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FUNGI COLONIZING DIFFERENT PARTS OF TOMATO PLANT (*LYCOPERSICON LYCOPERSICUM* (L.) KARST. IN PAKISTAN

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ABSTRACT

Tomato is vulnerable to attack by different fungal pathogens which are borne by soil, air and seed, and inflict colossal losses in production. Mycoflora associated with different parts of tomato plant was exclusively investigated for the first time in Pakistan. In all, 25 fungal species belonging to 16 genera were variably recovered and identified. Fungal species detected from leaves (*Alternaria alternata*, *A. solani*, *Phytophthora infestans*, *Septoria lycopersici*), from fruits (*Colletotrichum coccodes*, *Alternaria alternata*, *A. solani*, *Phytophthora infestans*, *Fusarium oxysporum* f.sp. *lycopersici*, *Botrytis cineria*, *F. solani*, *Rhizoctonia solani*, *Aspergillus* sp, *Rhizopus stolonifer*) and from roots were (*Fusarium oxysporum* f.sp. *lycopersici*, *F. solani*, *Rhizoctonia solani*, *Verticillium albo-atrum*) and some non-sporulating fungi. Seed-borne fungi were: *Curvularia lunata*, *Drechslera australiensis*, *Alternaria tenuissima*, *Chaetomium globosum*, *Penicillium digitatum*, *Rhizopus stolonifer* and species of *Fusarium* (*F. solani*, *F. moniliformae* and *F. oxysporum* f.sp. *lycopersici*), *Aspergillus* (*A. flavus*, *A. terreus*, *A. fumigates*, *A. niger* and *A. sulphureus*), *Mucor* and *Cladosporium* spp. *Alternaria solani* predominantly occurred on leaves and fruits (19%, 15.75%), respectively, whereas *Fusarium oxysporum* f. sp. *lycopersici* was isolated from roots to an extent of 17.50%. In case of seed-borne fungi, blotter method yielded more number of mycoflora with higher frequency over agar plate method. *Fusarium solani* and *Alternaria alternata*; the two main fungi accounted for high frequency of 15% and 11.20% in blotter test and 10.60% and 7.40% in agar plate method. *Rhizopus stolonifer* appeared in least frequency of 2.50% on tomato fruits and 1.35% on seeds of tomato. The pathogenic behaviors of all the fungal isolates were confirmed after multiplication of important pathogens and then under artificial conditions of inoculation on test plants. The study will help farmers of Southern Punjab, Pakistan to focus on appropriate management of emerging and recurrent fungal diseases of tomato.

Keywords: Detection methods, foliage, fruit, *Lycopersicon lycopersicum*, mycoflora, root and seed.

INTRODUCTION

Tomato (*Lycopersicon lycopersicum* (L.) Karst. (synm. *Lycopersicon esculentum* Mill.)) belonging to nightshade family (*Solanaceae*) is most widely consumed vegetable crop due to its savory fruit, flavor and nutritive values. Low calories, high contents of vitamin A and C, beta-carotene and potassium, presence of lycopene-a natural and powerful antioxidant that is known to reduce the risks of cancers and cardiovascular diseases associated with type 2 diabetes (Shidfar *et al.*, 2011) make tomato fruit beneficial to human health. Low cost of production and short-duration of attract tomato growers to cultivate tomato and gain high yields throughout the

year particularly in countries with warmer climates (Naika *et al.*, 2005). Diversified climatic conditions in Pakistan favor the production of good quality tomatoes throughout the year. It is cultivated on area of about 58.2 thousand hectares producing 574.0 thousand tons, Punjab alone produces 86.3 thousand tons from 6.6 thousand ha giving yield of 13.1 thousand tons per ha (GOP, 2013). Diseases are the major obstacles in crop production which often cause heavy losses ranging from minor to 100%. Tomato plant is prone to attack by numerous diseases caused by fungi, bacteria, viruses and nematodes (Agrios, 2005), but fungal pathogens constitute major cause of yield reduction (Stone *et al.*, 2000), because they attack tomato at all stages of growth and are carried by air, soil, water, seed and vector. Among these diseases, blights caused by *Phytophthora*

