soxhlet extraction method using hexane as solvent according to AOAC (2005a). Approximately 5 g of dried and crushed seed material was placed in extraction thimbles and refluxed for 4 hours. After extraction, the solvent was evaporated and the extracted oil was weighed. Oil percentage was calculated on dry weight basis. Total carbohydrate content was estimated by the Anthrone method (Hedge and Hofreiter, 1962). Hydrolyzed seed extracts were reacted with freshly prepared anthrone reagent under acidic conditions, producing a green-colored complex. The absorbance was recorded at 630 nm using a spectrophotometer, and carbohydrate content was calculated using a standard glucose curve.

Total soluble sugar content was estimated using the Phenol-Sulphuric Acid method described by Dubois *et al.* (1956). Ground seed samples were extracted with 80% methanol, and aliquots of the extract were reacted with phenol and concentrated sulphuric acid. The developed color intensity was measured at 490 nm spectrophotometrically and sugar concentration was calculated using glucose standards. Reducing sugar content was analyzed by the Nelson-Somogyi method (Somogyi, 1952). Methanolic extracts of seed samples were treated with alkaline copper reagent followed by arsenomolybdate reagent, and the absorbance was measured at 620 nm. The reducing sugar percentage was determined using the corresponding standard curve. Non-reducing sugar content was calculated by subtracting reducing sugar content from total soluble sugar content and expressed as percentage.

All biochemical estimations were carried out in triplicate to ensure accuracy and reproducibility of results. Standard analytical procedures and spectrophotometric methods were followed throughout the study for quantitative estimation of biochemical constituents in groundnut genotypes.

Results and Discussion

The biochemical characterization of groundnut genotypes revealed substantial variability among the tested materials. The findings generated from the present study are discussed under the following headings.

Moisture Content (%)

The moisture content of the twenty groundnut genotypes exhibited significant variation, indicating the presence of considerable diversity among the genotypes for this trait. The moisture content ranged from 5.90 to 8.97 % with an overall mean value of 7.27 % (Table 1; Fig. 1). Among the studied genotypes, TCGS-894 recorded the highest moisture content (8.97%), whereas the lowest moisture content (5.90%) was observed in genotype JB-1625. The variation in moisture content among the genotypes may be attributed to genetic differences. The present findings are in agreement with the results reported by Ayoola *et al.* (2012), who observed 7.40 % moisture content in groundnut seeds, which was comparable with the present investigation. Similarly, Alhassan *et al.* (2017) reported moisture content ranging from 4.9 to 6.8 % among different groundnut genotypes. However, Ingale and Shrivastav (2011) recorded comparatively lower moisture content (5.53%) in groundnut seeds than that observed in the present study.

Total Protein (%)

The total protein content among the twenty groundnut genotypes differed significantly, indicating substantial variability for this nutritional parameter. Protein content ranged from 20.41 to 23.12 % with a mean value of 21.90 % (Table 1; Fig. 2). The genotype JB-1677 recorded the highest protein content (23.12%), which was statistically at par with KDG-160, J-118, and TCGS-894. In contrast, the lowest protein content (20.41%) was observed in genotype GG-40, which was at par with JB-1625, JB-1628, JB-1647, and J-120. The results obtained in the present investigation are partially supported by earlier findings. Asibuo *et al.* (2008) reported protein contents of 25.69 and 30.53 % in subspecies *fastigiata* and Spanish market types, respectively. Campos *et al.* (2009) observed protein content ranging from 23.5 ± 1.2 to 26.6 ± 0.0 % among different groundnut varieties. Ingale and Shrivastav (2011) reported 25.20 % protein content in JL-24, which was comparatively higher than the present findings. Kulkarni *et al.* (2015) also observed crude protein content ranging from 28 to 38 % with a mean of 35.00 %.

