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**EFFECT OF CHARCOAL ROT (*M. PHASEOLINA*) ON YIELD OF SUNFLOWER
 (*HELIANTHUS ANNUUS* L.)**

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ABSTRACT

The yield potential of forty six different advanced lines/germplasm of sunflower (*Helianthus annuus* L.) was investigated at College of Agriculture (COA), Sargodha under artificial stress of charcoal rot (*Macrophomina phaseolina*). Head diameter (cm), number of achene per head and total yield (kg h⁻¹) of individual advanced line/germplasm of sunflower was further recorded. The yield potential of individual advance line/germplasm of sunflower were given the priority as it is the ultimate demand of the farmers. The results revealed that the two advanced lines/germplasm of sunflower HA-259 (2494.4 kg h⁻¹) and HA-65 (1857.6 kg h⁻¹) with highest yield potential were selected for induction of resistance and these advanced lines/germplasms also showed good result under the artificial inoculation condition with 2098.7 kg h⁻¹ and 1827.2 kg h⁻¹ respectively. The range of head diameter (cm) was recorded as 7.9 to 24.3 cm without artificial inoculation while it reduced up to 7.2 to 23.8 cm under stress condition of charcoal rot. Maximum 1120.66 number of achene per head was recorded in HA-259 while 17584 were exhibiting the minimum 620.77 without artificial inoculation. The head diameter, number of achene per head plays a significant role in the yield of sunflower. The selection of appropriate advanced line/germplasm with higher yield under charcoal rot stress condition would contribute for the yield improvement of sunflower.

Keywords: Sunflower, yield, head diameter, number of achene per head.

INTRODUCTION

The sunflower (*Helianthus annuus* L.) belongs to the family *Asteraceae* (Basu *et al.*, 2014; Talukder *et al.*, 2019) and originated from United States (Yadava *et al.*, 2012). 9 million tons of sunflower were produced annually in the world (Ali *et al.*, 2011). The sunflower oil is on 4th position among important vegetable oil (Fernández-Martínez *et al.*, 2004). Almost 50% of the sunflower world production is produced by India, Ukraine, Argentina and Russian Federation (Vollmann & Rajcan, 2009; Seiler & Gulya, 2016). During the year 1960's, the sunflower was first time introduced in Pakistan (Ullah *et al.*, 2010). It is included in major oilseed crops with rapeseed/mustard, cotton and canola (GOP, 2018). Pakistan is still a deficit in vegetable oil

production (Iqbal *et al.*, 2018). The production of sunflower fits in the local cropping system and it is getting an important position as a cash crop of Pakistan (Shah *et al.*, 2013; Iqbal *et al.*, 2018). The contribution of local production in edible oil consumption is about 12% (0.431 million tonnes) and remaining 88% (3.191 million tonnes) is imported from different countries with heavy costs about 320.893 billion (US\$ 3.063 billion) to the national economy (GOP, 2018). About 13 thousand acre reduction in sunflower area was recorded in 2018 and 11 thousand tones reduction in sunflower yield was attributed due to different biotic and abiotic factors (GOP, 2018). Many pests and pathogens are responsible in low yields of sunflower (Mirza & Beg, 1983). Among them, charcoal rot caused significant yield in all over the word (Hamid & Jalaluddin, 2007; Khan & Shuaib, 2007; Khalili *et al.*, 2016) and getting an alarming threat to this crop (Qamer *et al.*, 2018; Qamer *et al.*, 2019). In Pakistan, charcoal rot was first time reported in the sunflower growing areas of Faisalabad (Mirza, 1984) and also recorded in the NWFP and Sindh provinces as a major threat to sunflower crop sunflower (Mirza & Beg, 1983; Steven *et al.*, 1987). *M. phaseolina*

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can cause disease in 500 cultivated plant species (Khan, 2007). To improve the yield potential of sunflower, adapt such strategies that increase the production (Rashid *et al.*, 2016). The objective of this study was to determine the yield potential of different advanced lines in the presence and absence of *M. phaseolina* and impact of charcoal rot disease on agronomical characters of sunflower.

