RESEARCH PAPER International Journal of Agricultural Sciences, Vol. 7 Issue 1 (January, 2011) : 1-7

Association analysis and identification superior segregants for resistance to *Sclerotium rolfsii* and yield component traits in groundnut (*Arachis hypogaea* L.)

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ABSTRACT

Evaluation of 165 groundnut genotype along with parent (TAG 24 and R 9227) under artificial inoculation condition for stem rot, *Sclerotium rolfsii* indicated the significant difference among genotype, season and genotype x season interaction for disease, yield and yield related parameters. Significant positive correlation of plant population, primary branches, test weight, shelling percentage, oil content exhibited positive significant association with pod weight per plant and negatively correlated with disease at 30, 60, 90 days and was negatively correlated with plant population, And pod weight per plant was negatively correlated with plant population, plant height and disease at harvest. None of the genotypes was completely free from the disease. However, six genotypes, *i.e.* line number 21, 77,199, 25,165and 36 were resistant compared to resistant parent *i.e.*, R 9227. Among parent R 9227 showed resistance to stem and pod rot and TAG 24 showed susceptible to stem and pod rot and six genotype showed resistance to stem and pod rot compared to both parent.

Pujer, Santoshkumar, Satish, R.G. and Boranayaka, M.B. (2011). Association analysis and identification superior segregants for resistance to *Sclerotium rolfsii* and yield component traits in groundnut (*Arachis hypogaea* L.). *Internat. J. agric. Sci.*, **7**(1): 1-7.

Key words : *Sclerotium rolfsii,* Groundnut, Association analysis, Superior segregants, Yield and yield component traits

INTRODUCTION

Groundnut or peanut (*Arachis hypogaea* L.) is one of the important economic oilseed crops of the world. Groundnuts contain more protein than meat-about two and a half times more than eggs, and far more than any other vegetable food except soybean and yeast. The proteins in groundnut are well balanced, except for slight deficiency in some of the essential amino acids. As it happens, these amino acids are, abundant in milk which can be combined with groundnut products for better results. Groundnut oil primarily used in the manufacture of vegetable oil (vanaspati ghee). Groundnut seed contains about 45 per cent oil and 26 per cent protein. In addition to this, it contains 18 per cent carbohydrates. It is also very good source of mineral (calcium, magnesium and iron) and vitamins (B_1, B_2 and Niacin). Obviously, poor soil fertility, abiotic and biotic stress factors limit groundnut crop growth and yield to many ways. Among biotic stresses stem and pod rot disease caused by *Sclerotium rolfsii* Sacc. is one of the significant factors contributing to yield loss. Only limited resistance screening of germplasm has been attempted. There are very few reports of clear varietal differences for resistance to stem and pod rots. Although, no genotype was known to be immune or even highly resistant to *Sclerotium rolfsii*, several genotypes and advanced breeding lines have shown field resistance (Smith *et al.*, 1989; Grichar and Smith, 1992 and Shokes *et al.*, 1993).

As in the case of many other diseases, breeding for disease resistance is the best way of controlling the *S. rolfsii* to initiate breeding programme for resistance to any of the disease, understanding of the basic mechanism of disease resistance and its inheritance are pre-requisite. It is desirable to have a variety resistance to the disease, combined with other desirable economic characters. The knowledge of mode of inheritance, variability of resistance/ susceptibility is essential to have effective selection programme. Estimate of gene effects will help in predicting the effectiveness of selection. The relative variance will decide the breeding procedure to be followed through lot of information is available on the quantitative characters but less information is available about the inheritance of *S. rolfsii* resistance as well as its association with other morphological traits.

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MATERIALS AND METHODS

The experimental material comprised of TAG 24 ´ R 9227 cross. This cross was made by using susceptible (TAG 24) and resistant (R 9227) parents. Hybridization was forwarded to get F_1 (F_2 seeds) and F_2 generation (F_3 seeds) by selfing. The F_2 generation was advanced to F_3 through single seed decent method. Individual F_3 families were propagated as bulk in F_4 and F_5 generation. F_5 generation was selected and evaluated in F_6 generation and artificial inoculation conditions were created during summer 2009.

