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EVALUATION OF DIFFERENT SUGAR BEET VARIETIES AGAINST *FUSARIUM OXYSPORUM* F. SP. *BETAE* AND *ALTERNARIA ALTERNATA*

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ABSTRACT

Twelve different sugar beet varieties were used to evaluate resistance against *Fusarium oxysporum* f. sp. *betae* and *Alternaria alternata* at Sugar Crops Research Program, NARC, Islamabad during 2012. In case of *F. oxysporum* only one variety (SD-PAK-01/07) was found moderately resistant, two moderately susceptible (SD-PAK-03/06 and EB-0809), seven susceptible (SD-PAK-04/06, SD-PAK-07/07, SD-12970, EB-0618, EB-0726, Sandrina and Ernestina) and two (SD-PAK-09/07 and California) were found highly susceptible. While in case of *A. alternata* leaf spots, only two varieties were found moderately resistant (California and Ernestina), three moderately susceptible (SD-PAK-01/07, SD-PAK-09/07 and EB-0809), three susceptible (SD-PAK-04/06, EB-0618 and Sandrina) and four (SD-PAK-03/06, SD-PAK-07/07, S-12970 and EB-0726) were found highly susceptible.

Keywords: Sugar beet; variety; *Fusarium oxysporum*; *Alternaria alternata*.

INTRODUCTION

Sugar beet (*Beta vulgaris* L.) is an herbaceous dicot belonging to family *Chenopodiaceae*. It is a biennial plant completing its life cycle in two years, in its first year of growth it develops a rosette of leaves and a large fleshy root, which stores the food reserve in the form of sugar. If left to grow, it produces flowers and seed during the second year. As a sugar crop, however, it is grown annually and efforts are made for the maximization of sugar deposition in the root. In Pakistan, sugar beet has been cultivated in the NWFP for sugar production since the mid-sixties. Sugar beet was cultivated on an area of 2300 ha, with an average yield of 23.2 t/ha producing 53336 tons of beets during the year 2010-11 (FAO, 2011). *Alternaria* leaf spots (ALS) and *Fusarium* yellows of sugar beet are among the major diseases of sugar beet in Pakistan. The ALS are mostly oval spots, ranging from 0.3 to 1 cm in size and become larger rarely. These are light brown at early infection stages and get dark brown

to black brown with age. The typical concentric rings on the leaves are observed in the centre of the spots. Sometimes there is fusion between spots. In general, ALS occurs on flavescent and older leaves, but under normal conditions, the spots occur on healthy and young leaves as well, suggesting a higher aggressiveness or virulence of the pathogen. The severity of disease depends on the number of infected plants and on the degree of assimilatory surface reduction (Ragab *et al.*, 1994). Infection and disease development occurs during the winter months when the mean temperature is 7°C to 10°C, and humidity is high (Mcfarlane *et al.*, 1950).

Symptoms of *Fusarium* yellows of sugar beet include wilting of foliage, first appear on older leaves as chlorosis (yellowing) between the veins of the leaves (interveinal chlorosis) and a darkening of the rings in the taproot as the disease progresses. Plants can be affected at any stage from seedling until harvest; the majority of plant death appears to occur from seedling to the four-leaf stage of growth. When plants are removed and roots sliced in cross section, many show a yellow-brown to gray discoloration of the water-conducting,

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vascular tissues. The pathogen (*F. oxysporum* f. sp. *betae*) is a soil-borne fungus that invades water-conducting tissues of the root and grows upward into leaf petioles and stem of sugar beet. Optimum conditions for infection are a temperature of 25 - 27°C (the disease is favored by high soil temperature), combined with other stress factors such as herbicide, fertilizer, salinity damage to roots, soil compaction, moisture extremes, and poor water drainage (Schwartz *et al.*, 2003).

Keeping the importance of sugar beet and the adverse effects of ALS & Fusarium yellows of sugar beet on the crop, present research project was planned and carried out to study the resistance of sugar beet varieties against Alternaria leaf spot and Fusarium yellows under controlled conditions using artificial inoculation.

MATERIAL AND METHODS

Isolation and purification of fungal cultures: Beet leaf samples showing typical symptoms of Fusarium yellows (*F. oxysporum* f. sp. *betae*) and Alternaria leaf spots (*A. alternata*) received from Al-Moiz Sugar Mills, D. I. Khan, were brought to pathology laboratory of Sugar Crops Research Program, National Agricultural Research Center, Islamabad, Pakistan.

Leaves were first washed under running tap water to remove dust and other contaminants. Small pieces of about 2-3mm were cut from actively disease growing margins and sterilized with 1% sodium hypochlorite solution for 2 minutes and rinsed thrice in sterilized distilled water and dried on sterilized filter paper at room temperature. The tissue sections were placed on Potato Dextrose Agar (PDA) and incubated at 25±2°C for seven days. Subsequently pure cultures of pathogens were isolated and maintained on PDA.

