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Genetic variability and association pattern among nutritional traits in recombinant inbred lines of groundnut (*Arachis hypogaea* L.)

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Abstract

Groundnut (*Arachis hypogaea* L.) is the world's third most important source of oil and fourth most important source of vegetable protein. Oil content, protein content and fatty acid composition (O/L ratio) are the most important quality attributes of groundnut. A mapping population segregating for these traits was evaluated for genetic variability and correlation among the traits. The population exhibited significant variation among the genotypes, seasons and G x E interaction. Moderate magnitude of variability followed by higher heritability was observed for most of the quality traits. Negative correlation between oil and protein content, oleic and linoleic acid indicated their antagonistic nature. All the eight fatty acids were correlated with each other either positively or negatively. Superior RILs were identified for higher protein content, oil content, oleic acid and O/L ratio from the population.

Key words: Oil, Protein, oleic acid, O/L ratio, correlation, heritability

Introduction

Cultivated groundnut (*Arachis hypogaea* L) also known as peanut is an important oilseed crop of the world covering an area of 25.2 m ha with a production of 35.9 m t. In India it is spread over an area of 6.6 m ha with production of 5.9 m t. (FAO 2006). Groundnut is grown primarily for human consumption either as a whole seed or processed to make peanut butter, oil and other products. Peanut seeds are rich source of edible oil and contain 42-50% oil, 25-32% protein on a dry weight basis. Oil and protein contents and oil quality with respect to its fatty acid composition are most important quality traits both for oil and confectionary purposes. Seed oxidative stability is closely associated with oil composition. Peanut seeds with high oleic acid content and O/L ratio and low iodine value have improved stability against lipid peroxidation and also higher shelf life can be achieved as compared to low O/L ratio because oleic acid, the 18-carbon monounsaturated fatty acid and precursor to linoleic acid, is less reactive with oxygen.

The development of cultivars in groundnut varies with the purpose for which they are put to use [1]. For edible oil purpose, cultivars having high oil content with high O/L ratio and low iodine value are preferred where as, the quality requirement for confectionary groundnut is more stringent and distinctly different. This requires additional efforts to develop confectionary grade varieties with high protein and sugar, low oil, reduced aflatoxin risk and high O/L ratio and low iodine value [2].

For making desired progress in breeding for increased oil, protein and oleic acid, the available genetic variability in the cultivated peanut should be enlarged and the donors with improved desired traits in the breeding programme should be selected after their evaluation in the multi-season and multi-location trials [3]. The knowledge on the estimates of variability and its heritable components in the material with which the breeder is working is essential for chalking out selection strategies. Before formulating suitable strategies to breed varieties for better quality, understanding the relationship among the quality traits is also of paramount importance. Hence, the present study was aimed at evaluation of recombinant inbred lines segregating for important nutritional quality traits (protein, oil and fatty acid profile) and their association pattern for the improvement of low heritable traits through indirect selection of the highly heritable traits.

Material and methods

A mapping population comprising of 146 RILs ($F_{9:10}$) developed from the cross TG 26 x GPBD 4 was used for the study. The female parent TG 26 is a Spanish bunch variety derived from BARCG 1 x TG 23 cross

and it is an early maturing, semi-dwarf variety with high pod growth rate, higher harvest index, greater partitioning efficiency, tolerance to bud necrosis but susceptible to rust and late leaf spot [4, 5]. The male parent GPBD 4 is a Spanish bunch cultivar derived from the cross KRG1 x CS16. CS 16 is a *virginia* bunch interspecific derivative (*A. hypogaea* x *A. cardensii*). GPBD 4 is resistant to late leaf spot and rust, high yielder, early maturing, high in oil and O/L ratio and protein content [6].

Mapping population consisting of 146 RILs and parents were subjected for phenotypic evaluation for several quality traits *viz.*, protein content (%), oil content (%), fatty acid composition consisting of 8 fatty acids (Palmitic, stearic, oleic, linoleic, arachidic, eicosenoic and behenic acids). The experiment was carried out in randomized complete block design with two replications during Rainy and Post-rainy seasons (2007) at the Botany Garden located at University of Agricultural Sciences Dharwad. All the traits were estimated by near Infrared spectroscopy (NIRS) at seed quality testing and research lab. Oil stability indices *viz.,* Oleic/Linoleic acid (O/L) ratio, Iodine value (IV), Unsaturated/saturated (U/S) fatty acid ratio and % of saturated fatty acids (% S) were computed as follows:

- **Oleic/Linoleic acid (O/L) ratio:** % of Oleic acid (C18:1)/ % of Linoleic acid (C18:2).
- **Iodine value (IV):** (% Oleic x 0.8601)+(% Linoleic x 1.7321)+(% Eicosenoic x 0.7854) [7].
- **Unsaturated/saturated fatty acid (U/S):** % (Oleic + Linoleic + Eicosenoic)/% (Palmitic + Stearic + Arachidic + Behenic + Lignoseric).

% of Saturated fatty acids (% S): % (Palmitic + Stearic + Arachidic + Behenic + Lignoseric acid) [7].