MATERIALS AND METHODS

A total forty six sunflower lines were sown at College of Agriculture (COA), University of Sargodha during November 2016. Among all these, 41 were susceptible and remaining were moderately resistant lines in the previous experiment. From single varieties/lines, 20 sunflower seeds were sown with 75 cm row to row (R x R) and 45 cm plant to plant (P x P) distance. The Experiment was conducted in a randomized complete block design (RCBD) and replicated three times. The earthing was performed at a height of one foot. All the agronomical practices were conducted properly on time. Mass culture of *M. phaseolina* isolate MP₁₂ (GenBank accession No. MH277017) was prepared on millet grains and inoculum was spread at the time of seed bed preparation with the ratio of 3-4 g per meter (Shukla *et al.*, 2015). For spore suspension viable sclerotia from seven days old pure fungal culture of MP₁₂ were transferred to 250 mL of distilled water and mixed in haring blender at very low speed. The advanced lines were inoculated after 60 days from emergence (Keerio *et al.*, 2014). A parallel field of same germplasm of sunflower was also maintained which was inoculated with sterile millet grains with same plant to plant and row to row distance. Spore suspension was added after 60 days from emergence. In both experiments, the optimum cultural requirements from planting to harvesting were same to determine the expression of genetic potential of sunflower under inoculated and un-inoculated conditions. The data regarding various agronomic characters viz; number of achene per head and head diameter (cm) were recorded from the each row of individual germplasm of sunflower. The yield of both the set of experiments (with and without inoculum) was recorded to compare the yield (kg h⁻¹) under disease free condition and induction of disease artificially. The mean of each parameter was compared by the Tukey test ($p \leq 0.05$) in R statistical software (Heldwein *et al.*, 2014).

RESULTS AND DISCUSSION

In the present study, it was planned to investigate the yield potential of available germplasm under stress and stress free condition of charcoal rot. All the agronomical practices were conducted in both experiments where it required, as water stress can influence charcoal rot incidence (Mayek-Perez *et al.*, 2002). The typical symptoms of charcoal rot were observed in artificially inoculated advanced lines while no symptoms were

recorded in control. The infected plants showed early maturity, reduced head size and fewer numbers of grains setting. The variation in disease symptoms depends upon the climatic conditions of the area, inoculum pressure, soil type and genetics in host germplasm (Khan, 2007). *Macrophomina phaseolina* has marked effect on different growth and yield parameter of forty six germplasm.

Head diameter (cm)

The impact of charcoal rot on head diameter was studied with and without inoculum on forty six germplasm of sunflower during 2016 (Table 1). The range of head diameter was varied from 7.9 to 24.3 without stress of charcoal rot while it reduced up to 7.3 to 23.8 in the presence of inoculum under field conditions. The maximum (24.5 cm) head diameter was recorded in HA-259 without inoculum and 23.4 cm reduction was observed. The statistical mean comparison of head diameter confirmed that there was a significant difference in all germplasm.

According to the findings of Ahmad *et al.*, 2005, the maximum head size was recorded in hybrid sunflower cultivar (18.30 cm) while minimum was 15.37 cm in the absence of disease. Similar trend was observed in the study of Ali *et al.*, (2011) with maximum head diameter 19.87 cm while 16.12 cm minimum was computed. Present findings was in accordance with the investigation of Jalil *et al.*, (2014) who confirmed that head diameter always reduced under stress condition of charcoal rot and reduction in head diameter will ultimately effect on the yield, also conclude that there was a significant differences among artificially inoculated and un-inoculated plants of sunflower. The reduction in head diameter of sunflower was recorded in water deficient soil (Buriro *et al.*, 2015). Similar Findings was given by Youssef *et al.*, 2017 that head diameter of sunflower suppress in saline stress soil. An increase in plant population also reduces the sunflower head size (Khan *et al.*, 2018). Sunflower head size increased by the application of rizolex fungicide against pathogen, while reduction was recorded in the presence of charcoal rot without application of fungicide (Ahmed *et al.*, 2018). Abiotic factors were also responsible in the reduction of head size of sunflower (Soomro, *et al.*, 2015; Jamro *et al.*, 2018; Asghar *et al.*, 2019).