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infestans and *Alternaria solani*, leaf spots by *Colletotrichum* spp., wilt by *Fusarium oxysporum* f.sp. *lycopersici*, stem canker and fruit rot are the serious disease problems all over the world (Adebayo 2005, Carrillo-Fasio *et al.*, 2003). Under favorable climatic conditions, incidence of late and early blights can shoot up to 49-91% (Azam and Shah, 2003). Tomato is a perishable crop with a short shelf life and according to Fakir (2001) possesses high vulnerability to mycotic diseases. During extended storage, tomato fruits become prone to post harvest diseases caused by *Aspergillus niger*, *Fusarium oxysporum*, *F.solani*, *Penicillium* spp, *Botrytis cineria* and *Rhizopus stolonifer*.

Perveen and Ghaffar (1995) and Bhatti *et al*; (2010) have recorded *Fusarium solani*, *F. moniliformae*, *Alternaria alternata*, *Bipolaris* spp., *Curvularialunata* and *Drechslera australiensis* as main fungi on tomato seeds which cause seed-borne diseases such as pre and post emergence rot, seedling mortality and poor stand of crop in the field. The externally and internally seed-borne pathogens are present in the form of hyphae, chlamydospores, conidia and sclerotia (Agarwal and Sinclair, 1996). Considering the economic importance of seed-borne fungi, pathogen detection and screening methods have been developed and adopted by the seed pathologists in the world. Apart from these deteriorative effects, the fungal pathogens secrete a number of toxins which are lethal to humans and animals, and also affect seed metabolism at cellular level. About 300 fungal metabolites are known which contaminate more than 25% of the world cereals (Galvano *et al.*, 2001; Meah 2010). In view of the recurrent occurrence of fungal diseases on tomato and enormous losses caused by them, present investigations were carried out on the prevalence of fungi on tomato crop in typical specific production ecology in District Multan with particular emphasis on their identity.

MATERIALS AND METHODS

These studies were conducted in the Department of Plant Pathology, Bahauddin Zakariya University Multan (BZU) during 2012-2014, where laboratory and greenhouse facilities are adequately available.

Sample Collection: Tomato plants showing characteristic symptoms of fungal diseases on foliage and roots were collected at appropriate times from the open fields of irrigated tomato in the vicinity of Multan. Seed samples were collected from local tomato growers. The samples were collected in sterilized plastic bags,

tagged properly, brought to laboratory and stored in a refrigerator at 4°C until processed.

Isolation of fungi from tissue samples: Mycoflora infecting foliage and fruits was isolated by the standard isolation methods (Agrios, 2005). Infected leaf and fruit samples with typical symptoms, and apparently healthy tissue, were cut into small segments (1cm x 1cm), washed with tap water; surface sterilized with 70% ethyl alcohol for 1 minute, followed by three serial washings with sterile distilled water (Baudoni, 1988) and blot dried on sterile filter paper. Similarly, infected roots after thorough washing with tap water were rinsed with distilled water. These were cut into small pieces of 1cm size, surface sterilized with 2% sodium hypochlorite solution for 2 minutes and washed in three changes with sterile distilled water for 2-3 minutes. The excessive moisture was removed by placing them between two layers of sterilized filter paper. Ten segments of sample were aseptically placed on one of ten Petri plates each containing 20 ml PDA medium. In all, 400 segments of each sample replicated four times were processed. The plates were incubated at 25°C and observed periodically for the growth of colonies. Frequency percentage was calculated according to following formula given by (Fisher and Petrini, 1987).

$$\text{Percent frequency (\%)} = \frac{\text{Number of segments colonized by a specific fungus}}{\text{Total number of segments plated}} \times 100$$

Isolation from seed samples: Mycoflora associated with tomato seed samples, was determined by two methods as described by the International Seed Testing Association (ISTA, 1976).