S. rolfsii was isolated from diseased groundnut plant grown in a verticals and mass multiplication on sand – corn meal medium which was prepared in the proportion of 95:5 in order to get maximum sclerotial production (Abeygunwardhana and Wood, 1975). Two hundred gram of sand-corn meal medium was taken in 500 ml conical flasks and mixed with 30 per cent of distilled water and it was sterilized in autoclave at 121^0C at 1.1 kg per cm^2 pressure for 20 minutes. The pure culture of isolate of *S. rolfsii* was inoculated separately for flasks under aseptic condition and incubated at 27 $\pm$ 1^0C for 30 days. These flasks were shaked often to get uniform growth of isolate. The mass culture thus obtained was used for further studies. Inoculum containing mycelium and sclerotia along with corn meal and sand was applied to the soil surface around the base of the plants at 125 g per 2.5 m row, at 30 days after sowing or at flower initiation. Chopped sorghum stubbles (3 – 4 cm pieces) were scattered along the rows to enhance the fungal growth on soil. After two weeks, the inoculation was repeated. During summer season, the field was irrigated at five days intervals until pod formation to promote stem rot development. The interval was increased at 15 to promote pod infection.

RESULTS AND DISCUSSION

Yield is the result of combined effect of several component characters and environment. Understanding of the interaction of characters among themselves and with the environment is of great use in the plant breeding. Correlation studies provide information on the nature and extent of association between any two pairs of metric characters. Hence, it would be possible to bring genetic up gradation in one character by selection of other. Grafius (1959) opined that there may not be any gene for yield as such, but operate only through its components. Obviously, knowledge about character association will surely help to identify the character to make selection for

Table 1: Genotypic and phenotypic correlations of *Kharif* 2008

Characters	PP	PH	PB	SB	LL	LW	TW	SP	OC	DAH	PWP
Plant	1.000	-0.307*	0.348*	-0.073	-0.942*	-0.951*	-0.344*	0.184*	0.372*	0.483*	-0.332*
Population		-0.117**	0.016	-0.025	-0.164*	-0.131**	-0.061	0.071	0.080	0.206*	-0.152**
Plant height		1.000	-0.151**	0.057	0.354*	0.248*	0.155**	0.201*	-0.165*	-0.708*	-0.304*
			-0.049	0.051	0.200*	0.090	0.083	0.059	-0.129**	-0.124**	-0.239*
Primary branches			1.000	-0.078	-0.662*	-0.589*	-0.340*	-0.107	0.030	0.083	0.267*
				-0.036	-0.099	-0.091	0.058	-0.027	0.078	-0.031	0.086
Secondary branches				1.000	0.062	-0.166 *	0.017	-0.036	0.135**	-0.098	-0.114**
					0.032	-0.069	0.005	-0.024	0.107	-0.013	-0.107
Leaf length					1.000	0.566*	-0.098	0.048	-0.202*	-0.339*	-0.325*
						0.617*	0.030	0.004	-0.024	-0.090	-0.133**
Leaf width						1.000	0.324*	-0.196*	0.006	0.108	-0.187*
							0.037	-0.097	0.033	0.005	-0.006
Test weight							1.000	0.623*	0.109	-0.225*	0.051
								0.269*	0.033	0.039	0.058
Shelling percentage								1.000	0.114**	-0.533*	0.351*
									0.014	-0.046	0.224*
Oil content									1.000	-0.576*	0.146**
										0.102	0.092
Disease at harvest										1.000	-0.346*
											-0.330*
Pod weight per plant											1.000

* and ** indicate significance of values at P=0.05 and 0.01, respectively

higher yield. With a view to determine the extent and nature of relationship prevailing among yield contributing characters, an attempt has been made to study the character association in F_5 and F_6 material of resistant ´ susceptible crosses of groundnut both at phenotypic and genotypic levels.