Total number of achene per head

According to mean comparison of data charcoal rot showed negative influence on number of achene per head in forty six sunflower germplasm (Table 2). Results indicate that noticeable increased (1120.7) in number of achene was found in germplasm HA259 in the absence of fungus while it was reduce (1010.4) in the presence of charcoal rot. Lowest amount of number of seeds per head (632.7-624.9) was computed in germplasm B-291 with and without inoculum respectively.

Table 1: Mean comparison of head diameter (cm) of different sunflower germplasms after inoculation with *M. phaseolina*.

Sr. no.	Germplasm	Head diameter (cm)	
		Without Inoculation	With inoculation
1	B-224	21.31 b	20.82 b
2	17545	10.12 e	9.88 cd
3	17547	7.94 i	7.73 i-m
4	17548	8.79 e-i	8.15 g-m
5	17549	9.29 e-i	8.16 g-m
6	17550	9.76 e-g	9.08 d-j
7	17551	9.88 e-g	9.57 d-g
8	17552	10.22e	9.79 c-e
9	17553	9.68 e-h	9.36 d-h
10	17554	10.10 e	9.63 c-g
11	17555	9.65 e-h	8.92 d-j
12	17561	9.54 e-h	8.89 d-l
13	17565	9.91 ef	9.73 c-f
14	17566	9.37 e-i	8.51 d-m
15	17568	9.86 e-g	9.27 d-i
16	17569	9.53 e-h	9.10 d-j
17	17572	9.98 ef	9.77 c-f
18	17573	9.54 e-h	8.24 e-m
19	17574	9.78 e-g	9.08 d-j
20	17575	8.71 e-i	7.86 h-m
21	17576	9.16 e-i	8.62 d-m
22	17577	12.89 d	11.19 c
23	17578	8.68 e-i	7.36 k-m
24	17579	9.96 ef	9.40 d-h
25	17581	9.32 e-i	8.85 d-l
26	17582	19.84 bc	19.58 b
27	17584	8.54 fghi	7.18 m
28	17585	9.95 ef	9.01 d-j
29	17586	9.75 efg	9.27 d-i
30	17587	9.80 efg	9.59 d-g
31	17588	9.78 e-g	9.19 d-j
32	17589	8.51 f-i	7.86 h-m
33	17592	9.63 e-h	9.14 d-j
34	17594	9.39 e-i	8.42 d-m
35	17595	9.87 e-g	9.39 d-h
36	17596	9.10 e-i	8.23 f-m
37	3961	10.14 e	9.39 d-h
38	3962	9.97 ef	9.82 cd
39	3965	19.90 bc	19.63 b
40	3968	9.69 e-h	9.24 d-j
41	3969	9.73 e-h	9.48 d-g
42	B-291	8.34 g-i	7.35 lm
43	B-385	8.20 hi	7.69 j-m
44	B-64	9.30 e-i	8.90 d-k
45	HA-259	24.28 a	23.79 a

Bowen & Schapaugh, (1989) observed total number of seed increased in the absence of charcoal rot in soybean and vice versa. Present study was in accordance with the findings of Ahmad *et al.*, 2005 who investigated that maximum number of achene was recorded 952.9 in hybrid sunflower cultivar while minimum was 625.4 in the absence of

pathogen results also revealed number of seed play a vital role in yield. Similar trend was observed in the study of Ali *et al.*, (2011) author recorded (1509.75) maximum number of seed in Hysun -38 with maximum 25 cm plant to plant distance. Total number of seed in one head reduction was determined in water deficient soil (Buriro *et al.*, 2015).

Table 2: Mean comparison of number of achene per head of different sunflower germplasm after inoculation with *M. phaseolina*.