Blotter test: Seeds were surface sterilized with 1% sodium hypochlorite for 2 minutes and rinsed twice with sterile distilled water. Twenty seeds were plated in each plate (9 cm diameter) lined with three layers of moistened Whattmann filter paper No.1. The plates were incubated at 25±2°C under (NUV) near ultraviolet light altered with 12 hours darkness, for seven days. Four hundred seeds at random were subjected to the standard blotter paper method (SBM). Experiment was run in quadruplicate having five plates per replication. Seeds infected with fungal colonies were counted and expressed in percentage.

Agar plate method: Four hundred seeds in four replicates of 100 seeds each are surface sterilized in 2% sodium hypochlorite solution for 2-3 minutes, 20 seeds

were placed per plate under aseptic condition. The plates are incubated for seven days at 25±2°C under alternative cycles of 12/12 h of NUV and darkness and observed using stereo-binocular microscope. Results were expressed in percentages i.e No. of infected seeds /No. of seeds assessed x 100 (Habib *et al.*, 2007).

Identification of fungal pathogens: The fungal isolates were identified morphologically on the basis of color, spore types, colony texture and other growth characteristics, with the help of keys described by Barnett and Hunter (1972), Ellis (1971) and Dhingra and Sinclair (1985). These isolates were deposited at First Fungal Culture Bank of Pakistan.

Establishment of Koch Postulates: Pathogenicity tests of predominantly occurring fungi were carried out in the green house using healthy seedlings of susceptible tomato variety Money Maker. The fungal isolates were multiplied on PDA medium and used for artificial inoculation. Tomato seedlings were raised and grown in plastic pots of 10 cm diameter filled with formalin-treated soil mixture pH 7 (soil, manure and sand, 1:1:1). Young seedlings were

used because their roots are highly susceptible to pathogens which enter more rapidly in roots due to high levels of exudates (Olivain *et al.*, 2006).

Statistical analysis: The data was statistically analyzed by Analysis of Variance (ANOVA) followed by Tukey's-Honest Significant Difference (HSD) test using the Statistix 8.1 program (Analytical Software, 2005). Standard deviation on different parameters was also applied (Steel *et al.*, 1997).

RESULTS

A total of 25 fungal species distributed in sixteen taxonomic genera were isolated from the leaves, fruits, roots and seeds of tomato (Table 1). Based on mean occurrence/distribution and severity, the isolates were further classified as regularly epiphytotic established in the area, spreading and recurrent, major, common distributed and easily detectable and more than 15-25% incidence, moderate with 10-15% incidence, minor (incidence between 2-5%), and opportunist- the pathogens prevalent but awaiting favorable conditions for infection and disease development.

Table: 1. Genera of associated fungal species on tomato isolated from Multan.

