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RESEARCH ARTICLE

Effect of storage on biochemical changes in groundnut (*Arachis hypogaea* L.) during *Kharif* and summer

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SUMMARY

The biochemical changes during storage were studied in a set of fifty five spreading and semi spreading types of groundnut genotypes (*Arachis hypogea* L.). The studies indicated the effect of season and storage periods on these parameters. The genotypes behaved differently in different seasons and under different storage periods. Under storage both oil and protein showed marked reduction in their content. The variations were clearly observed in SDS PAGE analysis of protein and esterase isozymes. In the protein analysis 769 bands were observed in fresh *Kharif* 2010 seed whereas 696 bands were observed in stored summer 2010 seeds. The PIC (Polymorphism information content) of both summer and *Kharif* 2010 seed was found to be 0.94. Esterase analysis showed 181 bands in the fresh *Kharif* 2010 seed while, 101 bands were recorded in stored summer 2010 seed. The lower PIC value observed in *Kharif* and summer seasons for esterase isozymes as compared to proteins indicated that genotypic characterisation using the latter may be more reliable than former.

Key Words : Banding patterns, Isozymes, Oil content, Protein content, Storage period

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Groundnut (*Arachis hypogaea* L.) is an annual legume grown primarily for high quality edible oil (36 – 54% on dry matter basis) and easily digestible protein (12 – 36%) in its seeds. It is cultivated worldwide a world production of 35.9 million tonnes at an area of 25.2 million ha. In India, it is spread over an area of 6.6 million ha with production of 5.9 million tonnes (FAO, 2006). Among several seed quality attributes, the storage potential of seed plays an important role in meeting the demand for commercial crop production programme. The biochemical parameters such as oil content and protein content found to be affected by seasonal variations.

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MATERIAL AND METHODS

Fifty five genotypes of groundnut were sown in un-replicated trial during summer 2010 at Dept. of Agril. Botany AAU, Anand. A part of the groundnut produce harvested from summer 2010 was stored in cloth bags for laboratory studies. The rest of the groundnut produce was sown in *Kharif* 2010 in a Complete Randomized Block Design (RCBD) in three replications. The stored seed from summer 2010 was observed for biochemical parameters both in fresh seed as well as after three months of storage. The same sets of observations were studied in fresh *Kharif* 2010 seed also.

SDS PAGE :

The electrophoresis was carried out on vertical SDS-PAGE (12%) at 60 mA for 2 hours. The total protein content in the samples was estimated by Micro Kjeldal method in which the total nitrogen content in the sample was multiplied with 6.25 to obtain the crude protein value in the sample. (Sadasivam and Manickam,1996).

Table 1: Oil and protein percentage in fresh seed of *Kharif* 2010 and after six months of storage of summer 2010 seed

