academicresearchJournals

Vol. 7(2), pp. 57-62, March 2019
DOI: 10.14662/ARJASR2019.002

ISSN: 2360-7874
http://www.academicresearchjournals.org/ARJASR/Index.htm

Academic Research Journal of Agricultural Science and Research

Full Length Research

Genetic Variability for oil quality traits in Groundnut (Arachis *hypogaea* L.) cultivars

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Accepted 27 February 2019

Assessment of genetic variability with the help of suitable genetic parameters such as genetic coefficient of variation, heritability estimates, and genetic advance are absolutely necessary to start an efficient breeding program. Sixteen groundnut genotypes were evaluated for quantitative parameters. The crop was sown during 2015 wet season across four locations in Ethiopia. The experiment was laid out in an RCBD with two replications. The results of combined analysis of variance has shown that high heritability with high expected genetic advance were observed for stearic acid, arachidic acid, eicosenoic acid, lignoceric acid and oleic to linoleic acid ratio indicating the predominant role of additive gene action and the possibilities of effective selection for the improvement of these traits. However, heritability for oil content was low showing that direct selection for oil content is difficult; the possible improvement of oil content should be indirect selection through highly heritable traits.

Key words: Oleic to linoleic acid ratio, Additive gene action, Oil content, Heritability, Genetic advance.

Cite this article as: Zekeria, Y., Habtamu, Z., Wassu M., Shimelis H., Arno, H. (2019). Genetic Variability for oil quality traits in Groundnut (*Arachis hypogaea* L.) cultivars. Acad. Res. J. Agri. Sci. Res. 7(2): 57-62

INTRODUCTION

The basic key to bring about the genetic improvement to a crop is to utilize the available genetic variability (Ramalho *et al*., 2011). It is imperative to partition the observed variability into its heritable and non-heritable components and to have an understanding of parameters like genetic coefficient of variation, heritability and genetic advance, correlation and path analysis (Mukri *et al*., 2014). The presence of genetic variation in the breeding material at hand determines the success or failure of any breeding or bioengineering program. The measurement of genetic variation and understanding of mode of inheritance of quantitative traits, therefore, are essential steps in any crop improvement program. Heritability estimates provide authentic information about the faithfulness with which a particular genetic attribute will be transmitted to the successive generation. The higher the heritability, the simpler the selection process and greater the response to selection. A broad-sense heritability estimate provides information on the relative magnitude of genetic and environmental variation in the population (Dudley and Moll, 1969; Rafi and Nath, 2004). Genetic variability for oil traits is necessary to conduct groundnut breeding for oil yield, oil content and quality traits. Furthermore, no sufficient information is found on genetic Variability of oil traits in groundnut genotypes from Ethiopia. Therefore, the present study was undertaken with the objective of determining broad sense heritability and response to selection for yield, other agromorphological and oil traits in groundnut genotypes grown in Ethiopia.

MATERIALS AND METHODS

The field experiment was carried out across six locations viz. Babile, Fedis, Pawe, and Guba during 2015 growing season in Ethiopia under rain fed condition. The laboratory experiment was carried out for seed samples grown in four locations viz Fedis, Mechara (Locations in Eastern Ethiopia), Pawe, and Guba (locations in Western Ethiopia).Before running the laboratory experiment the moisture content of seed samples were reduced to 5%. The lab experiment was carried out in two replications by taking 10gm of seed samples from 16 groundnut genotypes grown across four locations. Oil content and fatty acid profile determination was carried out based on the following technique:

Total lipid from the seed sample was quantitatively extracted, according to the method of Folch *et al*. (1957), using chloroform and methanol in a ratio of 2:1. An antioxidant, butylated hydroxytoluene was added at a concentration of 0.001 % to the chloroform: methanol mixture Iodine value was determined with the Hanus method (AOAC nr. 920.158, 1990).Fatty acid methyl ester samples were identified by comparing the retention times of FAME peaks from samples with those of standards obtained from Supelco (Supelco 37 Component Fame Mix 47885-U, Sigma-Aldrich Aston Manor, Pretoria, South Africa). Nonadecanoic acid (C19:0) was also obtained from Supelco. All other reagents and solvents were of analytical grade and obtained from Merck Chemicals (Pty Ltd, Halfway House, Johannesburg, South Africa). Fatty acids were expressed as the proportion of each individual fatty acid to the total of all fatty acids present in the sample. The following fatty acid combinations were calculated: total saturated fatty acids (TS), total monounsaturated fatty acids (TMUS), polyunsaturated fatty acids (TPU), total unsaturated fatty acids (TUS) and TPUS/TS ratio.

