

SCREENING OF SESAME (*SESAMUM INDICUM* L.) GERMPLASMS FOR RESISTANCE AGAINST CHARCOAL ROT DISEASE CAUSED BY *MACROPHOMINA PHASEOLINA* (TASSI) GOID.

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ABSTRACT

Six commercial and 20 candidate line of sesame were screened during 2012-2013 against *M. phaseolina*. Resistance response of Sesame germplasms was determined in pots and field under natural conditions. Sick field was prepared by mixing test pathogen at the rate of 0.4% on the upper soil layer. Observation regarding resistance reaction of test germplasms was recorded by observing charcoal rot incidence at maturity using 0-9 visual rating scale. Results demonstrated variation in response of test germplasms to charcoal rot and none of the variety or line was found immune. In pot experiment seventeen varieties/lines were highly susceptible, three were moderately resistant while six varieties/lines showed susceptible reaction. On the basis of varietal response in pot experiment nine varieties were selected for screening in field experiment against *M. phaseolina*. The only line that exhibited 1-10% mortality in field was 87008, lines 87502 and 95002 were moderately resistant while 20011, black till, TS3 and 98002 showed susceptible response. The variety TH6 and candidate line 40009 exhibited more than 50% infection and were highly susceptible in their response against *M. phaseolina*

Key words: Sesame, Germplasm, *Macrophomina phaseolina*, Resistance, Charcoal rot

INTRODUCTION

Sesame (*Sesamum indicum* L.) is ancient crop. The archeological references reveal medicinal and edible importance of conventional oil seed crop of sub-continent. it is named as "til" and cultivated in Pakistan on an area of 79.8 thousand hectares with 33.4 thousands tons annual production and an average yield of 400 kg ha' (FAO, 2012). Seeds of sesame contain more than 50 % of high quality odor and color less semi-drying oil, that contains oleic and linoleic triglycerides. The presence of protein, carbohydrates, fats fibers and essential minerals has made sesame medicinally and nutritionally valuable crop for human use. Its oil strongly resists oxidative rancidity even after long exposure to air as a result of it containing endogenous antioxidants including lignin; sesamin and sesamolin and tocopherols (Akbar *et al.*, 2011; Lee *et al.*, 2008; Elleuch *et al.*, 2007;). Sesame oil provide raw material to many industries like paints, varnishes, soaps, perfumes, pharmaceuticals, and act as synergist which doubled the strength of insecticides like pyrethrum etc. (Jin *et al.*, 2001; Wang *et al.*, 2013). Sesamin also contain bactericide potential, besides this it has wide application against liver skin oral and dental ailments because of its antioxidant nature. Old civilizations of China and India were known for the use of sesame oil in mouth to treat bacteria or to relieve anxiety and insomnia (Annussek, 2001; Morris, 2002)

Sesame produced in Pakistan is not sufficient for country needs and among many other factors responsible for its low yield, one of the most common is wide range of fungal, bacterial, mycoplasma and viral diseases. Therefore for sustainable higher production of sesame, efforts should be made to integrate different cultural and management strategy to reduce the biotic stresses of the crop. Among major pathological constraints to sesame production, charcoal rot is most devastating in all the crop production zones. It is caused by *Macrophomina phaseolina*, one of the most damaging seed and soil borne pathogen in both agricultural and natural ecosystems (Mihail, 1992).The pathogen is extensively distributed in soils worldwide and have over 500 different kinds of plants as its host whereas at least 67 hosts have been recorded from Pakistan (Mirza and Qureshi, 1978; Shehzad *et al.*, 1988). Typical symptoms of charcoal rot include dark and irregular lesions on stem lower part wilting chlorosis early defoliation and finally plant death (Abwai and Pastor Corrales, 1990).There was little information available on statistics about resistance response in sesame cultivars in Pakistan. Therefore key objective of the present study was to monitor the resistance response of available sesame germplasm against charcoal.

MATERIALS AND METHODS

Collection of sesame varieties

Sesame germplasms material was collected from Oil Research Centre (ORC) of Ayub Agricultural research Institute (AARI), Faisalabad. Twenty six varieties of sesame namely, TH6, TS3, TH5 1005, 98002, 40009, 87006, 20006, 97006, 87002, 90005, 90004, 97005, 20003, L-24, , 87502, 92002, Till 90 95002,87008, Till 89 TH3, 20011, Black till, 96001, and 98002 were evaluated for resistance level against *M. phaseolina*

Green-house (pot experiment) Experiment

Inoculum preparation

Inoculum preparation of *M. phaseolina* was carried out using sorghum seeds were soaked overnight in distilled sterilized water. Boiled seeds were spread over paper towel to remove excessive moisture Seeds (300g) were filled in Poly propylene bags that were tightly closed by PVP pipe with cotton plug to devoid air. After autoclaving at 15-20 psi pressure for 20 min and proper cooling bags filled with sorghum seeds were inoculated with 9mm mycelial disc of *M. phaseolina* cut from 5 days old culture of test pathogen. After autoclaving bags were incubated for fungal growth at 28 ± 2 °C (Iqbal *et al.*, 2014).