Sr. No.	Genotype	Fresh seed of *Kharif*- 2010		Summer- 2010 seed stored for six months	
		Oil %	Protein %	Oil %	Protein %
1.	GG-2	46.18	29.26	31.47	26.00
2.	GG-3	41.70	24.79	36.69	21.52
3.	GG-4	41.69	28.12	31.28	24.33
4.	GG-5	48.39	27.43	34.49	23.87
5.	GG-7	42.48	28.77	33.56	25.95
6.	GAUG-10	45.68	28.39	25.53	24.47
7.	JL-24	43.79	24.96	35.27	18.43
8.	TG-37	43.38	21.73	35.26	19.75
9.	GG-12	49.19	18.93	34.94	15.07
10.	GG-13	44.10	20.17	31.35	15.28
11.	GG-14	45.74	22.13	27.42	16.60
12.	GG-15	44.06	20.47	37.37	14.00
13.	GG-16	41.94	19.19	37.05	16.17
14.	GG-20	46.78	22.62	36.51	18.67
15.	BAV-13	43.41	20.59	35.61	17.75
16.	KADIRI-3	50.18	19.85	32.40	14.06
17.	ICGS-1179	43.15	23.99	28.24	17.27
18.	ICGS-1703	49.05	20.12	32.68	13.67
19.	ICGS-4296	44.89	20.80	33.33	17.20
20.	ICGS-4849	48.57	21.64	37.60	16.01
21.	NRCG-6563	43.86	23.56	34.94	11.28
22.	NRCG--6663	50.31	21.20	33.55	15.33
23.	NRCG--6682	49.38	24.19	34.34	21.17
24.	NRCG--6705	44.65	23.37	33.49	20.38
25.	NRCG-6707	43.77	24.95	31.04	21.22
26.	NRCG-9000	47.56	21.11	32.42	16.33
27.	NRCG-9130	47.20	21.68	36.33	12.35
28.	NRCG-9185	49.15	28.05	32.25	24.00
29.	NRCG-9231	50.16	24.80	35.30	21.17
30.	NRCG-9747	47.18	22.35	34.53	17.73
31.	NRCG-9949	45.68	28.39	32.34	24.45
32.	ICGS-11615	50.21	24.27	32.01	22.33
33.	ICGS-13052	46.18	21.65	31.64	17.50
34.	ICGS-13128	45.91	23.11	35.77	16.30
35.	ICGS-13033	44.33	23.63	37.58	19.25
36.	ICGS-36	49.79	21.75	32.93	18.33
37.	ICGS-156	45.78	23.38	32.82	14.10
38.	ICGS-221	42.46	21.71	33.71	19.67
39.	ICGS-297	45.47	25.69	32.25	20.07
40.	ICGS-405	42.38	20.88	31.63	17.17
41.	ICGS-799	43.89	27.25	34.73	24.00
42.	ICGS-2738	47.85	22.31	32.40	15.87
43.	ICGS-4729	47.08	21.46	33.63	12.15
44.	ICGS-4750	49.50	22.59	28.67	20.52
45.	ICGS-5016	47.49	25.83	32.16	21.83
46.	ICGS-5236	46.37	21.26	33.85	19.33
47.	ICGS-7827	46.51	21.45	33.19	16.77
48.	ICGS-9157	43.70	20.61	32.44	18.58