Fungal genera	Spp. No.	Major species	Importance
<i>Alternaria</i> Nees ex Wallr.	3	<i>Alternaria alternata</i> (Fr.) Keissler, <i>A. solani</i> (Ell.&Mart.) Jones & Grout, <i>A. tenuissima</i> (Nees ex Fr.) Wilt	16%, major and epiphytotic
<i>Aspergillus</i> Mich. ex Fr.	5	<i>Aspergillus flavus</i> Link. ex Fr., <i>A. terreus</i> Thom., <i>A. fumigatus</i> Link ex Fr, <i>A. niger</i> van Tiegh. <i>Aspergillus sulphureus</i>	3%, Minor
<i>Botrytis</i> Pers. ex Fr.	1	<i>Botrytis cineria</i> Pers. ex Fr.	4.5%, Minor
<i>Cheatomium</i> Kunze ex Fr.	1	<i>Chaetomium globosum</i> Kunze ex Fr.	3%, Minor
<i>Cladosporium</i> Link. ex Fr.	1	<i>Cladosporium</i> sp.	5%, Minor
<i>Colletotrichum</i> Corda	1	<i>Colletotrichum coccodes</i>	15%, Moderate
<i>Curvularia</i> Boedijn.	1	<i>Curvularia lunata</i> (Wakker) Boed.	8%, Moderate
<i>Drechslera</i> Ito.	1	<i>Drechslera australiensis</i> (Bugn.) Subram & Jain.	2.5%, Minor
<i>Fusarium</i> Link. ex Fr	3	<i>Fusarium oxysporum</i> f.sp. <i>lycopersici</i> (Sacc.) Snyder & Hansen, <i>F. moniliformae</i> Sheldon, <i>F. solani</i> (Mart.) App. ex Wallenw	14%, Major and emerging
<i>Mucor</i> Mich. ex Fr	1	<i>Mucor</i> sp.	2%, Minor
<i>Penicillium</i> Link. ex Fr.	1	<i>Penicillium digitatum</i> Sacc.	3%, Minor
<i>Phytophthora</i> de Bary	1	<i>Phytophthora infestans</i> (Mont.) de Bary	15%, Major epiphytotic
<i>Rhizoctonia</i> DC ex. Fr	1	<i>Rhizoctonia solani</i> Kuhn	6%, Moderate
<i>Rhizopus</i> Ehrenb ex Corda	1	<i>Rhizopus stolonifer</i> (Ehrenb ex Fr) Lind	2.5%, Minor
<i>Septoria</i> Sacc.	1	<i>Septoria lycopersici</i> Speng	4%, Minor
<i>Verticillium</i> Nees ex Wallr	1	<i>Verticillium albo-atrum</i>	5%, Moderate and emerging
Non sporulating	-	Assorted	0.6%, Minor

Association of fungi with seeds of tomato: Using blotter paper and agar plate methods, 17 species belonging to 10 genera were isolated from tomato seeds (Table 2). The two methods showed results with significant differences, blotter paper method revealed 17 as against 13 fungi in agar plate method. Blotter and agar plate tests yielded *Fusarium solanito* an extent of 15.0% and 10.60% respectively. Blotter paper method supported *A. alternata* by 11.20% as against 7.4% in agar plate method. On the other hand, *Aspergillus flavus* developed 10.25% and 9.65

percent frequency. *Chaetomium globosum*, *Curvularia lunata* and *Drechslera australiensis* occurred with 9.85%, 7.10% and 6.90%, respectively, in blotter but 5.70%, 5.20% and 4.50 percent frequency in agar plate method. The lowest frequency of 1.4 percent was recorded in case of *Aspergillus sulphureus* in blotter test and agar plate also registered lowest frequency in case of *Rhizopus stolonifer* (1.35%). *Fusarium moniliformae*, *Aspergillus fumigatus*, *mucor* sp. and *Aspergillus sulphureus* did not developed in the agar plate method.

Table 2. Percentage Incidence of fungi isolated from tomato seeds using blotter and Agar plate methods.

Isolated Fungi	Blotter Paper Method		Agar Plate Method	
	*Mean±SD	Range	*Mean±SD	Range
<i>Alternaria alternata</i>	11.20 ± 1.84 b	7-20	7.40 ± 2.12 b	7-20
<i>Fusarium solani</i>	15.00 ± 1.93 a	10-20	10.60 ± 2.93 a	5-18
<i>Curvularia lunata</i>	7.10 ± 0.74 c	3-18	4.50 ± 0.93 cde	4-14
<i>Drechslera australiensis</i>	6.90 ± 1.48 c	3-18	5.20 ± 1.83 cd	3-14
<i>Fusarium moniliformae</i>	3.65 ± 0.77 fgh	3-12	0.00 ± 0.00 h	-
<i>F. oxysporum</i> f.sp <i>lycopersici</i>	4.90 ± 2.21 def	2-14	3.25 ± 1.02 defg	1-9
<i>Chaetomium globosum</i>	9.85 ± 1.84 b	3-18	5.70 ± 1.51 bc	3-14
<i>Aspergillus flavus</i>	10.25 ± 0.91 b	8-17	9.65 ± 0.50 a	7-17
<i>Alternaria tenuissima</i>	5.50 ± 1.25 cde	2-14	3.20 ± 0.54 defg	2-8
<i>Aspergillus terreus</i>	4.10 ± 1.16 efg	2-12	2.20 ± 1.18 fg	1-9
<i>Aspergillus fumigatus</i>	2.35 ± 0.98 ghi	1-8	0.00 ± 0.00 h	-
<i>Aspergillus niger</i>	6.00 ± 1.17 cd	3-16	3.70 ± 0.89 cdef	2-16
<i>Mucor</i> sp	2.20 ± 0.85 hi	1-8	0.00 ± 0.00 h	-
<i>Cladosporium</i> sp	4.10 ± 0.48 efg	2-12	3.00 ± 1.10 efg	1-9
<i>Penicillium digitatum</i>	1.80 ± 0.78 i	1-8	4.70 ± 1.11 cde	3-12
<i>Rhizopus stolonifer</i>	1.65 ± 0.25 i	1-6	1.35 ± 0.10 g	1-4
<i>Aspergillus sulphureus</i>	1.40 ± 0.28 i	1-5	0.00 ± 0.00 h	-
LSD at P= 0.05		1.7707		1.9358