Soil inoculation

Soil sterilized with 10% formalin was filled in 8X12 cm earthen pots. The pot soil was inoculated with *M. phaseolina*, thoroughly mixed at the upper layer of soil ten days before sowing. Inoculum of test pathogen was mixed at the rate of 4 g /kg. Pots were watered after sterile sand spread over to facilitate viability and establishment of pathogen.

Pot experiment

Screening of sesame germplasms was conducted in sesame growing season during 2013 and 2014 at field area of Institute of Agricultural Sciences, University of the Punjab. Sesame seeds before planting were surface sterilized by immersing in 1 % NaOCl for three minutes, washed several times with sterile water and dried aseptically. Ten seeds from different sesame varieties were sown separately in each pot. Experiment was arranged in a randomized complete block manner with three replicates for each variety/line.

Field experiment

Resistance level in selected sesame cultivars and test lines was studied in fields infested with *M. phaseolina*. Sick plots were infested with *M. phaseolina* at the rate 40g/m². Seeds of nine varieties selected from their expression of stable resistance in pot trial were sown in sick field with 45cm row to row and 30cm plant to plant distance. Field trial was also performed using a (RCBD) with three replications for each treatment. Irrigation times fertilizer dose, and plant protection measures against insect pests, were performed at proper times according to recommended agricultural practices for sesame production.

Data collection

Data regarding % disease incidence due to *M. phaseolina* was determined using following formula (0-9). Resistance response of test germplasm was determined using (0-9) disease rating scale. Response of test germplasm/lines in their resistance reaction against charcoal rot pathogen was observed and on the basis of calculated percentage disease incidence sesame cultivars were classified into six categories 0 = immune 1 = highly resistant 3= resistant 5= moderately resistant 7= susceptible 9= highly susceptible. Disease incidence was recorded by the given formula of Iram *et al.* (2003).

$$\text{Disease incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Total number of plants}} \times 10$$

RESULTS AND DISCUSSION

Resistance in host germplasms along with other cultural practices is considered one of the important effective strategy for the management of diseases in different crops (Salari *et al.*, 2012). In this context pot and field investigation was conducted during 2012-13 to screen sesame germplasm for resistance to *M. phaseolina*. Sick field was prepared by adding Pathogen in upper layer of pot and field soil. Sesame plants were regularly monitored for %

diseases incidence and data regarding disease parameter (% disease incidence and severity) was recorded at maturity using 0-9 severity rating scale (Irum *et al.*, 2003). Results demonstrated that sesame germplasms exhibited significantly $P < 0.05$ variable response in their resistance. Previously screening of different host crops against *M. phaseolina* proved that resistance in host crops is limited (Haseeb *et al.* 2013; Anis *et al.*, 2011; Iqbal *et al.*, 2010; Khan and Shuaib 2007; Thiyagu, *et al.*, 2007; El-Bramawy and Wahid 2006). In current investigation on screening of sesame no variety or line was found immune or disease free both in pots and field trials. In pot assay resistance level exhibited by test germplasm ranged from 5-9. On the basis of recorded cumulative resistance reaction during screening in pots, Till 90, TS3, Till 89, TH6, 97006, 1005, 90005, 40009, 20006, 87006, 87002, 98002, 90004, L-24, 97005, 92002, 20003, were found as highly susceptible. Three candidate lines 87502, 95002, 20011 exhibited moderately resistant reactions. Black till, 87008, TH3, TH5, 96001 and 98002 exhibited susceptible response (Table 1). Nine Sesame varieties/lines which showed resistance reaction in pot experiment were further screened against *M. phaseolina* in field. Screening in field exhibited that sesame germplasm/line 87008 showed 1-10% mortality and was grouped under resistant category whereas 87502 and 95002 were moderately resistant. Sesame varieties/line (germplasms) which falls in susceptible category was 20011, blacktill, TS3 and 98002. The varieties which exhibited highly susceptible reaction with more than 50% infection were TH6 and 40009 (Table 2). The results revealed that none of the variety/ line was immune against charcoal rot, except 87008 that exhibited resistant response in field trial. Therefore, it can be concluded that development of resistance in sesame against *M. phaseolina* along needs new and improved cultural management strategies.

Table1. Reaction of *Sesamum indicum* germplasms against charcoal rot disease in pot experiment under natural environmental conditions.

Disease severity scale (0-9)	Varieties	Percent disease incidence	Cultivar response category
0	Nil	0	Immune
1	Nil	1-10	Highly Resistant
3	Nil	11-21	resistant
5	87502, 95002, 20011	21-26	Moderately resistant
7	87008, Black till, TH3, TH5, 96001 and 98002	26-50	Susceptible
9	TH6, TS3, 1005, 40009, 87006, 20006, 97006, 87002, 90005, 90004, Till 90, 97005, L-24, 20003, 92002, 4001, Till 89	<50	Highly susceptible

Table2. Susceptibility reaction of test germplasms against Charcoal rot disease in field conditions.

Disease severity scale (0-9)	Varieties	Percent disease incidence	Cultivar response Category
0	Nil	0	Immune
1	Nil	1-10	Highly Resistant
3	87502	11-21	resistant
5	95002, 20011,	21-26	Moderately resistant
7	87008, blacktill, TS3, 98002	26-50	Susceptible
9	TH6, 40009	<50	Highly susceptible

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