49.	ICGS-10554	49.99	22.94	27.71	20.00
50.	ICGS-10890	45.62	24.15	31.30	19.58
51.	ICGS-12625	46.97	21.29	33.46	19.22
52.	ICGS-13941	41.41	28.17	31.55	14.17
53.	ICGS-13942	44.40	27.61	28.30	21.00
54.	AG-2006-14	48.67	23.33	31.22	18.92
55.	AG-2006-15	48.52	24.66	35.61	20.00
S.E.±		1.56	1.17	1.14	0.93
C.D. (0.05)		4.39	3.39	3.19	2.62
C.V. %		4.41	5.00	4.41	4.89

Oil content (%):

Extraction of oil from the sample was done using standard Soxhlet units and oil content (%) determined using gravimetric methods.

RESULTS AND DISCUSSION

The results obtained from the present investigation are presented in Table 1,2 and 3 and Fig. 1 to 8:

Bio- chemical parameters such as oil per cent, protein content (%) and SDS PAGE analysis for protein and esterase isozymes are good indicators for the degradation of the biological components during storage of summer groundnut seed. Oil and protein content estimated from fresh groundnut seed of *Kharif* and from summer groundnut seed stored for a period of six months showed significant differences among the genotypes (Table 1).

Oil content (%) and oil colour :

Oil content (%) recorded in the summer groundnut seed (2010) showed significant differences among genotypes when stored for six months. The maximum oil content was found in ICGS 4849 (37.60%) in the stored summer seed. Whereas the minimum oil content was recorded in the genotype GAUG 10 (25.53%). For fresh seed of *Kharif* 2010, significant higher oil content was recorded in NRCG 6663 (50.31%), while minimum was recorded for the genotype ICGS 13941 (41.41%). Regarding the colour of oil extracted from fresh *Kharif* seed and summer seed stored for six months, significant colour variation were not observed among genotypes except that the colour of the oil from *Kharif* seed appeared more dark yellow as compared to that of stored summer seed.