*Each figure is mean of four replicates along with Standard Deviation (SD). Means in each column not sharing a common letters are significantly different at P=0.05.

Mycoflora associated with tomato plant parts: Foliage, roots and fruits yielded twelve species of filamentous fungi belonging to ten genera (Table 3). The fungi that failed to sporulate were considered as sterile fungi. Among all the fungi, *Alternaria solani* was predominantly present on leaves with a frequency of 19%, followed by *Fusarium oxysporum* f. sp. *Lycopersici* (17.50%), and *Alternaria alternata* (15.75%). *Phytophthora infestans* and *Colletotrichum coccodes* appeared with a frequency of 15.25% but were statistically non-significant. Seven root infecting fungi as *Fusarium solani*, *Botrytis cineria*, *Rhizoctonia solani*, *Septoria lycopersici*, *Verticillium albo-atrum*, *Aspergillus* spp. and *Rhizopus stolonifer* were recovered in low frequencies ranging from 2.5-12.5% in 15.75%, 11.50% and 4.50% occurrence, respectively

(Table 2). Fungal species belonging to genus *Fusarium*, *Aspergillus*, *Rhizoctonia*, *Botrytis*, *Rhizopus* and *Verticillium* were not isolated from tomato leaves. Fungi infecting tomato fruits were: *A. solani*, *C. coccodes* and *P. infestans* were predominant over other isolates and statistically at par with each other. Both species of genus *Fusarium* (*F. oxysporum* f. sp. *lycopersici* and *F. solani*) and *A. alternata* showed moderate occurrence (13.75%, 10.25% and 13.0%, respectively) but *Aspergillus* spp. and *Rhizopus stolonifer* occurred in low frequencies of 3 and 2.5%, and might be occurring as saprophytic fungi. Only four fungal species were isolated from roots of tomato among which *F. oxysporum* f. sp. *lycopersici* shared 17.5% followed by *F. solani* (12.25%), *R. solani* (6.0%) and *Verticillium albo-atrum* (3.25%) of distribution (Table 3).

Table 3. Mean frequency distribution (%) of fungi isolated from various parts of tomato plant.