Table 1: Data on biochemical parameters of 20 genotypes of groundnut

Sr. No.	Genotypes	Moisture content (%)	Total protein content(%)	Oil content (%)	Total carbohydrate (%)	Total soluble sugar (%)	Reducing sugar (%)	Non-reducing sugar (%)
1.	VRI-11	6.84	22.18	49.47	18.59	9.23	2.55	6.69
2.	DH-256	7.54	21.03	51.72	19.44	9.66	2.16	7.49
3.	K-1812	7.38	22.30	50.49	18.77	**11.03**	2.66	8.37
4.	TCGS-894	**8.97**	22.75	49.33	17.68	9.05	**1.97**	7.07
5.	TCGS-1157	7.08	21.61	48.30	17.26	10.15	2.42	7.72
6.	KDG-160	6.35	23.09	49.35	18.29	9.55	1.99	7.56
7.	JL-1085	7.64	21.99	**47.94**	18.45	10.68	2.03	**8.65**
8.	JB-1613	7.39	22.33	50.72	16.94	10.28	2.76	7.52
9.	JB-1621	7.80	21.94	50.34	18.68	9.52	2.31	7.21
10.	JB-1625	**5.90**	20.76	51.47	21.04	10.11	2.12	7.99
11.	JB-1628	6.62	20.70	48.47	17.54	8.94	1.98	6.96
12.	JB-1632	8.09	22.33	**52.42**	17.40	9.18	2.29	6.89
13.	JB-1642	7.13	21.10	51.40	**21.43**	10.07	2.19	7.88
14.	JB-1647	8.17	20.85	49.20	19.58	9.11	2.07	7.04
15.	JB-1677	7.75	**23.12**	48.55	17.62	9.90	2.89	7.02
16.	J-116	6.93	22.54	48.52	20.48	9.41	2.50	6.92
17.	J-118	6.90	22.91	48.48	**16.00**	8.99	2.32	6.67
18.	J-120	6.36	20.73	49.36	18.99	9.00	**2.94**	6.06
19.	GG 39	7.28	21.85	51.08	18.49	**8.35**	2.49	**5.87**
20.	GG 40	7.22	**20.41**	52.16	18.31	9.67	2.43	7.24
Mean		7.27	21.83	49.94	18.55	9.59	2.35	7.24
Minimum		5.90	20.41	47.94	16.00	8.35	1.97	5.87
Maximum		8.97	23.12	52.42	21.43	11.03	2.94	8.65
S.Em ±		**0.10**	**0.23**	**1.02**	**0.29**	**0.12**	**0.02**	**0.12**
CD @ 5%		**0.28**	**0.67**	**2.93**	**0.82**	**0.34**	**0.06**	**0.33**
CV (%)		**2.36**	**1.85**	**3.55**	**2.69**	**2.14**	**1.56**	**2.80**

Oil Content (%)

Significant differences were observed among the twenty groundnut genotypes for oil content, demonstrating substantial variability among the studied materials. The oil content ranged from 47.94 to 52.42 % (Table 1; Fig. 3). The highest oil content was recorded in genotype JB-1632 (52.42%), followed by GG-40 (52.16%), whereas the lowest oil content (47.94%) was observed in genotype JL-1085. The variability in oil content among genotypes suggests the influence of genetic factors governing lipid biosynthesis and accumulation in groundnut seeds. The present findings are in close agreement with the reports of Asibuo *et al.* (2008), who found higher oil content in *Virginia* cultivars (49.7%) compared to *Spanish* and *Valencia* market types (47.3%). Pardeshi (2019) also reported oil extraction yields ranging from 39.45 to 41.48 % among different groundnut varieties.