Genetic variability Parameters were worked out as follows:

The analysis of variance was used to estimate genetic variances using the method of moments (Searle *et al*., 1992), i.e., the mean squares were equated to their respective expectations and the estimates of variance for each genotype were computed based on the following linear model was used to perform the analyses:

$$Y_{rge}=\mu + \propto_g+\beta_e +\rho_r(\beta_e) + \alpha_g\beta_e + \varepsilon_{rge}$$

where Y_{rge} is the measured trait of genotype in replication r at location e; μ is the grand mean; α_g&$\beta_e$are the genotype and location main effects; ρ_r (β_e) is the replication effect nested within location; $\alpha_g\beta_e$is the interaction between genotype and location; and $\varepsilon_{rge}$is residual or error of plot containing genotypes in replication *r* and environment e.

Phenotypic, genotypic, and environmental variances were computed from the respective mean squares following the procedures suggested by Allard (1960), and Singh and Chaundhary (1979).Total variation was partitioned into phenotypic ($\sigma^2{}_p$), genotypic ($\sigma^2{}_g$) and environmental ($\sigma^2{}_e$) variance based on expectation of mean square for respective source of variation described in ANOVA (Table 1) as suggested by Holland *et al*. (2003).

All these parameters were obtained from analysis of variance table 1 according to Comstock and Robinson (1952) Heritability in broad sense (H^2%) was estimated according to Falconer (1989). The heritability percentage was categorized as low when less than 40%, medium, 40–59%, moderately high, 60-79% and very high, 80% and above as indicated by Singh (2001).

The magnitude of genetic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) existing in a trait was estimated by formula given by Burton (1952):

GCV (%) = $\frac{\sqrt{\sigma^2{}_g}}{\bar{x}}$ x100, and PCV=$\frac{\sqrt{\sigma^2{}_p}}{\bar{x}}$ x100. The GCV and PCV values were categorized as low when less than 10%, moderate, 10-20% and high, greater than 20% as indicated by Deshmukh *et al.* (1986). Genetic advance (GA) was calculated with the method suggested by Allard (1960),and Singh and Chaudhury (1985): $GA=K\sigma_pH^2$: Where, GA: genetic advance; K: constant = 1.76 at 10% selection intensity; σ*p*: standard deviation of phenotypic variance ;H^2: Heritability in broad sense.GA as % of mean (GAM) = $\frac{GA}{\bar{x}}$ x100%. The Genetic advance (GA), expressed as a percentage of mean, was categorized as

Where MSg is the mean square for genotype; MSgxe: mean square for genotype X environment interaction; e:number of environments; r: number of replication; $\sigma^2_{\varepsilon\prime}$: residual variance; σ^2_e: variance due to plots or environments; σ^2_ε: error variance; σ^2_b: within plot variance or variance due to block effects; n: number of plants per plot;

Hb2: heritability in a broad sense based on entry or genotype mean basis.