Fungiisolated	Plant Parts		
	Leaf	Fruit	Root
<i>Colletotrichumcoccodes</i>	0.00 ± 0.00 e	15.25 ± 1.26 a	0.00 ± 0.00 e
<i>Alternariaalternata</i>	15.75 ± 0.96 b	13.00 ± 0.82 c	0.00 ± 0.00 e
<i>Phytophthora infestans</i>	11.50 ± 0.58 c	15.25 ± 0.50 b	0.00 ± 0.00 e
<i>Alternariasolani</i>	19.00 ± 0.82 a	15.75 ± 0.96 b	0.00 ± 0.00 e
<i>Fusariumoxysporum</i> f. sp. <i>lycopersici</i>	0.00 ± 0.00 e	13.75 ± 0.96 c	17.50 ± 0.58 a
<i>Rhizoctoniasolani</i>	0.00 ± 0.00 e	0.00 ± 0.00 g	6.00 ± 1.63 c
<i>Fusariumsolani</i>	0.00 ± 0.00 e	10.25 ± 0.50 d	12.25 ± 0.50 b
<i>Botrytis cineria</i>	0.00 ± 0.00 e	10.50 ± 2.38 d	0.00 ± 0.00 e
<i>Verticilliumalbo-atrum</i>	0.00 ± 0.00 e	0.00 ± 0.00 g	3.25 ± 1.71 d
<i>Aspergillus</i> sp.	0.00 ± 0.00 e	3.00 ± 1.15 ef	0.00 ± 0.00 e
<i>Rhizopusstolonifer</i>	0.00 ± 0.00 e	2.50 ± 1.29 f	0.00 ± 0.00 e
<i>Septorialycopercisi</i>	4.25 ± 0.50 d	0.00 ± 0.00 g	0.00 ± 0.00 e
Non sporulatingfungi	0.50 ± 0.58 e	0.75 ± 0.50 g	0.25 ± 0.50 e
LSD at P= 0.05,	0.9589	1.3707	1.6793

Each figure is mean of four replicates along with Standard Deviation (SD). Means in each column not sharing a common letter are significantly different at P=0.05.

Pathogenicity of fungal isolates: All the isolated fungi obtained from diseased tomato plants proved to be pathogenic and their pathogenicity varied. Among all, *Alternaria solani* was found highly pathogenic followed by *Fusarium oxysporum* f. sp. *lycopersici*, *Colletotrichum coccodes*, *Fusarium solani*, *Alternaria alternata*, *Phytophthora infestans*, *Aspergillus flavus*, *Chaetomium globosum*, *Curvularia lunata* and *Drechslera australiensis* respectively.

DISCUSSION

Fungal diseases infecting tomato are serious and demand accurate and rapid identification of the pathogens so that control measures are initiated in time. The present and systematic study clearly revealed that different fungal pathogens invade all parts of tomato plant including seed. Seed rot, poor emergence and seedling mortality caused by seed and soil-borne pathogens, early and late blights incited by *Alternaria solani* and *Phytophthora infestans*, wilts due to *Fusarium* and *Verticillium* species, and stem and fruit rot, anthracnose and postharvest diseases appeared to be the major and emerging problems in tomato production. These observations are in full conformity with those reported by Jones *et al.*, 1991) and Afroz *et al.*, 2008). Under the local conditions, isolates of *A. solani* seem to be highly virulent; an observation already made by Derbalah *et al.*, 2011). Vlutoglou and Kalogerakis (2000) had observed that susceptibility of tomato plants increased with maturity of plants resulting into premature defoliation.

Kumar *et al.*; (2008) reported that virulent isolates of *A. solani* on tomato could range between 73.90-83.35% while avirulent or less virulent isolates up to 42.07%. Association of *Alternariaalternata*, *Verticilliumalbo-atrum* and *Fusarium solani* can predispose and intensify foliar diseases of tomato (Booth 1971; Ewekeye *et al.*, 2013). Therefore, high incidence of early blight encountered and anticipated in the tomato fields may be attributed to those factors analyzed by these scientists.

Root diseases appeared to be one of the major problems in tomato production. Cwalina-Ambroziak and Nowak (2011) isolated 15 fungi from tomato roots, mainly *Fusarium* and *Colletotrichum* species, similar to those found in this study. Soil-borne nature of *Fusarium solani* and *R. solanias* predominant fungi causing damping off, wilt and root rot diseases was documented by several workers (El-Rafaiet *et al.*, 2003; Jiskani *et al.*, 2007; Moretti *et al.*, 2008; Mandal *et al.*, 2009; Abd-El Khair *et al.*, 2011; Haggag and El-Gamal 2012). The *Fusarium* wilt pathogen can persists in soil and debris for many years, thus infecting healthy plants grown in the infested field (Scheuerell *et al.*, 2005; Ignjatov *et al.*, 2012). In addition, *F. oxysporum* survived in debris and was detected with an accuracy of 85.42% on fruits of tomato through spectral technique (Hahn, 2002). It must be mentioned here that the Pakistani soils are rich in *Fusarium* species and the pathogens are mostly opportunists.

