

Original Research Article

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## Field Evaluation of Early and Late Leaf Spot Diseases in Advanced Breeding Lines of Groundnut (*Arachis hypogaea* L.)

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### ABSTRACT

#### Keywords

Advance backcross  
Lines, Dendrogram,  
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Leaf spots are the most serious foliar fungal diseases of groundnut (*Arachis hypogaea* L.) World wide. Early leaf spot and late leaf spots, singly or together can cause losses in pod yield of over 50% and reduce the quality of the pod and fodder. Screening of genotypes and advance breeding lines is one of the primary objective for resistance breeding in groundnut. Present investigation was carried out to screen 33 advance breeding lines and 7 released varieties of groundnut against early and late leaf spot diseases. Early leaf spot disease scoring was done at 35 and 45 days after sowing and late leaf spot scoring was done at 75 and 85 days after sowing on 1 to 9 scale. Dendrogram were constructed based on disease scoring data of both the diseases. Score 1-3 (0 to 20% disease) was given to highly resistance lines, 4-5 for moderately resistance (21 to 50%), 6-7 for susceptible (51 to 70%) and 8-9 for highly susceptible (71 to 100%) lines. For early leaf spot 13 highly resistant, 8 moderately resistant, 4 Susceptible and 15 highly susceptible lines were identified. For late leaf spot 13 highly resistant, 7 moderately resistant, 3 susceptible and 16 highly susceptible lines were identified. Total 12 highly resistant lines having resistant to both i.e., early and late leaf spot diseases were identified. Identified resistant lines can be used as source of resistance and for hybridization programme to develop new resistant variety.

### Introduction

Groundnut (*Arachis hypogaea* L.  $2n = 40$ ) is an important legume crop rich in oil, protein, vitamins and other micronutrients (Settaluri *et al.*, 2012). It is grown nearly in 114 tropical and subtropical countries, covering area of 26.5 million ha with total production of 43.9 million tons and productivity of 1654 kg/ha in 2014 (FAO, 2017). India ranks second in

groundnut production after China, followed by USA and Nigeria. In India, it is cultivated on about 3.7 million ha with the production and productivity of 6.7 million tons and 1810 kg/ha respectively during 2015-16 (Anonymous, 2017). Groundnut production is largely affected by multiple biotic and abiotic stresses, of which the two foliar fungal diseases, early leaf spot (ELS) caused by *Passalora arachidicola* (Hori) U. Braun

(syn. *Cercospora arachidicola*) and late leaf spot (LLS) caused by *Nothopassalora personata* (Berk. & M.A. Curtis) U. Braun, C. Nakash., Videira & Crous (syn. *Cercosporidium personatum*), are most important. Both fungal diseases produce lesions (up to 1 cm in diameter) on groundnut leaves, stems, petioles, and pegs (McDonald *et al.*, 1985, Subrahmanyam *et al.*, 1989). Leaf spots damage the plant by reducing the available photosynthetic area, lesion formation, and by stimulating leaflet abscission. Shedding of infected leaves upon disease progression can lead to complete defoliation in susceptible genotypes (Singh *et al.*, 2011). Figure 1 shows severe damage to groundnut crops caused by early and late leaf spot in the Gwalior, Madhya Pradesh, India. Worldwide, yield losses range from 10% to over 50%, but vary considerably from place to place and between seasons. Yield losses are generally substantial upto 70% when the crop is attacked by both leaf spots and rust. The cultivation of resistant and tolerant groundnut varieties does not only eliminate the crop losses caused by disease, it also contributes to reduce costs related to fungicide sprayings and other control methods. Identification of resistant sources and knowledge of components and mechanism of resistance are the pre-requisite for the success of disease resistance breeding programs. Insufficient disease incidence also complicates the selection of resistant plants in field experiments (Tshilenge *et al.*, 2012, Mondal *et al.*, 2014, Pramanik *et al.*, 2019). Chaudhary *et al.*, (2019) used a set of 340 diverse peanut genotypes and screened for LLS and rust resistance and yield traits across three locations in India under natural and artificial disease epiphytotic conditions. The study revealed significant variation among the genotypes for LLS and rust resistance at different environments. Recently, Chu *et al* (2019) developed an RIL population from crossing Florida-07 × GP-NC WS 16 and