. $\sigma^2{}_e$= MSE

$$\sigma^2_g = \frac{MS_g - MS_{gxe}}{er}$$

$$\sigma^2_p = \frac{MS_g}{er} = \sigma^2_g + \frac{\sigma^2_{gxe}}{e} + \frac{\sigma^2_{\varepsilon\prime}}{er}$$

$$\sigma^2_{\varepsilon\prime} = \sigma^2_{\varepsilon} + \frac{\sigma^2_b}{n}$$

$$H^2_{b(\text{entry mean basis})} = \frac{\sigma^2_g}{\sigma^2_p} = \frac{MS_g - MS_{gxe}/er}{MS_g/er} = \frac{\sigma^2_g}{\sigma^2_g + \frac{\sigma^2_{gxe}}{e} + \frac{\sigma^2_{\varepsilon\prime}}{er}}$$

Table 1. ANOVA layout for evaluation of genotypes (g) in replicated(r) trials across environments(e) in RCBD

Source of variation	Degree of freedom	Expected mean squares
Environment	e-1	$rg\sigma^2_e + r\sigma^2_{gxe} + g\sigma^2_b + \sigma^2_\varepsilon$
Rep(env)	(r-1)e	$g\sigma^2_b + \sigma^2_\varepsilon$
Genotype	g-1	$er\sigma^2_g + r\sigma^2_{gxe} + \sigma^2_\varepsilon$
Genotype x environment	(g-1)(e-1)	$r\sigma^2_{gxe} + \sigma^2_\varepsilon$
Error	(g-1)(e-1)e	σ^2_ε

high when it is above 20%, moderate, 10-20% and low when it is less than 10% based on Johnson *et al.* (1955).Data were subjected to combined analysis of variance (ANOVA) using the PROC MIXED procedure of SAS (SAS, 2011) with genotypes being considered as fixed effects, while locations, replications and blocks within locations as random effects. Homogeneity of variance was tested using Obrein test (Obrien, 1981).

RESULT AND DISCUSSION

The ANOVA showed that individual location ANOVA was significant and homogeneous. The results of combined analysis of variance showing mean squares for 17 oil traits and grain yield evaluated for 16 groundnut genotypes combined across four locations were presented in Table 2.Highly significant differences were detected among the genotypes, locations and genotype x location interactions of all the traits indicating prevalence of genetic variability.

The mean, range, coefficients of genotypic and phenotypic variations, heritability, and genetic advance of various oil traits and quality parameters were given in the Table 3.The genotypic coefficient of variation provides a measure to compare genetic variability present in quantitative parameters (Maurya *et al*., 2014). The GCV, in the present study, ranged from 1% for IV and TUS to 23% for stearic acid. High GCV was observed for stearic acid, eicosenoic acid, GY and OY indicating high degree of genetic variability. Moderate GCV was obtained for palmitic acid, arachidic acid, behenic acid, lignoceric acid, and O/L ratio indicating existence of genetic variability. Low GCV was observed for oil content, iodine value, oleic acid, linoleic acid, total saturated fatty acids (TS), total monounsaturated fatty acids (TMUS), total polyunsaturated fatty acids (TPUS) and total polyunsaturated to saturated fatty acids (TPUS/TS) ratio indicating the existence of little genetic variability with regard to these parameters and difficulty of improving such traits directly.

Phenotypic coefficient of variation which measures total relative variation was high for stearic acid, eicosenoic acid, GY and OY indicating high degree of genetic variability. Moderate PCV was obtained for palmitic acid, linoleic acid, arachidic acid, behenic acid, lignoceric acid, total polyunsaturated fatty acids, and O/L ratio. Low PCV was observed for oil content, iodine value, oleic acid, total saturated fatty acids (TS), total monounsaturated fatty acids (TMUS), total unsaturated fatty acids (TUS), TPUS/TS ratio, and TUS/TS ratio. These results are in accordance with the findings of Ashish (2013) in groundnut, Azharudheen *et al*. (2013) and Mukri *et al*. (2014) in groundnut, Archana *et al*. (2007) in soybean and Kavera *et al*. (2008) in groundnut where they observed greater magnitude of variations for stearic acid, oleic acid, linoleic acid content and O/L ratio.