Postharvest decay of fruits was monitored as a serious problem where *A. solani* and *C. coccodes* were the

common associates with *P. infestans* showing occurrence of 15.75, 15.25 and 15.25%, respectively. Chavan and Tawade (2012) and other scientists have reported association of various fungi in disease syndrome. Among the 14 fungal pathogens isolated by El-Katatny and Emam (2012) and 11 fungal species by Abdel-Malleket al; (1995) from rotted tomatoes, *A. alternata* was most predominant. Similarly, Feng and Zheng (2007) and Wang et al; (2008) found *A. alternata* as a most common postharvest pathogen that causes fruit rot of tomato. Kutama et al; (2007) concluded that fungal species of three genera; *Aspergillus*, *Rhizopus* and *Alternaria* are commonly accompanied with stored tomato fruits. Fontma et al; (1996) revealed that *A. solani* and *Phytophthora infestans* produced yield losses upto 67% despite regular sprays on tomato crop. While Byrne et al; (1997) recorded yield loss of 91.8% due to *Phytophthora* rot. The results obtained in this study are in complete agreement with several workers (Iqbal et al., 2003; Akhtar et al., 2004; Ali et al., 2005; Patel et al., 2005 and Wani 2011).

Seed pathology has been an extensive and interesting subject with many scientists in the world including Pakistan. The frequency of fungal species associated with seeds of different hosts greatly depends on the detection methods. Blotter paper method is internationally more popular, economical, consistent and provides reliable results (Begum and Momin 2000; Fakhr-un-nisa et al., 2006). Tomato seeds are known to harbor large number of fungi. Perveen and Ghaffar (1995) analyzed 24 seed samples from Pakistan and isolated 37 fungal species belonging to 20 genera, and a set of 17 fungal species distributed in ten genera identified in this study amply justify their findings. There are scattered reports on the association of *Fusarium* species with different seeds causing seedling blights, wilts and rots (Karim, 2005). Asha et al; (2011) reported the presence of *F. oxysporum* on seeds of a local tomato variety. Variations observed in this study may be attributed to various biotic and physical factors and type of seed as identified by Habib et al; (2007). Seeds of soybean, wheat, rice and cucurbits carried all these fungi (Shovan et al., 2008) but no potential relationship was found between fungal species isolated from tomato, except the methods of isolation (Ora et al., 2011; Hussain et al., 2013). Al-Kassim and Monawar (2000) and Chamling et al; (2011) preferred agar plate method over the standard blotter paper method, and isolated 12

fungal species by the former and 8 species by the later method without any differences in predominance. On the other hand, Dhekle and Bodke (2013) recovered 12 fungi through blotter method with predominance of *Fusarium moniliformae*, *Cladosporium* spp, *Aspergillus flavus*, *A. niger*, *A. nidulans*, *Rhizopus stolonifer*, *Curvularia lunata*, *Alternaria tenuis*, *A. nidulans* and *Drechslera* spp. Among the storage fungi on tomato, three species of *Aspergillus*; *Aspergillus flavus*, *A. niger* and *A. fumigatus*, which are also known to heavily infect hosts other than tomato (Rasheed et al., 2004; Tariq et al., 2005). Nonetheless, Bankole (1996) reported that these saprophytic fungi have no effect on germination of tomato seeds but only deteriorate quality of seed in storage.

CONCLUSION

Conclusively, *Alternaria solani* and *Fusarium oxysporum* f sp. *lycopersici* were isolated more frequently among all the fungal pathogens isolated from leaves and roots of tomato samples, whereas *F. solani* predominated seeds of tomato. Furthermore, the result of the study divulges that higher frequency of early blight and wilt causing fungi on tomato have adverse effect on its growth and production. Consequently, growers need to manage both seed borne and soil borne mycoflora to minimize crop losses and eventually increase the quality and yield of the tomato crop.

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