Genetic correlation between different characters of plant often arises because of either linkage or pleiotropy (Horland and Csinos, 1939). Hence, the study of character association through correlation will help in selecting the yield attribute. The association between two characters can be ascertained by phenotypic correlation which is determined from measurement of two characters in a number of individual of the segregating population. In general, the genotypic correlations were higher than their respective phenotypic correlation for most of the characters indicating strong intrinsic association reduced at phenotypic level due to environment and genotype x environment interaction components. Higher values of genotypic correlation than the phenotypic correlation coefficient between the pair of characters have been reported in soybean (Johanson *et al.*, 1955).

In the present study, phenotypic and genotypic correlations were studied for pod weight per plant and its component traits in F_5 and F_6 populations. Phenotypic correlation of plant population, primary branches, test weight, shelling percentage, oil content exhibited positive significant association with pod weight per plant. Similar results were reported by Badwal *et al.* (1967), Sharma and Vershney (1990), Manoharan and Sethupathiramalingam *et al.* (1990), Nagabhushanam and Prasad (1992) for primary branches, Mishra and Yadav (1992), Sharma *et al.* (1995), Pushkaran and Nair (1993) for shelling percentage, Sarala and Gowda (1998), Nagda *et al.* (2001) for 100-seed weight positively correlated with yield. But, in all the populations, pod weight per plant was negatively correlated with plant population, plant height (except F_6), disease at harvest. The suggesting possibility of identifying and isolating genotype with lesser plant population and plant and lower disease incidence. The characters plant population, plant height, primary branches, secondary branches, leaf length, leaf width, test weight, shelling percentage, oil content, disease at harvest were not only exhibited significant association with pod weight per plant, but also showed significant positive association among themselves. This suggests that, this character should be considered while selecting plants for yield improvement. In this F_5 and F_6 population, similar results reported for pod yield by Lin (1954) and for 100-kernel weight by Venkataraman (2001). In case of F_6 population disease at 30 days was negatively correlated with plant population. Disease at 60 days was negatively correlated with plant population and disease at 30 days and disease at 90 days was negatively correlated with plant population, disease at 30 days and disease at 60 days. This indicates the disease incidence leads to reduction in plant population and yield and yield components.Hundred seed weight was positively

Table 2 : Genotypic and phenotypic correlations of summer 2009

Characters	PP	PWP	SP	TW	OC	D 30	D 60	D 90	DAH
Plant population (PP)	1.000	-0.702**	0.352**	-0.139	-0.035	-0.563**	-0.690**	-0.613**	-0.116
		-0.288**	0.060	-0.021	0.020	-0.217**	-0.249**	-0.241**	-0.200**
Pod weight per plant (PWP)		1.000	-0.174**	0.048	-0.160*	-0.029	0.049	-0.032	0.179**
			-0.011	0.035	-0.138	-0.025	0.047	-0.028	0.040
Shelling percentage (SP)			1.000	-0.402**	-0.016	-0.167**	-0.041	0.015	-0.058
				0.069	0.057	-0.084	-0.026	0.003	0.098
Test weight (TW)				1.000	-0.295**	0.044	0.014	-0.029	-0.497**
					-0.078	0.009	0.006	-0.011	0.001
Oil content (OC)					1.000	-0.044	-0.078	0.113	0.133
						-0.042	-0.047	0.082	0.012
Disease at 30 days (D 30)						1.000	-0.178**	-0.224**	-0.009
							-0.173**	-0.222**	-0.273**
Disease at 60 days (D 60)							1.000	-0.300**	-0.458**
								-0.294**	-0.400**
Disease at 90 days (D90)								1.000	-0.100
									-0.308**
Disease at harvest (DAH)									1.000