In the present study, high heritability with high-expected genetic advance were observed for stearic acid, arachidic acid, eicosenoic acid, lignoceric acid, O/L ratio, GY and OY indicating the predominant role of additive gene action and the possibilities of effective selection for the improvement of these traits. Such estimate of high heritability with moderate to high genetic advance is indicating the chance of effective selection of these traits for improvement of oil quality traits. Johnson *et al*. (1955) suggested that heritability estimates along with genetic advance would be more useful in predicting desired trait

Table 2. ANOVA for oil traits evaluated for 16 groundnut varieties across four locations

Trait	Standard	Min	Max	Mean	CV	Std	MSenv	MSgen	MS gxe	MSE
Oil	NA	40.54	52.32	45.7	1.82	2.20	47.48**	11.53**	5.93**	0.69
IV	86-107	86.74	114	98.3	1.00	5.20	771.54**	15.33**	8.91**	0.97
Palmitic acid (C16:0)	8.0-14	8.08	12.55	9.67	1.58	0.96	0.40**	7.08**	0.19**	0.02
Stearic acid (C18:0)	1.0-4.5	1.27	5.76	2.67	3.94	1.00	17.51**	3.48**	0.46**	0.01
Oleic acid (C18:1)	35-69	38.47	62.34	49.4	0.72	5.69	692.38**	104.32**	10.34**	0.13
Linoleic acid(C18:2)	12-43	19.67	46.75	31.9	2.27	5.63	811.51**	74.76**	9.96**	0.53
Arachidic acid (C20:0)	1.0-2.0	0.56	1.96	1.17	6.53	0.29	1.15**	0.33**	0.04**	0.006
Eicosenoic acid (C20:1)	0.7-1.7	0.65	1.94	1.19	3.82	0.34	1.67**	0.51**	0.04**	0.002
Behenic (C22:0)	1.5-4.5	0.94	3.38	2.56	8.33	0.42	1.69**	0.76**	0.07**	0.05
Lignoceric (C24:0)	0.5-2.5	0.33	3.99	1.26	21.64	0.35	0.60**	0.34**	0.09**	0.07
TS	12-27.8	14.09	20.91	17.36	2.54	1.48	18.38**	11.79**	0.73**	0.19
TMUS	35.7-69	39.16	63.54	50.72	0.74	5.64	620.79**	112.85**	9.45**	0.14
TPUS	12-43.3	19.67	46.75	31.93	2.27	5.60	810.44**	74.73**	9.98**	0.53
TUS	---	79.09	85.91	82.64	0.53	1.48	18.38**	11.79**	0.73**	0.19
TPUS/TS	0.8 to 1.0	1.06	3.32	1.86	5.91	0.39	4.21**	0.19**	0.06**	0.01
O/L	2.0-4.0	0.82	3.17	1.64	2.99	0.53	6.59**	0.63**	0.13**	0.002
GY	NA	2049	9795	4856.4	14.79	1658.3	9.5E+06**	1.0E+07**	3.0E+06**	5.2E+05
OY	NA	889.7	4834.3	2219.7	14.9	758.9	1.9E+06**	2.2E+06**	6.1E+05**	1.1E+05

IV: iodine value; TS: total saturated fatty acids; TMUS: total monounsaturated fatty acids; TPUS: total polyunsaturated fatty acids; TUS: total unsaturated fatty acids; TPUS/TS: total polyunsaturated to total saturated fatty acids; O/L: oleic to linoleic acid ratio; TUS/TS: total unsaturated to total saturated fatty acids; GY: grain yield (kg/ha); OY: oil yield (kg/ha).

Table 3. Variance components and genetic variability parameters of 17 oil traits and quality parameters measured for 16 groundnut varieties