utilized this population to map QTLs associated with ELS and LLS resistance. Screening and identification of germplasm and advance breeding lines for foliar fungal diseases is one of the primary objective for resistant breeding. The present investigation was conducted for screening of foliar fungal disease early leaf spot and late leaf spot disease in groundnut at field conditions. This study reports the screened disease resistance genotypes at field conditions these genotypes can be used in breeding programme and development new variety of groundnut and application of resistance genotypes used in marker assisted selection for further rust-resistant breeding programs in groundnut.

## **Materials and Methods**

Total thirty three advance breeding lines of groundnut received from ICRISAT and six popular genotypes i.e., KDG-128 TG-26 GANGAPURI SUNOLIC 95-R ICGS-44 JGN-3 GPBD-4 were screened for leaf spot diseases during Kharif seasons 2019. Genotypes were grown in RBD with three replications at KVK, Research Farm, College of Agriculture, RVSKVV, Gwalior, Madhya Pradesh. Scoring for diseases was done at 35 and 45 days for early leaf spot and at 75 and 85 days for late leaf spot. Disease scoring was based on 1-9 scale visual score indicated as 1= highly resistance 0%, 1-3= resistance 1-20%, 4-5= moderate resistance 21-50%, 6-7= susceptible 51-70% and 8-9= highly susceptible 70-100% (ICRISAT, 1995) (Table 1). Dendrogram was constructed and Principal coordinates analysis was done using Darwin software 6.0.21 versions.

## **Results and Discussion**

### **Early Leaf Spots Disease (ELS)**

Disease scoring data for early leaf spots (ELS) was analyzed using Darwin software

and constructed dendrogram showed different groups of the groundnut genotypes (Table 1 & Table 2). Cluster analysis revealed that genotypes of groundnut under study divided into two groups' major group I and minor Groups II (Fig 2). Major group I contained 21 genotypes divided into two sub grouped "A" and "B". Sub grouped "A" contained 13 genotypes were resistant as they were having disease score 1-3 (0-20%) including, ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21, Sunolic-95R (Table 2). Sub grouped "B" contained 8 genotypes namely ABL-13, ABL-9, ABL-2, ABL-16, ABL-5, Gangapuri, ICGS-44, and ABL-1 showing moderate resistance against the early leaf spot disease having disease score was 4-5 (21-50%). Groups II divided into two sub groups "C" and "D" sub grouped "C" contained 4 genotypes *i.e.*, ABL-31, ABL-27, GPBD-4 and KDG-4 showing susceptible lines (51-70%) and disease score was 6-7. Sub groups "D" contains 15 genotypes namely ABL-14, ABL-25, ABL-28, ABL-8, ABL-11, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-26, ABL-12, ABL-7, JGN-3 and TG-26 were highly susceptible (70-100%) and disease score was 8-9.

### **Late Leaf Spots Disease (LLS)**

Characterization of advance breeding lines of groundnut was done for late leaf spot (LLS) at 75 and 85 days after sowing and data were analyzed using Darwin software and a dendrogram was constructed. Cluster analysis constructed different group on the basis of disease score data 1-9 scale. In dendrogram two groups *i.e.*, I (major) and II (minor) were formed (Fig 3). Major group I consisted 13 genotypes *i.e.*, ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21 and ABL-16 having disease score 1-3 showed resistance (0-20%) against the late leaf spot disease. Groups II divided into two sub

groups "A" and "B". Sub grouped "A" contained 19 genotypes and sub grouped "B" contained 8 genotypes. Sub grouped "A" further divided into two sub grouped "C" and "D". Sub grouped "C" contained 16 genotypes namely ABL-14, ABL-11, GPBD-4, ABL-28, JGN-3, ABL-25, TG-26, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-8, ABL-7, ABL-12 and ABL-26 were highly susceptible (70-100%) and disease score was 8-9. Sub grouped "D" contained 3 genotypes *i.e.*, ABL-31, ABL-27 and KDG-128 with disease susceptible (51-70%) and sub grouped "B" contained 7 genotypes *i.e.*, ABL-5, ABL-1, ABL-2, ABL-13, ICGS-44, SUNOLIC 95-R, and Gangapuri showed moderately resistance against late leaf spots disease (Table 1 & 3).