Total Carbohydrate (%)

The analysis of total carbohydrate content revealed significant variation among the twenty groundnut genotypes. The carbohydrate content ranged from 16.00 to 21.43 % with a mean value of 18.55 % (Table 1; Fig. 4). The maximum carbohydrate content (21.43%) was recorded in genotype JB-1642, which was statistically at par with JB-1625, whereas the minimum carbohydrate content (16.00%) was observed in genotype J-118. The present findings are comparable with earlier reports. Ingale and Shrivastav (2011) recorded 21.26 % carbohydrate content in the JL-24 variety. Ayoola *et al.* (2012) reported 17.41 % carbohydrate content in groundnut seeds, which was similar to the present investigation. Likewise, Alhassan *et al.* (2017) reported carbohydrate content ranging from 21 to 37 % among different groundnut accessions.

Total Soluble Sugar (%)

The total soluble sugar content among the twenty groundnut genotypes showed significant differences. The total soluble sugar content ranged from 8.35 to 11.03 % (Table 1; Fig. 5). Genotype K-1812 recorded the highest total soluble sugar content (11.03%), whereas the lowest value (8.35%) was observed in genotype GG-39. The findings of the present study are supported by the results of Shad *et al.* (2009), who reported total soluble sugar content ranging from 7.64 ± 0.06 to 8.30 ± 0.12 %. Kulkarni *et al.* (2015) observed total sugar content ranging from 6.10 to 9.00 % with a mean value of 7.53 %, which was comparatively lower than the values recorded in the present investigation.

Reducing Sugar (%)

Significant variation was observed among the groundnut genotypes for reducing sugar content. The reducing sugar content ranged from 1.97 to 2.94 % with an average value of 2.35 % (Table 1; Fig. 6). The highest reducing sugar content was recorded in genotype J-120 (3.05%), which was statistically at par with genotype JB-1677 (2.89%). On the other hand, genotype TCGS-894 exhibited the lowest reducing sugar content (1.97%), which was at par with JB-1628 and KDG-160. The results obtained are in agreement with the findings of Shad *et al.* (2009), who reported reducing sugar content ranging from 1.64 ± 0.11 to 1.97 ± 0.08 %. Similarly, Kulkarni *et al.* (2015) observed reducing sugar content ranging from 1.02 to 1.60 %, which supports the present findings.

Non-reducing Sugar (%)

Significant variation was observed among the twenty groundnut genotypes for non-reducing sugar content. The non-reducing sugar content ranged from 5.87 to 8.65 % (Table 1; Fig. 7). Among the evaluated genotypes, JL-1085 recorded the highest non-reducing sugar content (8.65%), which was statistically at par with genotype K-1812 (8.37%). In contrast, the lowest non-reducing sugar content (5.87%) was observed in genotype GG-39, followed by genotype J-120 (6.06%). The present findings are in close agreement with the results reported by Shad *et al.* (2009), who recorded non-reducing sugar content ranging from 5.67 ± 0.21 to 6.67 ± 0.24 %. Similarly, Kulkarni *et al.* (2015) observed non-reducing sugar content varying from 4.88 to 7.73 % among different groundnut genotypes.

Cluster analysis based on biochemical parameters was carried out to assess the genetic similarity and diversity among the groundnut genotypes. Biochemical characterization provides important information on nutritional and metabolic variability, while similarity analysis helps in understanding genetic relationships and identifying diverse genotypes for breeding programmes. Therefore, the present study was undertaken to evaluate the diversity pattern among twenty groundnut genotypes based on biochemical traits.