Preparation of Soil and Filling of Bags: Sand and field soil was mixed (1:1) and a heap was made for fumigation. Cotton balls dipped in Formaldehyde solution were inserted in heap at different points and the heap was covered with black plastic sheet and left for 7 days i.e. until the smell of Formaldehyde vanished. Plastic bags (15 x 7.5 cm) were filled with fumigated soil and then brought to pathology Laboratory of Sugar Crops Research Program, NARC, Islamabad. Seeds of all twelve varieties were sown in bags after treatment with Ridomil Gold MZ 68WP (metalaxyl-M 4% + mancozeb 64%). After sowing of nursery bags were shifted to the growth chamber with 8 hours light and 16 hours dark period and a temperature range of 20-25°C. Germination was completed within a week.

Preparation of Spore Suspension: Sterile distilled water was added to plates containing 2 weeks old *F. oxysporum* f. sp. *betae* and *A. alternata* culture and the surface of the agar was scraped with a sterilized glass rod to remove hyphae and spores. The contents of several plates were poured through muslin cloth into a beaker to remove agar and large fragments of mycelium. The spore concentration was determined with a hemacytometer and adjusted to approximately 10⁴ spores per ml of *F. oxysporum* f. sp. *betae* (Hanson and Hill, 2004) and 5x 10⁵ spores per ml of *A. alternata* (Kohmoto *et al.*, 1991).

Inoculation of Test Plants: Healthy and vigorous plants at 2-3 true leaf stage were selected after 4 weeks of germination for inoculation of *F. oxysporum* f. sp. *betae*. Ten plants were inoculated with spore suspension of pathogen which was directly injected in root zones and other 10 plants were used as control. Same process was repeated for all 12 varieties used in the study.

In case of *A. alternata* after 4 weeks of germination, twenty vigorously growing plants of each variety at 2-3 true leaf stage were selected and 10 plants were sprayed with *A. alternata* spore suspension and other 10 plants were used as control and shifted to another growth chamber at same conditions. Same process was repeated for other 11 varieties.

Data collection: Plants inoculated with *Fusarium* and *Alternaria* isolates were independently examined for two weeks. A 0 to 4 rating scale described by Hanson and Hill, 2004 (where; 0 = no disease, plants green and healthy, 1 = plants slightly stunted to extremely stunted, leaves may be wilted, 2 = leaves chlorotic, necrosis at edges of leaves, 3 = crown becoming dried and brown to black in color, leaves dying, and 4 = death of the entire plant) was used in case of *F. oxysporum* f. sp. *betae*. While a 0-4 rating scale described by Abd-El-Kareem, 2007 (where: 0 = No leaf lesions, 1 = 25 % or less, 2 = 26 to 50, 3 = 51 to 75, 4 = 76 to 100 % infected leaf area) was used for rating ALS.

RESULTS AND DISCUSSION

None of twelve different sugar beet varieties showed a complete resistance against *F. oxysporum* f. sp. *betae*. However, one variety (SD-PAK-01/07) was found moderately resistant, two moderately susceptible (SD-PAK-03/06 and EB-0809); seven susceptible (SD-PAK-04/06, SD-PAK-07/07, SD-12970, EB-0618, EB-0726, Sandrina and Ernestina) and only two (SD-PAK-09/07 and California) were found highly susceptible to *F.*

oxysporum (Table 1). All of the studied sugar beet varieties showed a varied degree of resistance against *Alternaria* leaf spots. It was noted that no variety was completely resistant to ALS. However, two varieties were found moderately resistant (California and Ernestina), three moderately susceptible (SD-PAK-01/07, SD-PAK-09/07 and EB-0809), three susceptible (SD-PAK-04/06, EB-0618 and Sandrina) and four (SD-PAK-03/06, SD-PAK-07/07, S-12970 and EB-0726) were found highly susceptible to ALS (Table 2). Based on these studies we can recommend that the varieties having moderate resistance and susceptibility may be cultivated as they can escape *Alternaria* leaf spots and *Fusarium* yellows of sugar beet if appropriate phytosanitary measures and integrated disease management practices are utilized, until they are replaced by resistant varieties through breeding or introduction of exotic varieties having resistance against *Alternaria* leaf spots and *Fusarium* yellows of sugar beet.

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Table 1. Results for screening of sugar beet varieties against *Fusarium oxysporum* f. sp. *Betae*.

Varieties	Attribute
SD-PAK-01/07	Moderately Resistant
SD-PAK-03/06 and EB-0809	Moderately Susceptible
SD-PAK-04/06, SD-PAK-07/07, SD-12970, EB-0618, EB-0726, Sandrina and Ernestina	Susceptible
SD-PAK-09/07 and California	Highly Susceptible

Table 2. Results for screening of sugar beet varieties against *Alternaria alternata*.

Varieties	Attribute
California and Ernestina	Moderately Resistant
SD-PAK-01/07, SD-PAK-09/07 and EB-0809	Moderately Susceptible
SD-PAK-04/06, EB-0618 and Sandrina	Susceptible
SD-PAK-03/06, SD-PAK-07/07, S-12970 and EB-0726	Highly Susceptible

Response of each variety against *F. oxysporum* f. sp. *betae* was different as it has been reported in other studies (Hanson *et al.*, 2009).

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