### **Principal coordinates analysis (PCoA)**

#### **Early Leaf Spots**

Principal coordinates analysis (PCoA) is a member of the factorial analysis family working on distance matrices. Factorial analysis methods constitute two very different approaches for the representation of data structure. Principal coordinates analysis (PCoA) of groundnut genotypes showed relationship of early leaf spots disease resistant. According to PCoA of present study, 13 genotypes were grouped together namely ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21, and Sunolic-95R showing resistance, 8 genotypes namely ABL-13, ABL-9, ABL-2, ABL-16, ABL-5, Gangapuri, ICGS-44, ABL-1 showed moderate resistant, 4 genotypes ABL-31, ABL-27, GPBD-4 and KDG-4 grouped together showing susceptible, while ABL-14, ABL-25, ABL-28, ABL-8, ABL-11, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-26, ABL-12, ABL-7, JGN-3 and TG-26 found highly susceptible against early leaf spots (Fig 4).

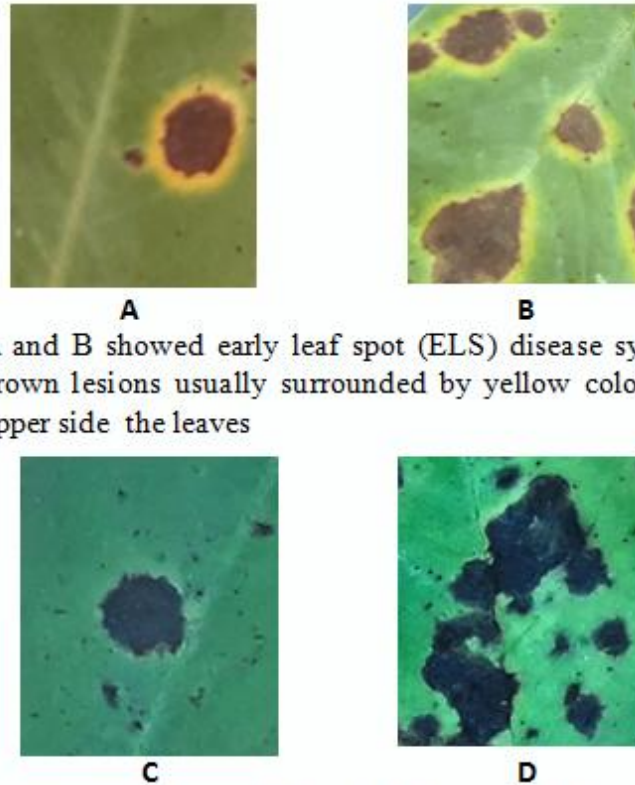
## Principal coordinates analysis (PCoA)

### Late Leaf Spots

Principal coordinates analysis (PCoA) of groundnut genotypes for late leaf spot grouped resistant (0-20%) lines including ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21 and ABL-16. Seven groundnut genotypes *i.e.*, ABL-5, ABL-1, ABL-2, ABL-13, ICGS-44, SUNOLIC 95-R, and Gangapuri showed moderate resistant (21-50%), ABL-31, ABL-27 and KDG-1 showed susceptible (51-70%), while ABL-14, ABL-11, GPBD-4, ABL-28,

JGN-3, ABL-25, TG-26, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-8, ABL-7, ABL-12 and ABL-26 showed highly susceptible (70-100%) pattern for late leaf spot.

