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Resistance Screening of Groundnut Advanced Breeding Lines against Collar Rot and Stem Rot Pathogens

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ABSTRACT

Forty groundnut advanced breeding lines along with susceptible checks JL-24, J-11 and TMV-2 were used for collar rot (Aspergillus niger) and stem rot (Sclerotium rolfsii) disease screening. Based on the per cent number of plants affected by the collar rot pathogen, the advanced breeding lines were categorized into four groups. The lines present in group I (Resistant) having < 15% incidence, group II (Moderately resistant) having 15.1 to 30%, group III (Susceptible) having 30.1 to 45 % and group IV (Highly susceptible) having > 45% incidence. Similarly among 40 breeding lines only three lines (ICGV86699, ICGV91114 and ICGV 89280) have shown stem rot disease reaction below 3 (up to 25 % plants were symptomatic) and considered to be moderately resistant to stem rot pathogen. The advanced breeding line ICGV99058 has recorded a disease reaction of 5 scale (> 50 % of the plants symptomatic) equal to the susceptible checks which is considered to be highly susceptible to stem rot pathogen.

Key words: *Screening, Advanced breeding lines, Groundnut, Stem rot, Collar rot*

INTRODUCTION

Groundnut (*Arachis hypogaea* L.) is one of the important oilseed crops grown in India. Major groundnut growing states of India include Andhra Pradesh, Telangana, Gujarat, Karnataka and Tamil Nadu. Of these, Andhra Pradesh, Telangana and Gujarat contribute to more than half the crop area in the country [6]. Groundnut cultivation in India as a rainfed crop is often subjected to significant yield losses annually due to biotic and abiotic stresses are the major limiting factors for attaining high productivity in India. Of them, stem rot caused by *Sclerotium rolfsii* Sacc. and collar rot caused by *Aspergillus niger* Van Tieghem are serious soil borne diseases that cause significant losses worldwide. On simultaneous infections, these diseases are responsible for yield losses ranging from 13 to 59% [12].

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Individually, collar rot can cause yield losses up to 26% [11] whereas, in stem rot the losses in pod yield range from 16 to 30% [13]. Growing resistant cultivars against collar rot and stem rot diseases is a cost-effective, sustainable strategy and ideally fits into integrated disease management. Unfortunately, high degree of resistance to these soilborne diseases is not available among cultivable germplasm. Previous reports on the commercial release of resistant varieties to these diseases available in different countries such as, C-99R [7], Florida MDR 98 [8], and Georgia-03 L [2] are available. However, identification of potential resistant sources from germplasm assumes significance for their further use in breeding programmes. Earlier reports on identification of resistance to stem rot based on greenhouse screening techniques were available[17], but differential reactions were observed by the same genotypes under controlled and field conditions. For example, the groundnut genotypes, NC 2 and NC Ac 18016 which showed resistance to *S. rolfsii* in the field were less resistant under greenhouse conditions[16]. Consistent results with respect to disease resistance under greenhouse and field conditions however are also available[1]. Reports on resistance screening to collar rot disease is however scanty**[4]**. Therefore, systematic screening of various groundnut germplasm sets such as minicore, core, and advanced breeding lines for identifying resistant sources will help in identification of elite lines with superior resistance to these diseases. Hence the present study was taken up to identify the resistance against collar rot and stem rot pathogens.

MATERIALS AND METHODS

The experiment was conducted and repeated twice during 2013 and 2014 under greenhouse conditions. Forty groundnut advanced breeding lines along with susceptible checks JL-24, J-11 and TMV-2 were included in this study The test plants were grown in plastic pots (5" diameter with sterilized soil) under greenhouse conditions and the temperature was adjusted to 26 ± 2°C. Pots were arranged in a RCBD fashion with three replications and 5 pots per replication with three plants per pot.