* and ** indicate significance of values at P=0.05 and 0.01, respectively

Table 3 : Percentage of superior segrgeants over TAG-24 and R-9227

Characters	TAG 24	R 9227	Number of lines	% of superior segregants
Test weight	42.56	46.28	19	11.52
Shelling percentage	65.30	69.52	8	4.85
Oil content	44.68	41.85	21	12.73
Pod weight per plant	14.18	10.74	26	15.76
Disease at harvest	42.17	26.88	6	3.60

correlated with shelling percentage. Similar results were observed by Ramanathan and Raman (1968). Test weight had significant positive correlation with shelling per cent and oil content. Similar results were reported for shelling per cent by Ramanathan and Raman (1968) and Sangha (1973), disease at harvest was positively correlated with plant population, whereas negatively correlated with plant height, number of primary branches, oil content and pod yield per plant. Similar results were reported by Nagaraja (2003). Eventhough, the segregating populations can be usually assessed using their mean and variance. These parameters alone will not give complete merit of different population. The ultimate worth of a population can mainly be judged by the superiority of derived recombinants over the existing varieties for different characters. The segregants which equaled or exceeded the means of best parent *i.e.*, TAG 24 and R 9227 for test weight, shelling percentage, oil content, pod weight per plant, disease at harvest were considered important in the present study (Table 3).

In the study, the superior segregants of TAG 24 x R

Table 4 : Superior TAG 24 × R 9227 segregants identified for test weight, shelling percentage, oil content, pod weight per plant and disease at harvest

Lines/ parents	Test weight	Lines/ parents	Shelling percentage	Lines/ parents	Oil content	Lines/ parents	Pod weight per plant	Lines/ parents	Disease at harvest
TAG-24	42.56	TAG-24	65.30	TAG-24	44.68	TAG-24	14.18	TAG-24	42.17
R-9227	46.28	R-9227	69.52	R-9227	41.85	R-9227	10.74	R-9227	26.88
162	46.36	134	69.52	146	44.72	38	14.58	21	19.79
73	46.44	82	70.06	41	44.74	110	14.60	77	20.04
47	46.69	61	70.57	65	44.76	14	14.77	109	22.04
163	46.69	137	70.72	8	44.78	136	15.06	25	24.72
137	46.95	148	71.08	115	44.82	10	15.21	165	24.92
50	47.11	64	71.40	153	44.83	8	15.33	36	25.92
138	47.15	21	72.41	92	44.86	66	15.35		
160	47.27	20	75.65	121	44.89	49	15.45		
78	47.62			90	44.94	154	15.59		
152	47.86			55	44.95	4	15.93		
28	47.94			129	44.97	72	16.23		
142	48.59			49	45.12	132	16.45		
74	48.87			39	45.31	130	16.68		
63	48.88			140	45.31	128	16.73		
136	49.54			82	45.36	91	16.88		
38	50.46			118	45.70	2	17.14		
9	50.48			135	45.70	135	17.33		
116	50.73			89	45.77	121	18.33		
161	51.48			104	45.87	160	18.95		
				4	45.92	158	19.14		
				117	45.94	124	20.45		
				122	46.06				
				127	46.30				
				116	46.42				
				9	46.54				
				2	46.93				
S.E.±	3.60		6.20		1.08		2.15		1.19
C.D. (P=0.05)	10.25		17.33		3.02		6.40		3.33
CV	12.53		15.09		2.51		9.25		14.69

9227 population were selected. The percentage of desirable segregants was rather more for the oil content in population (Table 4). This may be because of quantitative inheritance with modifier genes, which enhances the phenotypic expression of gene at loci controlling resistance (Walls and Wynne, 1985). Further, it has also been reported that the trait is controlled by recessive alleles at five different loci (Nevill, 1982) and by two complementary recessive mutant genes (Soriano, 1988). Resistance was shown to be influenced by additive genetic variance (Green and Wynne, 1986). The cross TAG 24 x R 9227 segregants gave relatively higher proportion of superior recombinant for oil content, test weight and pod weight per plant. This may be because of the cross involved parent exhibiting superiority for these characters. On the other hand, the cross TAG 24 x R 9227 produced less proportion of desirable segregants for shelling percentage and disease at harvest. This can also be owed to the performance of the parents. The available variability within the population of TAG 24 x R 9227 could be further utilized to evaluate the concealed variability in a phased manner until the uniformity is attained within the population for most of the characters under study. This would accumulate most of the genes dispersed among the genotype in a population and offer greater scope for selection.