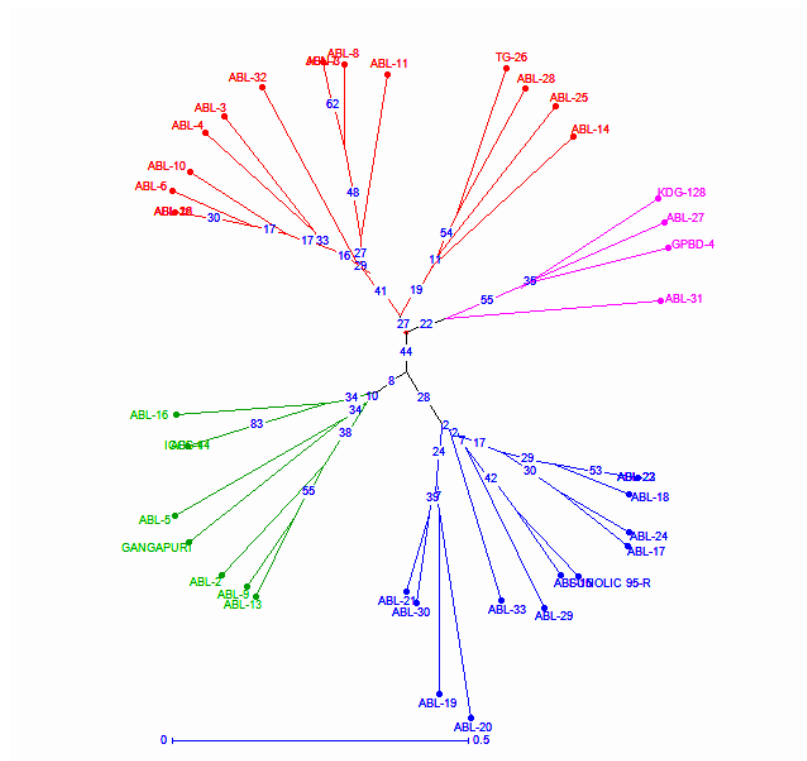
For early leaf spot 13 highly resistant, 8 moderately resistant, 4 Susceptible and 15 highly susceptible lines were identified. For late leaf spot 13 highly resistant, 7 moderately resistant, 3 susceptible and 16 highly susceptible lines were identified. Total 12 highly resistant lines having resistant to both *i.e.*, early and late leaf spot diseases were identified.



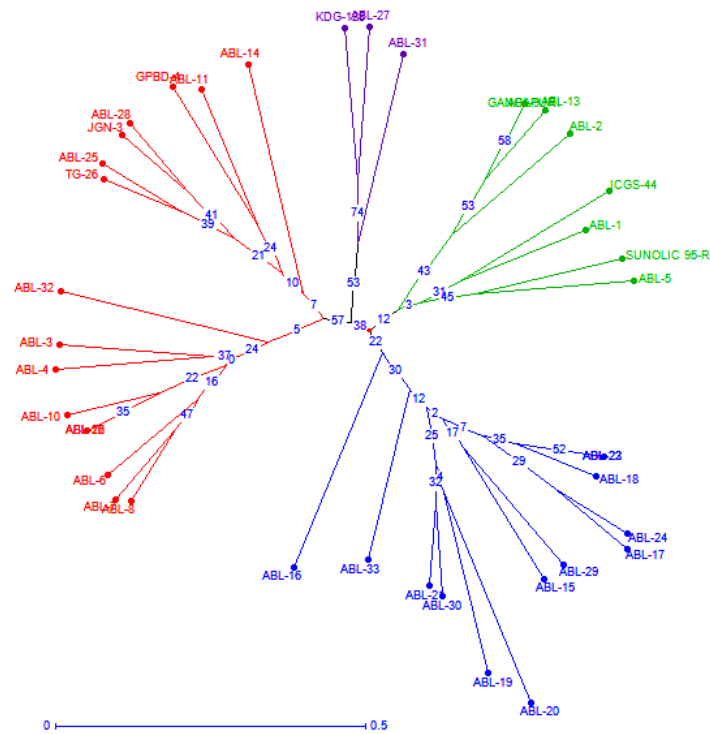
A and B showed early leaf spot (ELS) disease symptoms brown lesions usually surrounded by yellow color on the upper side the leaves