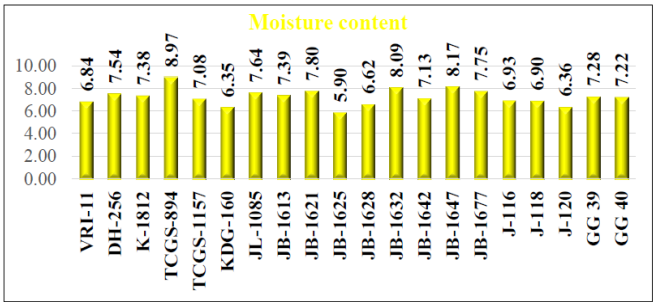

Fig. 1 : Comparative moisture content among 20 genotypes of groundnut

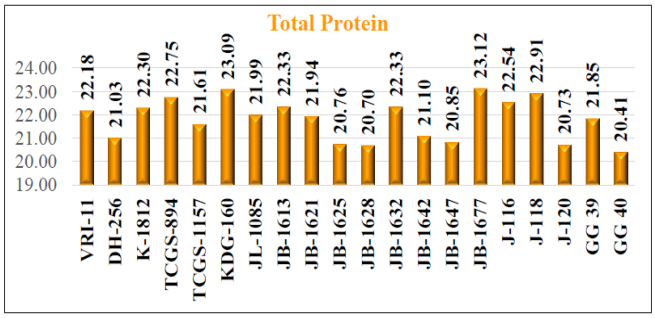

Fig. 2 : Comparative total protein among 20 genotypes of groundnut

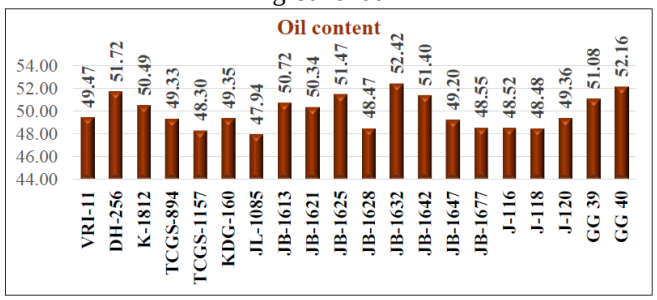

Fig. 3 : Comparative oil content among 20 genotypes of groundnut

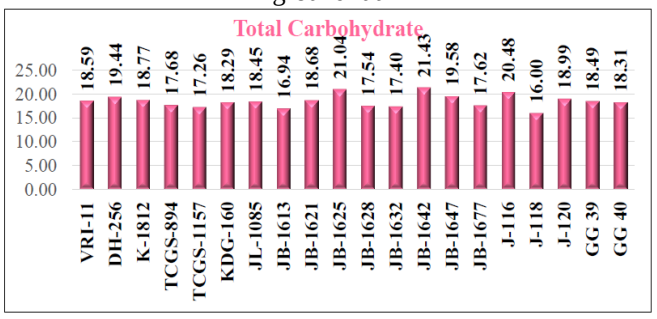

Fig. 4 : Comparative total carbohydrate among 20 genotypes of groundnut

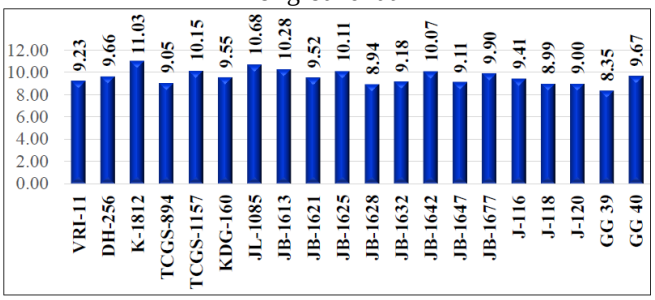

Fig. 5 : Comparative total soluble sugar among 20 genotypes of groundnut

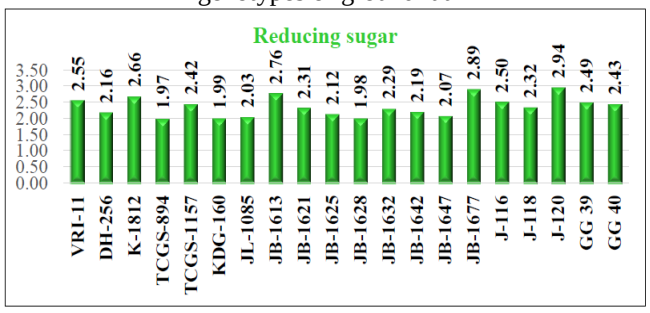

Fig. 6: Comparative reducing sugar among 20 genotypes of groundnut

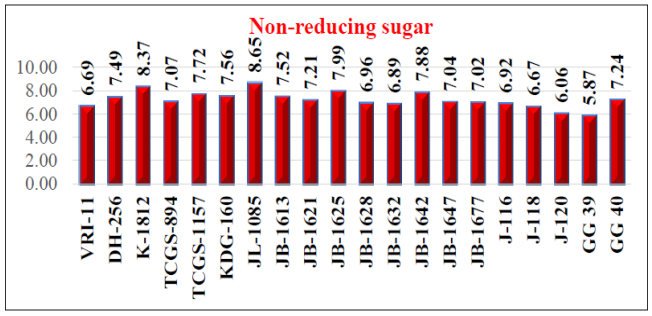

Fig. 7 : Comparative non-reducing sugar among 20 genotypes of groundnut

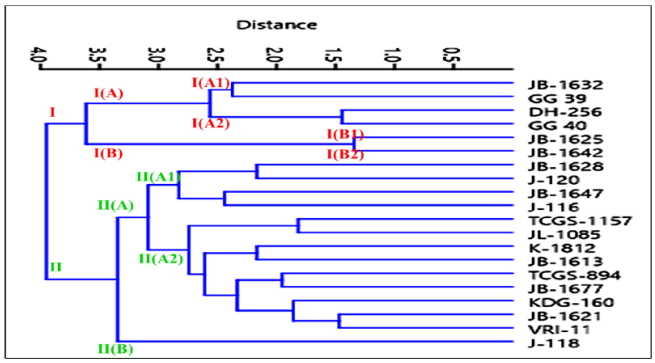

Fig. 8: Dendrogram of Biochemical data analysis of 20 groundnut genotypes

Similarity analysis of groundnut genotypes grouped them into two main clusters, Cluster I and Cluster II. Cluster I was further divided into two groups, A and B. Group A was subdivided into A1 and A2, where A1 included closely related genotypes such as JB-1632 and GG-39, while A2 comprised genotypes like DH-256 and GG-40. Group B was divided into B1 and B2, B1 including genotype JB-1625 and B2 JB-1642. Cluster II was further grouped into A and B, where Group A was subdivided into A1 and A2, A1 including genotypes such as JB-1628, J-120, JB-1647 and J-116 while A2 including genotypes TCGS-1157, JL-1085, K-1812, JB-1613, TCGS-894, JB-1677, KDG-160, JB-1621 and VRI-11. Group B included only one genotype J-118. Overall, the clustering pattern clearly indicates the genetic similarity and diversity among the groundnut genotypes (Fig. 8).

Conclusion

Based on the biochemical evaluation and cluster analysis of 20 groundnut genotypes, the study clearly demonstrated the presence of significant genetic variability among the tested materials for all the biochemical traits analyzed. Among the studied genotypes, JB-1677 was identified as a superior genotype for protein content, whereas JB-1632 and GG-40 were promising for high oil content. JB-1642 and JB-1625 exhibited higher carbohydrate content, while K-1812 and JL-1085 showed superior sugar content. TCGS-894 recorded the highest moisture

content, whereas JB-1625 showed the lowest moisture percentage, which may be advantageous for storage stability. These differences among genotypes indicate the influence of genetic factors on biochemical composition and suggest opportunities for selecting desirable parental lines in breeding programmes.

The cluster analysis grouped the twenty genotypes into two major clusters with several sub-clusters, reflecting varying degrees of genetic similarity and divergence among the genotypes. Genotypes grouped in separate clusters were genetically more diverse, indicating their potential usefulness in hybridization programmes to obtain superior recombinants and broaden the genetic base of cultivated groundnut. The clustering pattern also revealed that biochemical traits are effective tools for assessing genetic diversity and phylogenetic relationships among groundnut genotypes.

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