Table 5 : Potential segregants identified for test weight, shelling percentage, oil content, pod weight per plant in TAG 24 × R 9227

Test weight(g)	Shelling percentage (%)	Oil content (%)	Pod weight per plants (g)	Disease at harvest
		2 (46.93)	2 (17.14)	
		4 (45.92)	4 (15.93)	
		8 (44.78)	8 (15.33)	
9 (50.48)		9 (46.54)		
	21(72.41)			21 (19.79)
38 (50.46)			38 (14.58)	
		49 (45.12)	49 (15.45)	
	82(70.06)	82 (45.36)		
116 (50.73)		116 (46.42)		
		121 (44.89)	121 (18.33)	
136 (50.46)			136 (15.06)	
137 (46.95)	137(70.72)			
160 (47.27)			160 (18.95)	

The population of TAG 24 x R 9227, 19 lines were observed to be superior segregants (11.52%) than both the parents for test weight and also 8 lines as superior segregant (4.85 %) in shelling percentage, 21 lines as superior segregants (12.73%) for oil content. Less number of superior segregants (3.6%) was observed in case of disease at harvest *i.e.*, 6 lines, while, 26 lines were found to be superior segregant (15.76 %) in case of pod weight per plant. Similar results were reported by Bhat (1994). Out of 165 lines, six recombinant inbred lines (21, 25, 26, 36, 109 and 165) had better disease resistance recording low disease incidence (19.79% to 25.92%). Among the six lines, the line No. 21 had less disease incidence (19.79%) in combination with shelling percentage (72.41%).

Generally, genotypes which are potential for one or more character, nevertheless in nature we do find occasionally, some genotypes resistant/tolerance to disease. The potential genotypes were tested for test weight, shelling percentage, oil content, pod weight per plant, disease at harvest (Table 5). Some of the genotypes showed higher values in many traits *i.e.*, the line No. 2, 4 and 8 showed higher values than the parent with respect to oil content, pod weight per plant and test weight. The line No. 38, 136 showed higher values in case of test weight and pod weight per plant. But, line No. 116 showed higher value in case of test weight and oil content. The genetic studies revealed quantitative inheritance with

Table 6 : Frequency distribution of superior recombinant with respect to disease resistance and productivity parameters

Combination	Superior lines	Frequency	Percentage (%)
Test weight + Shelling percentage	1	0.006	0.60
Test weight + Oil content	2	0.012	1.21
Test weight + Pod weight/plant	3	0.018	1.81
Test weight + Disease at harvest	0	0	0
Shelling percentage + Oil content	1	0.006	0.60
Shelling percentage + Pod weight/plant	0	0	0
Shelling percentage + Disease at harvest	1	0.060	0.60
Oil content + Pod weight/plant	5	0.030	3.03
Oil content + Disease at harvest	0	0	0
Pod weight/plant + Disease at harvest	0	0	0

modifier gene affecting the phenotypic expression of gene at loci controlling resistance (Walls and Wynne, 1985), where as it was reported to be controlled by recessive alleles at five loci (Nevill, 1982) and by two complimentary recessive mutant gene (Soriano, 1988). Resistance was shown to be influenced by additive genetic variance (Green and Wynne, 1986; Choroenrath *et al*., 1989). Because of the low frequency of resistant plants, Nevill (1982) opined that the selection of resistant plants in the F_2 as part of a pedigree selection programme would be inefficient. He suggested alternative procedures such as intermating the progenies of resistant segregants selected in early generation or retaining high yielding but susceptible material in bulk from which resistant forms could be selected in advanced generation.

In the present study the test weight combination like oil content, pod weight per plant (0.012, 0.018, respectively) showed moderate frequency and also the combination of shelling percentage with oil content, disease at harvest (0.60%) resulted with less frequency of superiority. But none of the combination was observed in case of shelling percentage with pod weight per plant, test weight with disease at harvest, disease at harvest with oil content and pod weight per plant. However, very high frequency was observed for oil content combined with pod weight per plant (0.03) in Table 6. Similar results were reported by Krishnakanth (1998) and Prabhu (1993). Hence this material has to be further tested in large scale and also can be utilized from development of breeding material.

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Received : May, 2010; Accepted : July, 2010