Collar rot: Pots were inoculated at three days before sowing with collar rot pathogen (*A. niger*) multiplied on sorghum grains. Forty grams of sorghum grains containing pathogen was applied to the soil at 10 cm below the surface layer and covered with soil. After inoculation, the pots were watered daily to maintain soil moisture. The soil was collected from ICRISAT groundnut fields and was used in a ratio of 1part sand and 2 parts soil. There were three seeds per pot for each accession and were sown equi-distantly at a depth of 4 cm. Observations on per cent disease incidence[3] was recorded at 60 days after sowing (DAS) as follows;

$$\text{Per cent Disease Incidence (PDI)} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 100$$

Stem rot: stem rot pathogen, *S. rolfsii* that was multiplied on sorghum grains was added to the pots at 45 DAS. The pots were immediately watered for two consecutive days after inoculation. The pots were maintained on a greenhouse bench at 26±2 ^{0} C and at RH 90% until harvest. Stem rot disease severity was measured for every 15 days starting from the day of inoculation. Stem rot disease severity was measured on 1-5 scale according to **(18)** wherein, 1= Healthy plant; 2= Lesions on stem only; 3= Up to 25% of the plants symptomatic (wilted, dead or decaying); 4=26-50% of the plants symptomatic and 5=> 50% of the plants symptomatic.

The following Advanced breeding lines were selected for the present study

1 **Spanish bunch with medium duration:** ICGV 03042, 06100, 89280 and ICGS 44.
2 **Spanish bunch with medium duration and Foliar disease resistance:** ICGV 99058, 99072, 00162, 00187, 00189,

00191, 00201, 00202, 00203, 00206, 00211, 00213, 86590, 06146, 93260 and 93261

3 **Spanish bunch with medium duration and Drought resistant:** 03057, 07220, 07222, 05155, 02266, 00348, 00350 and 00351

4 **Spanish bunch with short duration**: ICGV 91114, 00308, 03042, 93468, 92195, 92035, JL-24, J-11, and TMV-2.

5 **Virginia bunch with medium duration**: ICGS 76

6 **Virginia bunch with medium duration and Foliar disease resistance:** ICGV 00241, 00246, 00247 and 86699

7 **Virginia bunch with medium duration and Drought resistant:** ICGV 87846 and ICR 48

RESULTS AND DISCUSSION

Host plant resistance is the one of the effective methods in managing the soilborne disease. Identification of resistant sources is an important factor in breeding methodology in selecting the resistant donors for incorporation of resistance. A total of 40 advanced breeding lines along with three standard checks were screened under greenhouse conditions for their resistance against collar rot and stem rot pathogens and the results are presented in the Table 1, 2 & 3.

Collar rot (*Aspergillus niger*)

Based on the per cent number of plants affected by the collar rot pathogen, the advanced breeding lines were categorized into four groups. The lines present in group I (Resistant) having < 15% incidence, group II (Moderately resistant) having 15.1 to 30%, group III (Susceptible) having 30.1 to 45 % and group IV (Highly susceptible) having > 45% incidence (Table 1).

Screening results on reaction of 40 advanced breeding lines along with three standard checks on collar rot disease in greenhouse have shown that 10 out of 40lines have shown less than 15% disease incidence and were considered as resistant. These resistant genotypes include ICGV 00202, ICGV 00211, ICGV 86590, ICGV 91114, ICGV 05155, ICGV 00350, ICGV 93261, ICGV 92195, ICGV 92035 and ICR 48. However, the differences among these lines based on the per cent collar rot incidence were not significant. A total of 19 genotypes were present under II category (moderately resistant), showing collar rot incidence in the range of 15.1 to 30% and the difference among these genotypes were not significant (Table 2). Ten genotypes were in the III category (susceptible) with percent collar rot incidence ranging from 30.1 to 45%. In IV category (Highly), the genotypes, ICGV 86699, J 11, JL 24 and TMV 2 were present and recorded the incidence of above 45%. The per cent collar rot incidences for these genotypes were 45.8, 54.7, 81.5 and 97.5 respectively.