The replicated data over two seasons for the above traits were subjected for statistical analysis *viz.,* Analysis of variation (ANOVA), mean, range, genetic variability components such as phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV), heritability (h^2) and genetic advance as per cent mean (GAM) and correlation analysis. A statistical software SPAR was used for the analysis.

Results and dscussion

Analysis of variance revealed significant variation among the RILs, seasons and RILs x season interaction for all the fourteen quality traits (Table 1). Based on the mean values, GPBD 4 was higher value parent for

Table 1. Analysis of variance pooled over two seasons (rainy and post rainy 2007) for nutritional quality traits in TG 26 x GPBD 4 mapping population

Traits/Source of variation	Mean sum of square							
	S	R	S x R	G	S x G	Error	CV	SE (+)
df	1	1	1	145	145	290		
Protein content (%)	65.31**	2.5	2.19	26.69**	13.66**	1.45	4.02	0.7
Oil content (%)	38.87**	0.63	0.06	9.23**	5.96**	1.18	2.38	0.63
Palmitic acid	333.55**	0.65	0.27	1.52**	0.64**	0.13	3.5	0.26
Stearic acid	0.14**	0.31	0.13	0.72**	0.31**	0.15	11.08	0.27
Oleic acid	292.63**	0.38	0.63	82.84**	35.81**	2.13	3.16	1.03
Linoleic acid	1678.63**	0.06	0.06	59.68**	27.03**	1.72	4.08	0.93
Arachidic acid	29.74**	0.01	0.01	0.049**	0.029**	0.02	6.39	0.08
Eicosenoic acid	24.59**	0.01	0.01	0.03**	0.014**	0.01	7.58	0.04
Behenic acid	4.40**	0.10	0.01	0.26**	0.13**	0.03	4.6	0.13
Lignoseric acid	17.71**	0.02	0.00	0.05**	0.021**	0.01	7.8	0.08
O/L ratio	3.36**	0.00	0.01	0.01*	0.26**	0.01	1.2	0.81
Iodine value (IV)	2741.00**	2.00	1.5	31.4**	15.38**	1.32	1.2	0.81
U/S ratio	19.87**	0.005	0.01	0.17**	0.08**	0.01	3.11	0.08
%S	351.25**	0.031	0.19	2.99**	1.20**	0.25	2.08	0.35

S- Season R-Replication G- Genotypes

protein, oil content, oleic acid, eicosenoic acid, lignoseric acid, O/L ratio and U/S ratio, whereas, TG 26 was a higher value parent for palmitic acid, stearic acid, arachidic acid, behenic acid, iodine value and % S. Hence, GPBD 4 is superior parent for all the important nutritional traits (Protein, oil, oleic and O/L ratio) compared to TG 26. Transgressive segregants were observed in both the directions for all the traits as revealed by the range of variation indicating the contribution of favorable alleles from both the parents (Table 2).

Genetic variability components revealed low to moderate magnitude of variation (PCV, GCV) and genetic advance with very high heritability for protein but lower magnitude of variation with higher heritability and lower genetic advance for oil content in individual seasons (data not shown). Across the seasons, there was reduction in values of components of variability for both the traits, but the reduction was more for oil compared to protein indicating preponderance of G x E interaction for oil as compared to protein. Hence, in spite of high heritability, there is better scope for selection for protein compared to oil content in this population (Table 2).

All the oil quality parameters except O/L ratio had low to moderate PCV and GCV with high to very high heritability coupled with low to moderate GAM. O/L ratio recorded higher magnitude and heritable variation as evidenced by high PCV, GCV, heritability and GAM. (Table 2).

Before formulating suitable strategies to breed varieties for better quality, understanding the relationship among oil quality traits is of paramount importance. All the fatty acids are linked in the biosynthetic pathway through modifications such as elongation and desaturation. Hence, alteration in biosynthetic steps influences the whole fatty acid profile and the relationships among different fatty acids. These correlations may reflect precursor-product relations in some instances but probably also reflect genetic linkages of various enzymes involved in the conversions [8].

Like previous reports [9-12], the correlation between oil and protein content was negative. Such a relationship could be advantageous while developing cultivars for confectionary purpose where low oil and high protein is preferred. Oil content showed positive correlation (Table 3) with oleic, eicosenoic acid, lignoseric acid, O/L ratio and U/S ratio and negative correlation with stearic, arachidic and behenic acids which is in accordance with previous reports [3].