C and D showed late leaf spot (LLS) disease symptoms circular and darker spots appear on lower surface the leaves

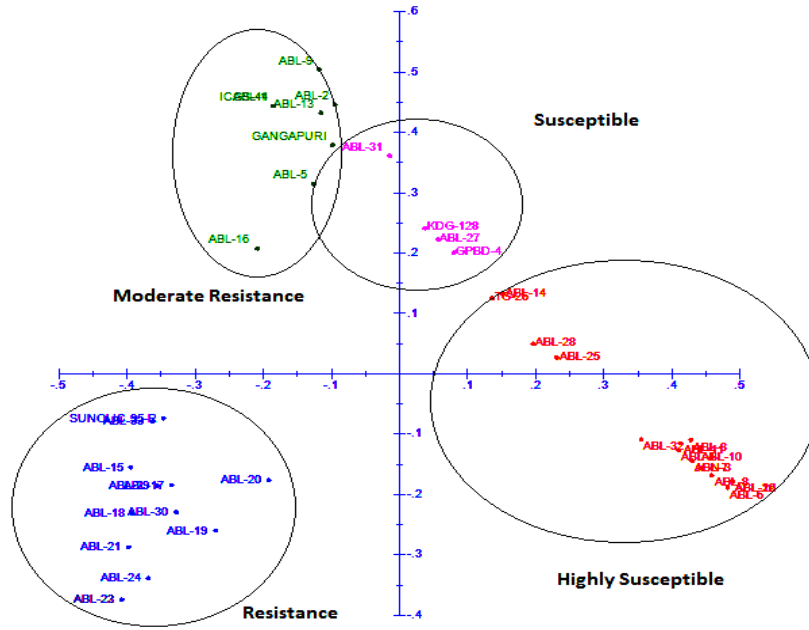
**Figure.1** Disease symptoms of early leaf spot (ELS) and late leaf spot (LLS) in advance breeding lines of groundnut



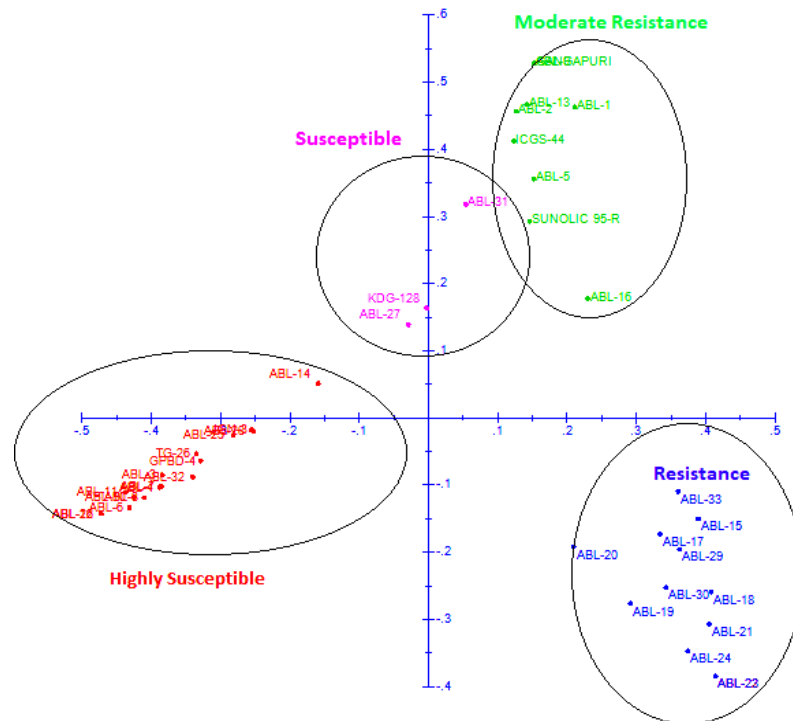
**Fig.2** Dendrogram show disease relationship of early leaf spot disease resistance in 40 groundnut (*Arachis hypogaea L*) germplasm