Stem rot (*Sclerotium rolfsii*)

The results of the present study revealed that there was a gradual increase in the stem rot severity from 15 DAI to 60 DAI and the genotypes showed considerable variation to the stem rot incidence caused by *S. rolfsii*. At 15 DAI the disease severity ranged from 2 to 3.93, 2.4 to 4.7 at 30 DAI, 2.5 to 4.9 at 45 DAI and while at 90 DAI the severity was in the range 2.7 to 5.0. Of the 40 advanced breeding lines screened against stem rot of groundnut under greenhouse conditions, three lines (ICGV 86699, ICGV 91114 and ICGV 89280) have shown disease reaction of below 3 (up to 25% plant parts symptomatic) which were considered as resistant with disease scales up to 2.9 though significant differences in disease reaction were noticed in these lines (Table 3). A total of 19 lines have shown disease reaction of less than 4 considered as moderately susceptible/partially resistant. While 16 advanced breeding lines showed disease reaction of 4-5 (scale) was observed for the remaining 16 genotypes, indicating 26-50% of the plant parts with stem rot symptoms and these genotypes were considered as highly susceptible. The genotype ICGV99058 recorded a disease reaction of 5 with more than 50% of the plants showing stem rot symptoms which is considered to be a susceptible reaction and is equal to the

performance of standard susceptible checks used in the study (TMV-2 and JL-24).

Germplasm screening is an important aspect for identifying resistant lines against plant diseases. Elite lines with showing strong resistance reaction to specific diseases will further be used in breeding programmes for infusion of resistance to cultivable germplasm. Stem rot and collar rot diseases are causing significant yield losses at global level. Host plant resistance is an important component in IDM of these soil borne diseases since other options seldom offer satisfactory control. However, satisfactory levels of resistance are not available in cultivable germplasm for these diseases and the present study on identifying elite germplasm lines assumes significance. Since the pathogen *A. niger* is soilborne in nature which often limit the effective management of this pathogen. However, the cultural practices along with resistant cultivars can increase the efficiency of disease management because single method of control is not successful to control the soilborne disease [10]. An attempt was made to know the resistance of 40 advanced breeding lines along with three standard checks under the greenhouse conditions. Almost all the breeding lines were more or less infected by the *A. niger* and none of them have shown immune reaction. Compare to Virginia bunch types the breeding lines with Spanish bunch type of growth habit showed more resistance to the collar rot pathogen. Previous reports also indicate the resistance reaction of Spanish bunch types to the collar rot pathogen where they evaluated the groundnut germplasm to collar rot pathogen under field conditions. The lines C 421 and C No 1780 with Spanish bunch type growth habit showed resistance reaction to both seed rot and collar rot[8]. Similarly, screening of 734 world collection of groundnut germplasm accessions against collar rot pathogen revealed that 20 lines showed complete resistance (Zero per cent incidence) to collar rot pathogen in which eleven varieties with spreading growth habit and 7 varieties with bunchy growth habit and remaining two were under unclassified group and in the remaining germplasm the disease incidence was in the range 3.7 to 100 per cent[4].

Identification of resistant breeding lines to stem rot of groundnut caused by *S. rolfsii* could improve the efforts to select the genotypes that are resistant to stem rot. Field trials alone do not allow the selection of resistant varieties, but comparisons of field, microplot and greenhouse trials may help to identify the different components of resistance[16]. In our present studies almost all the genotypes have shown considerable attack by the stem rot pathogen and none of them were immune to the disease. Similar results were reported by earlier researchers[9]. Among the advance breeding lines which showed resistant and moderately resistant reaction, most of them were Spanish bunch type (having upright branches) growth habit and similar type of resistance was observed in Spanish bunch types against stem rot pathogen was observed by earlier workers [14]. The plant type and its growth habits in determining the incidence of *S. rolfsii* on different plant types and resistance levels of these plant types is also important while selecting the resistant lines [16]. In our present studies the checks TMV-2, JL-24 and the advanced breeding line ICGV99058 were found to be highly susceptible to the stem rot pathogen and the susceptibility of these TMV-2 and JL-24 was also reported by earlier workers [15] where they found more than 50 per cent incidence under the field conditions. In the present studies the varieties which have shown less than 15 % collar incidence under control condition can be evaluated in the field with artificial inoculated conditions and may use for evolving the desirable varieties with high resistance to collar rot pathogen *A. niger*. Similarly lines which have resistant and moderately resistant reactions to stem rot should be evaluated in the field conditions in different locations under varied environmental conditions and can be used in the resistance breeding programme. Though, germplasm screening has been a continuous process against these diseases, integrating the host-plant resistance with other sustainable options under IDM is an ideal strategy over long run.