Protein content (%) :

Significant differences in protein content was observed among the genotypes of summer groundnut when stored for six months. The stored seed recorded higher protein content in genotype GG-2 (26.00) whereas the lower protein content was observed in NRCG 6563 (11.28%). In freshly harvested *Kharif* seed, significant higher protein content was found in the genotype GG 2 (29.26%) while, lower protein content was found in the genotype GG 12 (18.93%). Similar results for difference in oil content and protein content due to seasons was reported by Valizadeh (2001), Yaw *et al.* (2008) and Nkafamiya *et al.* (2010).

SDS PAGE analysis :

SDS PAGE analysis of groundnut seed protein and esterase isozyme for summer and *Kharif* revealed that considerable number of genotypes showed (Table 2) maximum similarity (1.00) irrespective of the seasons which was fortyone for summer and thirty nine for *Kharif.* Cluster analysis using Jaccard co-efficient (NTSYS software) revealed three and four major clusters in *Kharif* and summer, respectively. It was observed that genotypes with common phylogeny and geographical orientation tend to cluster together. Similarly for esterase isozyme, fifty three and fortysix genotypes showed 100 per cent genetic similarity between them. In this case also, *Kharif* and summer groundnut produced three and four clusters, respectively. The protein characterisation studies using SDS PAGE analysis revealed various types of banding patterns for stored summer and fresh *Kharif* seeds. One hundred percent polymorphism was observed both in *Kharif* and summer with PIC value 0.94 in both cases.

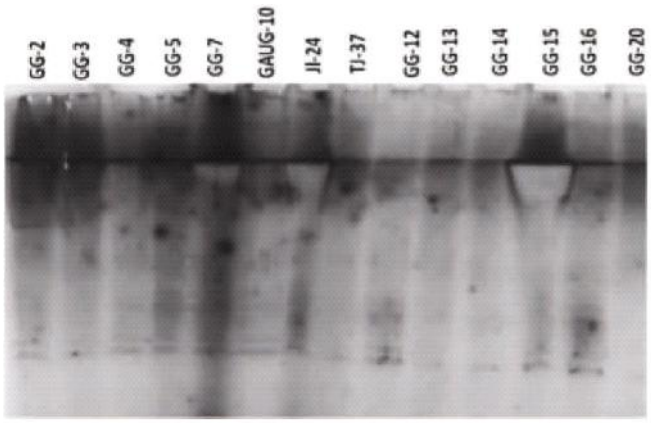
Similarly isozyme studies also indicated (Table 3) that the storage of summer seed contributed to less number of polymorphic bands in summer (101) as compared to *Kharif* (181). The lower PIC values observed for isozyme *i.e*, 0.73 (*Kharif*) and 0.69 (summer) indicated that characterisation using protein may give a more reliable estimate rather than isozymes. However, the number of polymorphic bands observed in fresh *Kharif* seeds (769) and stored summer seeds (696) were different indicating that protein denaturation must have occurred during storage. Esterase isozymes which are related to viability and germinability of seeds get progressively deactivated during storage (Hassanein, 1999, Aung and Donald,1995).

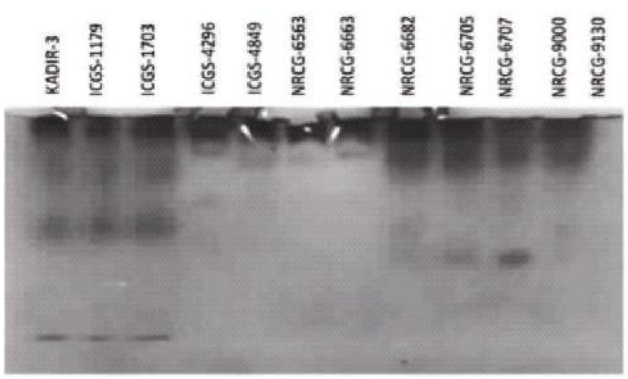
Table 2 : Details of polymorphism for protein studies in 55 groundnut genotypes for *Kharif* (fresh) and stored summer seed through SDS-PAGE analysis