Table 2. Mean, range and genetic variability components for nutritional quality traits in individual and pooled data across the seasons

	Parental means		RILs					
Traits	TG 26	GPBD 4	Mean	Range	PCV	GCV	h^{2b}	GAM
Protein (%)	25.44	33.51	29.98	24.18-35.42	6.35	4.92	60.2	12.35
Oil (%)	43.88	47.98	45.61	41.77-49.22	2.88	1.62	31.7	2.1
Palmitic acid	10.64	9.48	10.22	6.32-13.37	5.8	4.59	62.7	7.53
Stearic acid	3.06	2.27	3.5	1.68-5.26	14.34	9.1	40.3	11.99
Oleic acid	37.07	51.65	46.17	27.89-65.90	8.07	7.43	84.6	14.08
Linoleic acid	41.39	28.9	32.11	16.11-49.11	9.79	8.9	82.6	16.66
Arachidic acid	2.03	1.58	1.86	0.71-2.46	7.44	3.81	26.2	3.76
Eicosenoic acid	0.68	0.95	0.83	0.21-1.25	10.37	7.07	46.6	9.65
Behenic acid	4.14	3.69	3.99	2.85-5.12	6.34	4.37	47.5	6.26
Lignoseric acid	0.95	1.38	1.24	0.55-1.84	9.79	6.45	43.4	8.84
O/L ratio	0.91	1.79	1.51	0.57-4.06	18.78	17.08	82.7	31.89
Iodine value (IV)	104.11	95.23	95.98	84.78-109.46	2.41	2.09	75.2	3.72
U/S ratio	3.81	4.42	3.82	3.10-5.15	5.02	3.94	61.6	6.28
%S	23.81	21.43	23.82	19.25-27.35	3.49	2.81	64.6	4.66

PCV-Phenotypic coefficient of variation, GCV-Genotypic coefficient of variation h^{2b}-broad sense heritability, GAM-Genetic advance as % mean

Table 3. Correlation pattern among the nutritional quality traits pooled over two seasons in TG 26 x GPBD 4 mapping population

Trait	Protein	Oil	Palmitic	Stearic	Oleic	Linoleic	Arachidic	Eicosenoic	Behenic	Lignoseric	O/L ratio	IV	U/S	%S
Protein content	1													
Oil content	-0.199*	1												
Palmitic acid	0.263**	-0.071	1											
Stearic acid	0.069	-0.452**	-0.024	1										
Oleic acid	-0.302**	0.334**	-0.721**	-0.252**	1									
Linoleic acid	0.316**	-0.311**	0.685**	0.139*	-0.987**	1								
Arachidic acid	-0.102	-0.208*	-0.433**	0.342**	-0.032	0.021	1							
Eicosenoic acid	-0.212*	0.450**	-0.081	-0.500**	0.425**	-0.428**	-0.441**	1						
Behenic acid	-0.034	-0.287**	0.085	0.347**	-0.591**	0.551**	0.359**	-0.262**	1					
Lignoseric acid	-0.198*	0.476**	-0.03	-0.658**	0.360**	-0.343**	-0.369**	0.855**	-0.214*	1				
O/L ratio	-0.350**	0.298**	-0.694**	-0.172*	0.983**	-0.987**	0.004	0.406**	-0.579**	0.345**	1			
Iodine value (IV)	0.319**	-0.254**	0.611**	-0.036	-0.926**	0.974**	-0.009	-0.389**	0.472**	-0.283**	-0.949**	1		
U/S ratio	-0.189*	0.395**	-0.647**	-0.652**	0.798**	-0.693**	-0.126	0.385**	-0.593**	0.406**	0.729**	-0.514**	1	
%S	0.171*	-0.377**	0.647**	0.653**	-0.799**	0.693**	0.118	-0.387**	0.602**	-0.399**	-0.724**	0.513**	-0.997**	1

Among the fatty acids, oleic acid, a major fatty acid had a strong negative correlation with palmitic acid, linoleic acid, behenic acid, Iodine value and % S and it had a strong positive correlation with O/L and U/S ratio. The inverse relationship of oleic acid with palmitic acid and linoleic acid was also evident from the earlier studies [1, 12-14]. The strong negative relationship between palmitic acid and oleic acid (r = 0.721) most likely represents an increased rate of palmitic acid elongation to stearic acid, with rapid desaturation to oleic acid via Δ-9 desaturase [15]. The strong negative correlation between oleic and linoleic acids (r = 0.987) results from there being the chief acyl groups in the oil so that one cannot increase much without decrease in the other. Hence, increased oleic acid normally resulted in reduced palmitic acid, linoleic acid and iodine value which is desirable from the point of health and stability. Linoleic acid, a polyunsaturated fatty acid is unstable at higher temperature and has an inverse relationship with oil stability [16, 17]. Stearic acid, a neutral fatty acid with respect to cardiovascular disease was negatively correlated with eicosenoic (r = 0.500) and lignoseric acids (r=0.658). Eicosenoic acid had a strong positive correlation with lignoseric (r = 0.855) acid and it is also in agreement with earlier reports [12, 18].

Superior RILs for protein (7), oil content (7), oleic acid (14) and O/L ratio (10) more than GPBD 4 along with lower oil content (12), low linoleic acid (14), and low iodine value (11) than TG 26 and low palmitic acid (11) than GPBD 4 were identified (data not shown) based on their mean values over seasons. Among these, one RI line (95) was showing exceptionally high oleic acid (58.88 %), low linoleic acid (21.25 %), high O/L ratio (2.98), low Iodine value (88.19) thus combining several favorable traits and it could be used in future breeding program for developing varieties with improved nutritional quality. However, none of the RILs had a combination of high protein, high/low oil with high oleic acid like GPBD 4. A large segregating population or intercrossing among the selected lines could be an ideal strategy to develop nutritionally superior genotypes.

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