**Fig.3** Dendrogram show disease relationship of late leaf spot disease resistance in 40 groundnut (*Arachis hypogaea L*) germplasm



**Fig.4** Principal coordinates analysis (PCoA) early leaf spots disease relationship 40 groundnut genotypes



**Fig.5** Principal coordinates analysis (PCoA) late leaf spots disease relationship 40 groundnut genotypes

**Table 1.** Disease scoring for early leaf spot and late leaf spot data of groundnut at different time intervals

S.No.	Name of Genotypes	Disease score for early leaf spots (ELS)						Disease score for late leaf spots (LLS)					
		30 DAS			45 DAS			75 DAS			85 DAS		
		R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>
1	ABL-1	4	3	4	3	3	3	5	4	4	5	4	4
2	ABL-2	5	5	6	5	4	4	6	6	5	5	4	4
3	ABL-3	8	8	9	7	8	7	8	7	8	9	9	9
4	ABL-4	8	9	9	8	8	9	9	8	8	9	9	9
5	ABL-5	4	5	5	4	4	4	5	4	6	6	5	4
6	ABL-6	8	8	9	8	9	9	8	9	9	9	9	9
7	ABL-7	7	7	8	8	7	7	8	9	8	9	9	8
8	ABL-8	6	7	8	8	7	8	8	9	9	9	9	8
9	ABL-9	5	6	5	4	4	4	5	6	6	5	4	4
10	ABL-10	7	7	7	6	6	6	7	8	9	9	9	9
11	ABL-11	8	7	8	7	8	7	8	8	9	8	9	8
12	ABL-12	8	9	9	9	9	9	8	8	9	9	9	9
13	ABL-13	6	5	5	4	4	4	4	6	6	5	4	4
14	ABL-14	7	6	6	5	5	6	7	7	7	8	8	8
15	ABL-15	3	3	4	3	3	3	3	4	3	3	3	4
16	ABL-16	6	5	5	4	4	4	5	4	4	4	4	3
17	ABL-17	3	2	2	2	2	2	2	3	3	3	3	4
18	ABL-18	5	4	4	3	3	3	3	3	3	3	4	3
19	ABL-19	3	3	3	3	2	3	2	3	3	2	2	3
20	ABL-20	4	3	3	3	3	3	3	2	3	2	2	2
21	ABL-21	3	4	3	2	2	3	3	4	3	3	2	3
22	ABL-22	3	2	2	2	2	2	3	3	3	3	3	3
23	ABL-23	4	3	3	2	3	3	3	3	3	3	3	3
24	ABL-24	3	3	2	2	2	2	2	3	3	3	3	3
25	ABL-25	7	8	7	7	6	6	7	8	8	8	7	8
26	ABL-26	7	8	7	7	7	7	8	8	9	9	9	9
27	ABL-27	7	6	6	6	6	6	6	7	7	7	7	6
28	ABL-28	7	6	7	7	6	7	7	8	8	8	8	7
29	ABL-29	3	4	3	3	2	3	3	4	4	3	3	3
30	ABL-30	3	3	3	3	2	2	3	4	3	2	2	3
31	ABL-31	6	5	5	4	4	5	6	7	7	5	4	5
32	ABL-32	8	9	8	9	9	9	9	9	9	8	9	9
33	ABL-33	4	5	4	3	3	3	4	4	3	3	4	3
34	KDG-128	6	7	6	7	6	6	6	7	7	6	6	6
35	TG-26	7	8	7	7	8	7	8	8	8	8	7	8
36	GANGAPURI	5	5	5	6	4	4	5	6	6	5	4	4
37	SUNOLIC 95-R	4	4	3	3	3	4	6	4	4	6	5	4
38	ICGS-44	5	4	4	5	4	4	5	5	4	5	5	4
39	JGN-3	8	9	8	9	9	8	7	8	8	8	7	7
40	GPBD-4	7	7	7	7	6	6	8	8	9	8	8	7