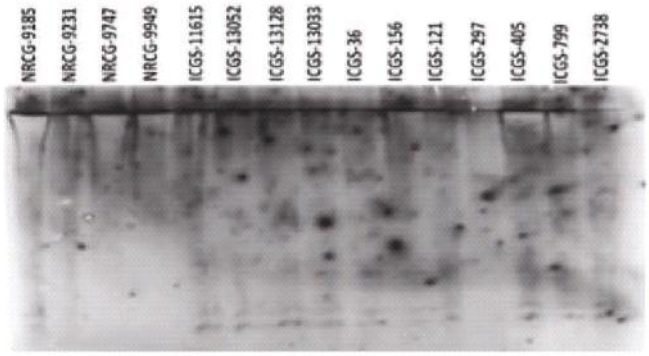
Sr. No	Markers	Seed season	Monomorphic band	Polymorphic bands (A)	Total (B)	Polymorphic band %, A/B× 100	PIC value
1.	Protein	Fresh *Kharif* 2010 seed	0	769	769	100 %	0.94
		Summer 2010 seed	0	696	696	100 %	0.94

Table 3 : Details of polymorphism for esterase isozyme studies in 55 groundnut genotypes for *Kharif* (fresh) and stored summer seed through SDS-PAGE analysis

Sr. No.	Markers	Seed season	Monomorphic band	Polymorphic bands (A)	Total (B)	Polymorphic band %, A/B× 100	PIC value
1.	Esterases	Fresh *Kharif* 2010 seed	0	181	181	100 %	0.73
		Summer 2010 seed	0	101	101	100 %	0.69







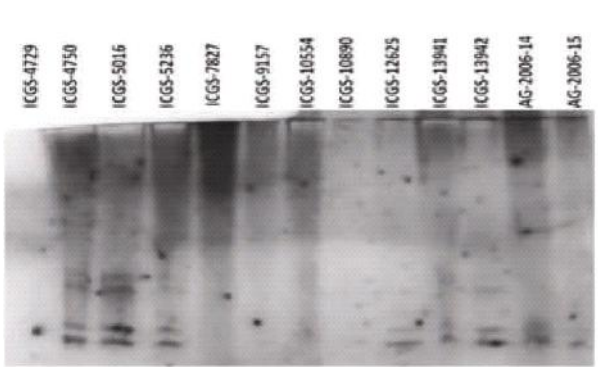
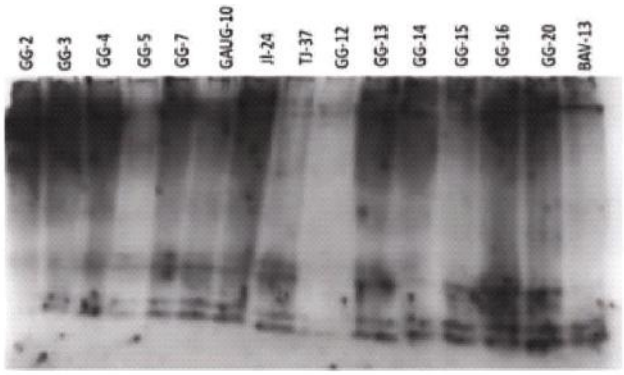
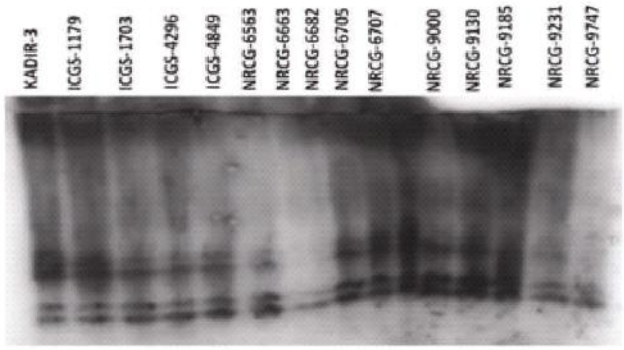
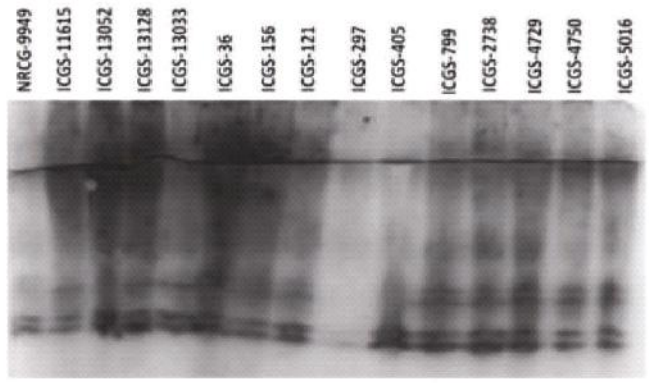