Sr. no.	Germplasm	No of Achene/ head	
		Without inoculation	With inoculation
1	B-224	1054 ab	1030.22 ab
2	17545	781.67 d-f	766.10 de
3	17547	658.32 h-j	642.78 f-i
4	17548	687 e-j	663.01 e-i
5	17549	694.56 e-j	667.55 e-i
6	17550	735.11 e-j	710.67 e-i
7	17551	764.67 e-h	749.10 ef
8	17552	787.56 de	758.67 de
9	17553	761.11 e-h	734.43 e-i
10	17554	772.33 d-h	744.56 e-g
11	17555	732.56 e-j	723.21 e-i
12	17561	727.11 e-j	716.43 e-i
13	17565	767.89 e-h	753.22 ef
14	17566	702.11 e-j	704.43 e-i
15	17568	742.43 e-i	739.78 e-h
16	17569	733.89 e-j	728.43 e-i
17	17572	782.01 d-f	772 de
18	17573	693.56 e-j	687.32 e-i
19	17574	740.22 e-i	727.78 e-i
20	17575	672.43f-j	666.23e-i
21	17576	717.67 e-j	710.67 e-i
22	17577	882.78 cd	870.32 cd
23	17578	634 ij	628.78 hi
24	17579	751.56 e-h	745.56 e-g
25	17581	727.67 e-j	722.00 e-i
26	17582	981.23 bc	961.67 bc
27	17584	620.78 j	632.00 g-i
28	17585	739.21 e-i	732 e-i
29	17586	739.10 e-i	733.43 e-i
30	17587	776.10 d-g	767.67 de
31	17588	745.67 e-i	729.43 e-i
32	17589	665.89 g-j	664.01 e-i
33	17592	728.21 e-j	724.10 e-i
34	17594	700.67 e-j	690.56 e-i
35	17595	757.56 e-h	751.56 ef
36	17596	691.56 e-j	681.67 e-i
37	3961	766.32 e-h	739.67 e-h
38	3962	777.56 d-g	762 de
39	3965	971.21 bc	949.21 bc
40	3968	741.32 e-i	714.67 e-i
41	3969	769.78 d-h	743.1 e-h
42	B-291	632.67 ij	624.89 i
43	B-385	660.56 h-j	658.23 e-i
44	B-64	713.56 e-j	715.11 e-i
45	HA-259	1120.67 a	1076.78 a
46	HA-65	1021.56 ab	1010.43 ab

Yield of sunflower germplasm (kg h⁻¹)

Significant difference in yield (kg h⁻¹) was observed in all sunflower germplasm in the presence and absence of *M.*

phaseolina (Table 3). In case of germplasm HA-259 exhibited (2494.4 kg h⁻¹) the maximum yield without inoculum while yield was suppressed (2098.7 kg h⁻¹) in

the presence of pathogen. According to mean comparison minimum yield (494.2-428.1 kg h⁻¹) was recorded in germplasm 17584 with and without inoculum of charcoal rot. The HA-259 was exhibiting the maximum head

diameter (cm) and a number of achene per head in both conditions and it is one of the main reasons that it exhibited the maximum yield (kg h⁻¹) among all germplasm.

Table 3: Mean comparison of yield kg h⁻¹ of different sunflower germplasm after inoculation with *M. phaseolina*.