Table 1: Grouping of groundnut advanced breeding lines based on per cent collar rot incidence

Groups	Genotype reaction	Per cent Incidence	Number of genotypes	Details of genotypes
Group I	**Resistant**	<15 %	10	ICGV 00202, ICGV 00211, ICGV 86590, ICGV 91114, ICGV 05155, ICGV 00350, ICGV 93261, ICGV 92195, ICGV 92035 and ICR 48
Group II	**Moderately resistant**	15.1-30%	19	ICGV 99058, ICGV 99072, ICGV 00162, ICGV 00187, ICGV 00189, ICGV 00203, ICGV 00206, ICGV 00241, ICGV 00246, ICGV 00308, ICGV 03042 ICGV 89280, ICGV 07220, ICGV 06146, ICGV 02266, ICGV 00351, ICGV 87846, CGV 93260 and ICGV 03057
Group III	**Susceptible**	30.1- 45%	10	ICGV 00191, ICGV 00201, ICGV 00213, ICGV 00247, ICGV 93468, ICGV 00348, ICGS 44, ICGS 76, ICGV 07222 and ICGV 06100
Group IV	**Highly Susceptible**	> 45%	4	ICGV 86699, J 11, TMV 2 and JL 24

Table 2: Screening of advanced breeding lines of groundnut for collar rot (Seedling blight) disease resistance under greenhouse conditions

Genotype	Collar rot incidene (%)
ICGV 99058	22.1 (**27.96)
ICGV 99072	23.2 (26.60)
ICGV 00162	28.8 (32.39)
ICGV 00187	25.7 (29.72)
ICGV 00189	26.8 (31.14)
ICGV 00191	31.9 (33.97)
ICGV 00201	31.1 (33.35)
ICGV 00202	14.9 (22.64)
ICGV 00203	23.3 (28.32)
ICGV 00206	20.4 (26.49)
ICGV 00211	12.5 (20.42)
ICGV 00213	30.6 (33.45)
ICGV 00241	16.7 (24.08)
ICGV 00246	21.6 (26.04)
ICGV 00247	30.6 (33.04)
ICGV 86590	2.8 (6.81)
ICGV 86699	45.8 (42.58)
ICGV 91114	14.5 (22.38)
ICGV 00308	18.5 (25.43)
ICGV 03042	25.8 (30.38)
ICGV 03057	18.3 (24.77)
ICGV 06100	40.5 (39.46)
ICGV 07222	41.4 (40.02)
ICGV 07220	18.8 (25.61)
ICGV 05155	11.0 (19.23)
ICGV 06146	17.0 (24.32)
ICGV 02266	23.2 (28.22)
ICGV 87846	17.6 (24.59)
ICGV 93468	35.7 (35.49)
ICGV 00348	33.1 (35.01)
ICGV 00350	13.1 (20.55)
ICGV 00351	18.3 (24.07)
ICGV 93260	25.0 (29.94)
ICGV 93261	8.3 (16.77)
ICGV 89280	18.0 (24.23)
ICGV 92195	9.9 (18.08)
ICGV 92035	12.4 (20.32)
ICGS 44	30.3 (33.28)
ICGS 76	35.0 (34.78)
ICR 48	13.2 (20.96)
JL 24	81.5 (64.96)
J 11	54.7 (47.77)
TMV 2	97.5 (83.52)
CD at 5 % SE(d) SE(m) CV%	16.87 8.33 5.89 27.69