Fig. 1 : Protein profile of 55 groundnut genotypes form stored summer seed (2010)







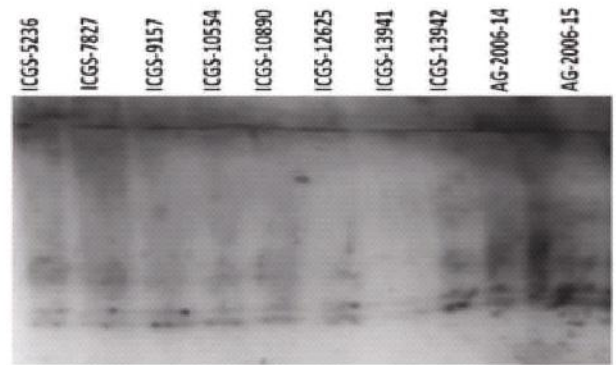


Fig. 2 : Protein profile of 55 groundnut genotypes form *Kharif* (2010)

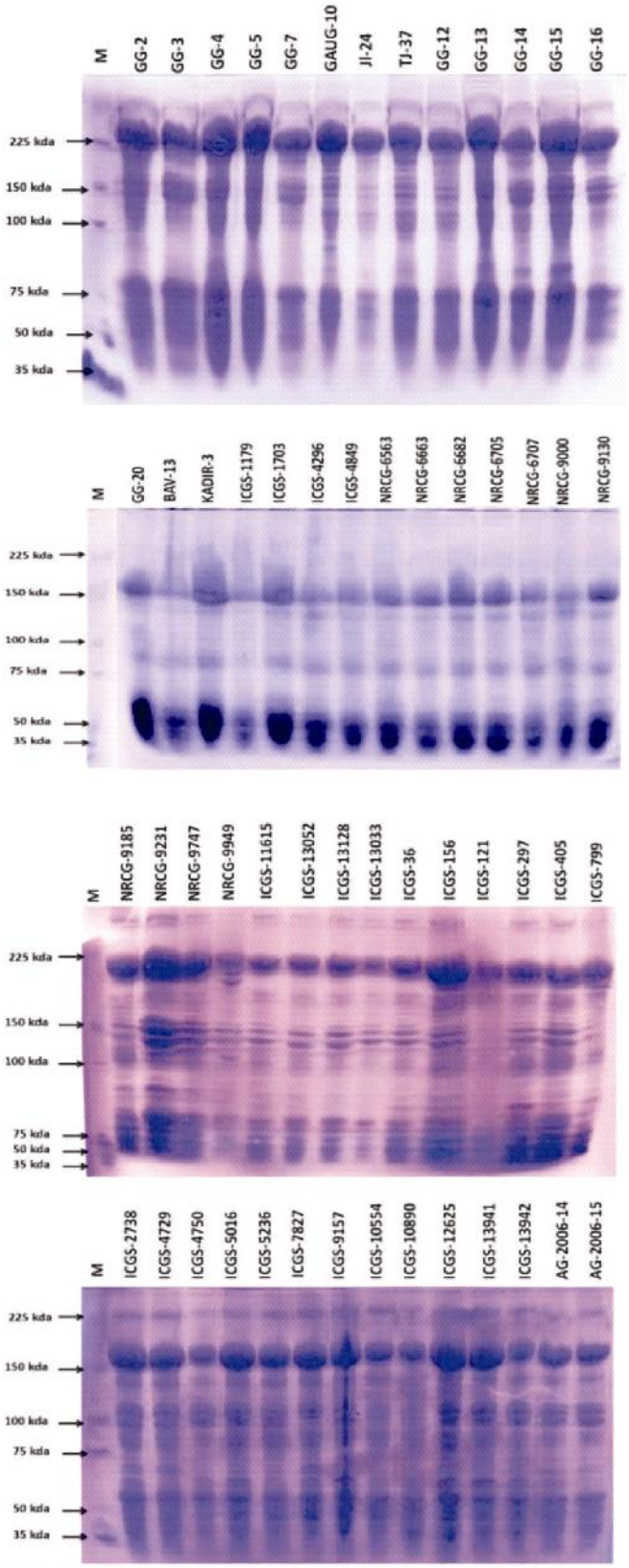


Fig. 3 : Esterase isozymes banding pattern in 55 groundnut genotypes from stored summer seed (2010)