**Table.2** Early leaf spots disease characterization of groundnut genotypes

S.No.	Name of Category	Name of Genotype
1	Highly Resistance (0%)	ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21, Sunolic-95R.
2	Moderate Resistance (21-50%)	ABL-13, ABL-9, ABL-2, ABL-16, ABL-5, Gangapuri, ICGS-44, ABL-1
3	Susceptible (50-70)%	ABL-31, ABL-27, GPBD-4, KDG-4
4	Highly Susceptible (70-100%)	ABL-14, ABL-25, ABL-28, ABL-8, ABL-11, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-26, ABL-12, ABL-7, JGN-3 TG-26

**Table.3** Late leaf spots disease characterization of 40 groundnut genotypes

S.No.	Name of Category	Name of Genotype
1	Highly Resistance (0%)	ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21 and ABL-16
2	Moderate Resistance (21-50%)	ABL-5, ABL-1, ABL-2, ABL-13, ICGS-44, SUNOLIC 95-R, Gangapuri
3	Susceptible (50-70)%	ABL-31, ABL-27, KDG-128
4	Highly Susceptible (70-100%)	BL-14, ABL-11, GPBD-4, ABL-28, JGN-3, ABL-25, TG-26, ABL-32, ABL-3, ABL-4, ABL-10, ABL-6, ABL-8, ABL-7, ABL-12 and ABL-26

Although early and LLS diseases occur naturally in all the peanut growing areas, the disease epidemic and severity is influenced by the history of leaf spot incidence, crop rotation and fungicide application (Fulmer, 2017). Early leaf spots are brown to reddish brown in color and always have yellow spots. Late leaves spots are characterized by dark brown to black spots and usually do not have yellow spots. When the condition is favorable for infection, leaf spot lesions begin to appear within 3-5 weeks after planting for ELS and about 1 month later for LLS. Since it only takes 10 to 15 days for the newly emerged lesions to sporulate, both diseases can go through many cycles of reproduction before harvest; therefore, with no fungicide applications, both leaf spot diseases can result in severe pod yield loss. In this study, the

field evaluation was conducted without fungicide applications, which created environmental conditions, highly favorable to disease incident. Chu *et al.*, (2019) also did screening for ELS and LLS and the field evaluation was conducted without fungicide applications and reported a SNP-based genetic map, consistent QTLs for resistance to ELS and LLS diseases were identified on chromosomes 3 and 5, respectively. Both ELS and LLS agents have the ability to over winter in the soil where the conidia are deposited on the debris of plant tissue. Beginning around mid-season, the pathogens progressively encroach upon peanut plants starting from the leaves closest to the ground and migrating to the upper layers of the canopy. If fungicides are not applied after the appearance of symptoms, both ELS and LLS will cause



defoliation toward the later stages of disease progression. Day *et al.*, (2016) conducted an experiments to investigate the effect of date of sowing on groundnut cultivars against late leaf spot disease severity, defoliation, frequency and size of the leaf spot which revealed that all these aspects were significantly influenced by the date of sowing and crop varieties which directly affect the pod yield and test weight.

Present study investigation was screening of genotypes against early and late leaf spot in groundnut based on 1-9 disease score in field condition. Groundnut yield is constrained mainly due to two major foliar fungal diseases as early leaf spots and late leaf spot disease affects not only seed yield but also fodder yield and quality. These diseases cause yield loss up to 70 % in the crop (Subrahmanyam *et al.*, 1985) and development of resistant cultivars is one of the best means of reducing crop yield losses from late leaf spot and also the best strategy to overcome additional cost of production (Prabhu *et al.*, 2015). The obtained results reported early leaf spots resistant lines i.e., ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21, Sunolic-95R and late leaf spots resistant genotypes i.e., ABL-22, ABL-23, ABL-18, ABL-24, ABL-17, ABL-15, ABL-29, ABL-33, ABL-20, ABL-19, ABL-30, ABL-21 and ABL-16. Further marker assisted selection will be applied for selection of resistant lines. These selected resistant lines may be used as a gene pool to obtain superior commercial types and to improve early and late leaf spot resistance in groundnut.

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