Trait	Ve	Vg	Vp	ECV(%)	PCV(%)	GCV(%)	H^2(%)	GAM(%)
Oil	0.69	0.7	1.44	2.0	3.0	2.0	49	2.0
IV	0.97	0.80	1.92	1.0	1.0	1.0	42	1.0
Palmitic acid	0.02	0.86	0.89	2.0	1.0	1.0	97	17
Stearic acid	0.01	0.38	0.44	4.0	25	23	87	38
Oleic acid	0.13	11.75	13.04	1.0	7.0	7.0	90	12
Linoleic acid	0.53	8.10	9.35	2.0	1.0	9.0	87	15
Arachidic acid	0.006	0.04	0.04	7.0	17	16	88	27
Eicosenoic acid	0.002	0.06	0.06	4.0	21	20	92	34
Behenic acid	0.05	0.09	0.10	9.0	12	12	91	19
Lignoceric	0.07	0.03	0.04	21	16	14	74	21
TS	0.19	1.38	1.47	3.0	7.0	7.0	94	12
TMUS	0.14	12.93	14.11	1.0	7.0	7.0	92	12
TPUS	0.53	8.09	9.34	2.0	1.0	9.0	87	15
TUS	0.19	1.38	1.47	1.0	1.0	1.0	94	2.0
TPUS/TS	0.01	0.02	0.02	5.0	8.0	7.0	68	1.0
O/L	0.002	0.06	0.08	3.0	17	15	79	24
GY	5.2E+05	9.0E+05	1.3E+06	15	23	20	71	29
OY	1.1E+05	2.0E+05	2.8E+06	15	24	20	73	30

TS: total saturated fatty acids; TMUS: total monounsaturated fatty acids; TPUS: total polyunsaturated fatty acids; TUS: total unsaturated fatty acids; TPUS/TS: total polyunsaturated to total saturated fatty acids; O/L: oleic to linoleic acid ratio; total unsaturated to total saturated fatty acids;OY: oil yield (kg/ha);ECV: environmental coefficient of variation; PCV: phenotypic coefficient of variation; GCV: genotypic coefficient of variation;H^2: heritability in broad sense; GA: genetic advance; GAM: genetic advance as a percent of mean.

under phenotypic selection than heritability estimate alone. High heritability with moderate genetic advance were observed for palmitic acid, oleic acid, linoleic acid, behenic acid, total saturated fatty acids (TS), total monounsaturated fatty acids (TMUS), total polyunsaturated fatty acids (TPUS), TPU/TS and TUS/TS. The present result was in accordance with previous report by Azharudheen *et al*. (2013) who obtained high heritability with high genetic advance for the majority of oil traits.

Low heritability with low genetic advance were observed for oil content, iodine value and total unsaturated fatty acids(TUS) indicating low genetic potentials for these traits, high effect of the environment in determining measured traits and absence of predominant role of additive gene action instead environmental factors were more important for such traits. Heritability for oil content is low showing that direct selection for oil content is difficult; the possible improvement of oil content should be through indirect selection. Predictability of high performance and hence selection of materials based on the above criteria may lead to successful groundnut breeding program. This finding was not agreement with the previous reports of Ashish (2013), Kavera *et al*. (2008), Sarvamangala *et al*. (2011), Noubissie *et al*. (2012) who have got high heritability for oil content, Mollers and Schierhold (2002) suggested low to moderate broad sense heritability indicates the greater influence of environment in the expression of these traits. However, genetic advance can help to predict the extent of genetic improvement that can be achieved for the traits.

A high genetic gain along with the high heritability would suggest that character is governed by additive gene action which is suitable for making effective selection. The estimated genetic advance was high for the traits like oil yield, pod yield and kernel yield (Sharma and Gupta, 2011). The high genetic advance coupled with high heritability estimates for these traits suggested the importance of additive genetic variance and improvement of these traits could be made by simple phenotypic selection.

CONCLUSION

The biochemical analysis of oil traits will have greater contribution for the future groundnut breeding program in Ethiopia. The present study has found that stearic acid, arachidic acid, eicosenoic acid, lignoceric acid, O/L ratio, palmitic acid, oleic acid, linoleic acid, behenic acid, total saturated fatty acids (TS), total monounsaturated fatty acids (TMUS), total polyunsaturated fatty acids (TPUS), TPUS/TS and OY were more variable traits among evaluated genotypes. These traits have potential in breeding groundnut for oil traits. However, low genetic variability for oil content and total unsaturated fatty acids (TUS) and iodine value (IV) was observed indicating that breeding for oil content should follow indirect selection through other traits due to low genetic advance for oil content trait.

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