**Values in the parenthesis are angular transformed values and are means of three replications

Table 3: Screening of advanced breeding lines of groundnut for stem rot disease resistance under greenhouse conditions

Groundnut genotype	* Days after inoculation of *Sclerotium rolfsii* and Stem rot severity rating			
	15 days	30 days	45 days	60 days
ICGV 99058	3.73	4.80	4.9	5.0
ICGV 99072	2.93	3.62	4.0	4.4
ICGV 00162	3.93	4.67	4.7	4.8
ICGV 00187	3.68	4.03	4.5	4.7
ICGV 00189	3.18	4.30	4.4	4.5
ICGV 00191	3.27	3.93	3.9	4.4
ICGV 00201	3.29	4.04	4.5	4.7
ICGV 00202	3.75	4.53	4.5	4.7
ICGV 00203	2.87	3.20	3.4	3.8
ICGV 00206	2.83	3.45	3.3	4.3
ICGV 00211	2.73	3.00	3.5	3.7
ICGV 00213	2.85	3.68	3.8	4.1
ICGV 00241	2.87	2.98	3.0	3.0
ICGV 00246	3.42	4.11	4.1	4.1
ICGV 00247	2.92	3.47	3.8	3.7
ICGV 86590	2.20	2.80	3.0	3.2
ICGV 86699	2.00	2.53	2.8	2.9
ICGV 91114	2.00	2.80	2.8	2.8
ICGV 00308	2.33	3.20	3.3	3.7
ICGV 03042	2.00	3.17	3.3	3.9
ICGV 03057	2.58	3.13	3.4	3.9
ICGV 06100	2.00	2.50	3.0	3.3
ICGV 07222	2.33	3.33	3.3	3.7
ICGV 07220	3.47	4.00	4.3	4.5
ICGV 05155	2.80	3.73	3.9	3.7
ICGV 06146	2.60	3.40	3.5	3.8
ICGV 02266	3.20	3.87	3.9	4.2
ICGV 87846	3.67	3.67	4.0	4.3
ICGV 93468	3.68	4.47	4.5	4.6
ICGV 00348	3.53	4.20	4.3	4.4
ICGV 00350	2.40	2.87	3.1	3.4
ICGV 00351	2.47	3.50	3.8	3.9
ICGV 93260	2.75	4.00	4.2	4.1
ICGV 93261	2.82	3.12	3.2	3.3
ICGV 89280	2.27	2.53	2.8	2.9
ICGV 92195	2.13	2.67	2.9	3.1
ICGV 92035	2.33	2.73	2.9	3.1
ICGS 44	2.53	3.67	3.6	3.6
ICGS 76	2.47	3.67	3.7	3.9
ICR 48	2.20	2.93	3.1	3.1
JL 24	2.13	2.67	2.9	3.2
J 11	2.00	2.40	2.5	2.7
TMV 2	2.00	3.67	4.4	4.9
CD	0.66	0.88	0.78	0.68
SE(d)	0.33	0.44	0.39	0.34
SE(m)	0.23	0.31	0.27	0.24
CV	15.21	16.10	13.43	11.10

* Groundnut plants were inoculated 45 days after sowing

CONCLUSION

Screening of 40 advanced breeding lines against collar rot and stem rot pathogens under greenhouse conditions indicated that out of 40 advanced breeding lines ten lines were recorded less than 15 per cent collar rot incidence. Similarly among 40 breeding lines only three lines ICGVs 86669, 91114 and 89820 have recorded low stem rot diseases severity. The advanced breeding line which has recorded lowest disease reaction for both stem rot and collar rot diseases identified and which can be utilized for further infusion in resistance breeding programmes.

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