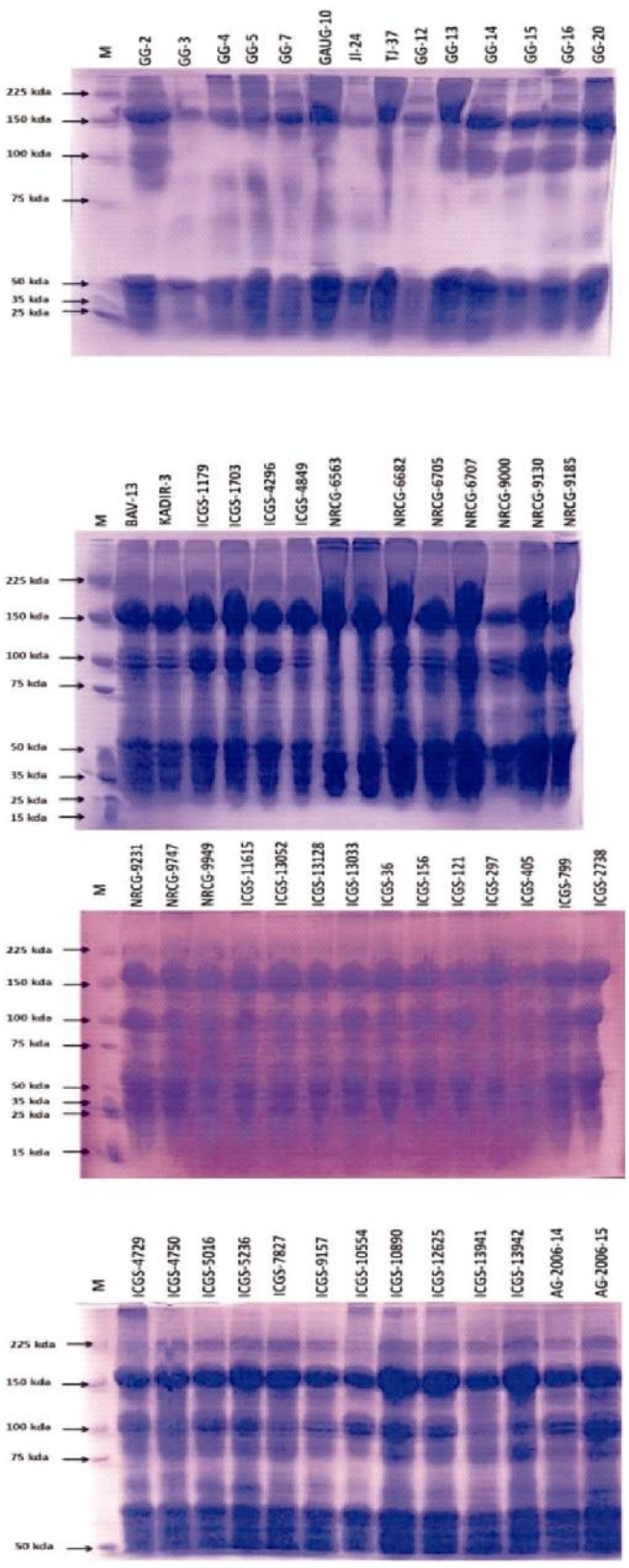


Fig.4: Esterase isozyme banding pattern in 55 groundnut genotypes (*Kharif* 2010)

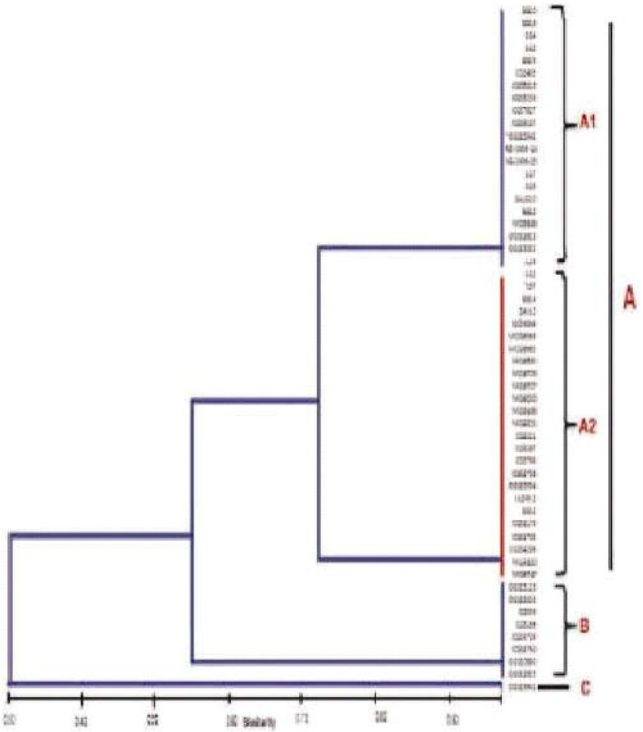


Fig. 5: Dendogram for esterase isoenzymes in fifty five groundnut genotypes developed through SDS-PAGE analysis (*Kharif* 2010)