Sr. no.	Germplasm	Yield kg h ⁻¹	
		Without Inoculation	With inoculation
1	B-224	2205.74 b	1893.26 b
2	17545	760.87 e-i	725.83 d-f
3	17547	553.49 j-m	630.33 d-j
4	17548	580.16 i-m	542.99 f-l
5	17549	623.91 f-m	560.42 e-l
6	17550	716.10 f-k	681.97 d-i
7	17551	778.12 e-h	704.54 d-g
8	17552	803.24 ef	708.61 d-g
9	17553	759.30 e-i	692.54 d-h
10	17554	751.98 e-i	696.71 d-h
11	17555	936.18 e	773.40 d
12	17561	686.15 f-m	617.44 d-l
13	17565	764.35 e-i	737.04 de
14	17566	636.34 f-m	594.35 d-l
15	17568	700.88 f-k	669.22 d-i
16	17569	688.17 f-l	645.85 d-i
17	17572	744.84 e-j	689.59 d-h
18	17573	608.28 g-m	561.75 e-l
19	17574	705.06 f-k	662.78 d-i
20	17575	591.07 h-m	516.89 g-l
21	17576	646.47 f-m	603.09 d-l
22	17577	1427.87 d	1173.86 c
23	17578	500.56 lm	447.73 j-l
24	17579	729.62 f-k	699.69 d-g
25	17581	668.53 f-m	604.12 d-l
26	17582	1820.87 c	1743.17 b
27	17584	494.18 m	428.07 l
28	17585	690.36 f-l	649.82 d-i
29	17586	695.29 f-k	672.35 d-i
30	17587	784.38 e-g	740.45 de
31	17588	701.54 f-k	650.42 d-i
32	17589	558.04 j-m	491.98 i-l
33	17592	698.57 f-k	659.70 d-i
34	17594	648.39 f-m	584.25 d-l
35	17595	732.80 f-j	701.27 d-g
36	17596	616.38 f-m	562.78 e-l
37	3961	654.60 f-m	623.08 d-k
38	3962	680.17 f-m	712.27 d-f
39	3965	1784.26 c	1723.25 b
40	3968	694.81 f-k	655.25 d-i
41	3969	745.00 e-j	709.44 d-f
42	B-291	498.53 lm	436.16 kl
43	B-385	539.65 k-m	506.07 h-l
44	B-64	650.01 f-m	607.41 d-l
45	HA-259	2494.42 a	2098.71 a
46	HA-65	1857.59 c	1827.23 b

Yield of cluster bean (*Cyamopsis tetragonoloba*) was recorded high with compost of pearl millet in the presence of charcoal rot while was observed low yield in control (Lodha *et al.*, 2002). Charcoal rot is very prevalent disease that suppress yield up to 30% in Bolivia, fifty percent in Brazil, seventy seven percent in India, in the presence of charcoal in soybean (Wrather *et al.*, 2010). Results was supported by Iqbal *et al.* (2010) who observed sixteen line susceptible against charcoal rot in black gram and found that *M. phaseolina* cause 100% yield losses in stress condition. *M. phaseolina* affect the quality cause wilting and discoloration that lead to the reduction in yield of mung bean (Zhang *et al.*, 2011). Our results were similar with the findings of (Jalil *et al.*, 2014) author studied about yield of 24 lines of sunflower and observed a reduction in yield with *M. phaseolina* than healthy plants. The reduction in yield was more evident in chick pea cultivars (Dhingani & Solanky 2016), strawberry (Bowen & Schapaugh 1989; Sánchez *et al.*, 2016), soyabean (Wrather *et al.*, 2006; Nagasubramanian *et al.*, 2018; Doubledee *et al.*, 2018) by this pathogen. *M. phaseolina* is a root inhibiting fungus and cause 25-30 % yield losses in storage and field conditions (Thiribhuvanamala *et al.*, 2018). Similarly fungus cause reduction in yield and block the vascular bundles to uptake the water (Doubledee *et al.*, 2018). Present results were also conformity with Adhikary *et al.*, 2019, who studied the impact of charcoal rot on yield parameter of *Sesamum indicum* in two consecutive years (2017-2018) and found that yield suppressed due to this notorious pathogen.

CONCLUSION

The conclusion revealed through present study that the germplasm HA-259 exhibited a great yield potential with maximum head diameter and number of achene per head under stress of charcoal rot. It is further, suggested to develop HA-259 as a variety of sunflower after confirmation of other agronomical traits.

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Contribution of Authors:

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| Muhammad Usman Ghazanfar | : | Design research, manuscript write up. |
| Misbah Iqbal Qamar | : | Conduct research, manuscript write up. |