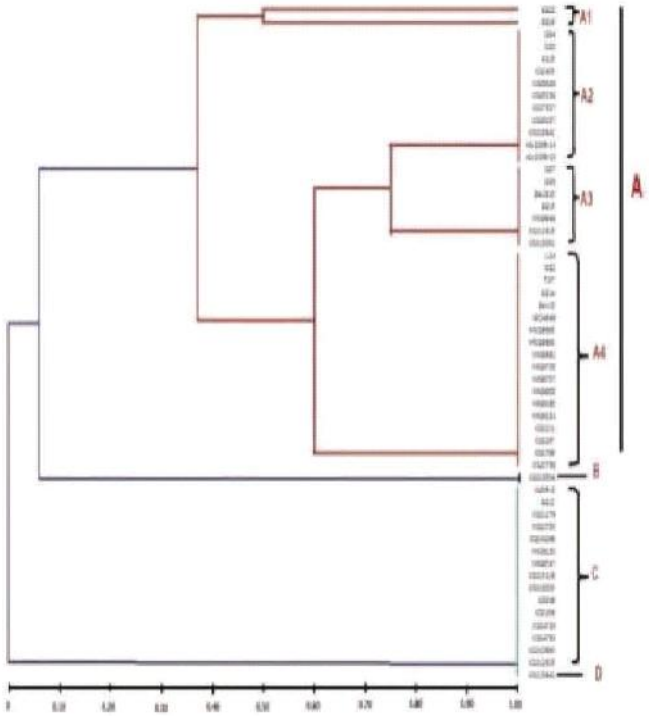


Fig. 6: Dendogram for esterase isoenzymes in fifty five groundnut genotypes developed through SDS-PAGE analysis (Summer 2010)

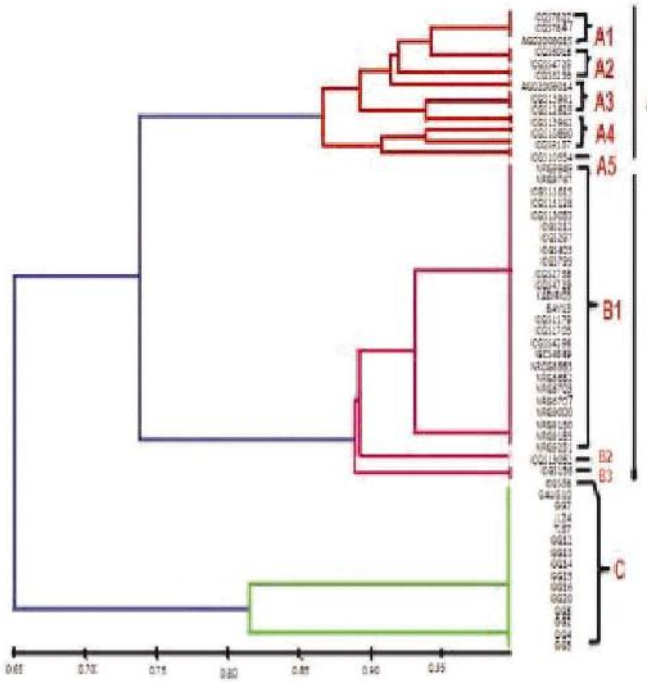


Fig. 7: Dendogram for protein in fifty five groundnut genotypes developed through SDS-PAGE analysis (*Kharif* 2010)

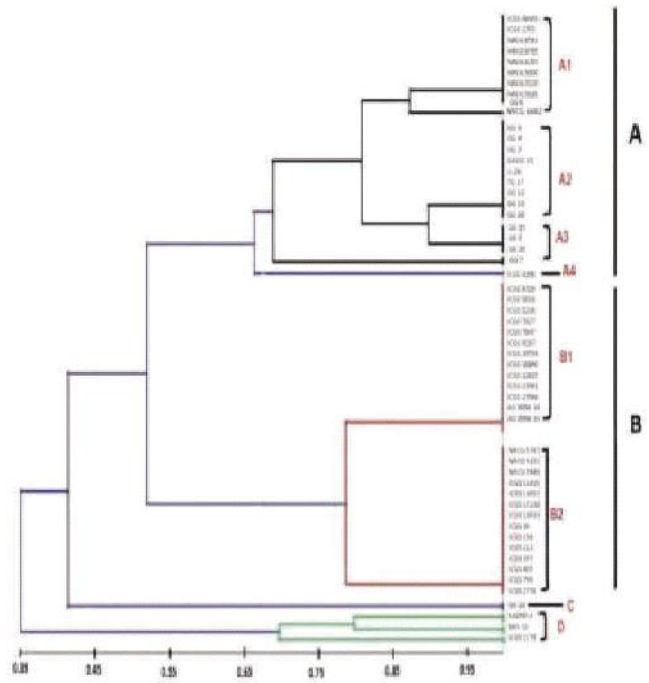


Fig. 8: Dendogram for protein in fifty five groundnut genotypes developed through SDS-PAGE analysis (Summer 2010)

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