

IMPACT OF ORGANIC MATTER AND SOIL TYPES ON THE DEVELOPMENT OF FUSARIUM WILT OF CHILLI

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The research was conducted to find out the most significant concentration of organic matter and soil types for diminishing disease incidence on chilli. In contemporary studies, two experiments were conducted to evaluate the impact of organic manures (OM) and soil types on the development of Fusarium wilt in greenhouse in pots which are filled with infested soil, under completely randomized design (CRD). In first experiment, different levels (0.5, 1, 1.5 and 2%) of cattle and poultry manures were evaluated. Cow manure expressed 33.23, 25.20, 22.26 and 17.23% and poultry manure exhibited 38.23, 27.26, 23.30 and 19.30% disease incidence at 0.5, 1, 1.5 and 2% OM, respectively. In second experiment, two varieties of chilli pepper viz. Desi and Maxi were grown in different soil types i.e. sandy, clay and sandy loam soils. These varieties (Maxi and Desi) expressed 63.27 and 49.27 percent disease incidence respectively in sandy loam soil whereas similar varieties exhibited 36.27 and 29.57 percent disease incidence in clay soil. Similarly, these varieties showed 25.23 and 23.43 percent disease incidence in sandy loam soil. It was statistically concluded that minimum disease was expressed by cow manure at 2% OM and sandy loam soil among other soil types.

Keywords: Chilli, wilt disease, *fusarium oxysporum* f. sp. *capsici*, organic matter, soil types.

INTRODUCTION

Chilli (*Capsicum annum* L.) is an important summer vegetable crop of Pakistan. It belongs to family *Solanaceae* (Bosland and Votava, 2000; Ziino *et al.*, 2009). It was originated from South and Central America. In 1500 AD, it was introduced in South Asia and dominates the world's spice trade. Its world production was assessed to be 3.47 million tones (Hussain and Abid, 2011). In Pakistan, it is cultivated on an area of 62.7 thousand hectares and total production of 150,300 tons (Bashir *et al.*, 2016). It has high nutritional value as it contains carotene (175 µg), thiamine (0.190 mg), riboflavin (0.190 mg), niacin (0.900 mg) and vitamin C (111 mg) per 100 grams (Joshi *et al.*, 2012).

Successful production of chilli is facing numerous problems of fungal, bacterial and viral diseases owing to non-availability of disease free seeds. Among these constrains, Fusarium wilt caused by *Fusarium oxysporum* (Schlect.) Emend. Synd. and Hans. f. sp. *capsici* Riv. is the most important one, which is a destructive soil born fungal pathogen (Matthew *et al.*, 2006). In the world, this disease reduced \$65300.00 million revenue with 48 percent disease incidence and yield losses of 100kg bags/ha (Matthew *et al.*, 2006) while in Pakistan, the overall disease incidence is 21.9 percent in vegetables followed by 16.6 percent in hot pepper

and decrease the production of 115.5 to 90.5 thousand tons during 1999-2005 (Hussain and Abid, 2011).

Appearance of yellow leaves, decaying and stunting are the characteristic symptoms of Fusarium wilt of green chilli. The whole plant appears to be brown and sunken with discoloration and girdling of cankers at the base (Matthew *et al.*, 2006). Plants wilt due to pathogen attack expressed distinct morphological appearance as compared to abiotic stress. Pathogen enters through fibrous roots from soil, its existence in xylem and phloem tissues, blocks the entire vascular system of plants (El-Kazzaz *et al.*, 2008).

Microbial communities are greatly influenced by soil types. Microconidia of *Fusarium oxysporum* f. sp. *capsici* (Foc) has different modes of damage in cultivated as well as uncultivated soils owing to the change in the density of spore and soil types. Sandy loam soils have less water holding capacity which is unsuitable for fungal growth because water is the foremost component which is deficient in sandy soils as compared to clay loam soil (Latiffah *et al.*, 2009). Temperature has an adequate effect on the growth of soil borne pathogens. High temperature reduces the manufacturing of antimicrobial compounds such as capsaicinoids, capsaicine and phenolics which provide a place for pathogen attack in the absence of these antimicrobial compounds (Estrella *et al.*, 2003).

Amendments in soil organic matter (OM) not only improve physical and chemical properties of soil but also suppress the population of *Fusarium oxysporum* (Martin, 2003; Bonanomi *et al.*, 2010; Nikoli and Matsi, 2011). It was observed that application of OM in the soil reduced Fusarium wilt disease up to 80% (Rochette *et al.*, 2006; Reijs *et al.*, 2007; Darby *et al.*, 2006). Randall *et al.* (2000) used OM for the management of Fusarium wilt and found promising results but he also described that reduction in disease depends upon source and time of application of OM. That is why in present study different levels and sources of OM were evaluated against Fusarium wilt of chilli.

MATERIALS AND METHODS

Impact of soil types on Fusarium: Impact of soil factors including soil types was determined under greenhouse conditions in pots. Three types of soils i.e. sandy soil (T₁), clay soil (T₂) and sandy loam soil (T₃) were prepared. For the preparation of sandy soil 90% sand, 7% silt and 3% clay was mixed while in case of sandy loam 49% sand, 44% silt and 7% clay and for clay soil, 90% clay, 7% sand and 3% silt was mixed (Rauf *et al.*, 2007). Sandy loam soil served as control treatment. These soils were surface sterilized with formalin (1:320) (Khan *et al.*, 2016) and inoculated with ¼ petri plat of fully grown 7 days old culture of *Foc* with concentration of 1×10⁵ spores /ml (adjusted with haemocytometer) to check the influence of various soil types on disease incidence (%). Each treatment with three replications and a control was evaluated under completely randomized design (CRD).

Impact of different levels of organic matter (OM) on incidence (%) of Fusarium wilt: Impact of OM on the development of chilli wilt was evaluated under greenhouse conditions. Two different organic manures viz. cattle manure (T₁) and poultry manure (T₂) were analyzed on dry weight bases from Institute of Animal Husbandry, University of Agriculture, Faisalabad (UAF) to examine the original amount of organic matter present in the manures. In poultry manure 25.7, 6.7 and 10.1 while in cow manure 4.9, 4.5 and 1.6 g/Kg amount of NPK was determined by using Total Kjeldhal Nitrogen (TKN) Scarf apparatus (made in Quic Fit, England) and atomic absorption spectrophotometer (Z-8200, Hitachi, Japan) respectively for NPK. Different levels of manures (0.5, 1, 1.5 and 2%) after analysis and tedious mathematical calculations were adjusted and mixed in soil homogeneously (Wong *et al.*, 1983). This soil was analyzed in the Department of Soil Chemistry (UAF) for the confirmation of adjusted levels of OM. Then, soil was inoculated with actively growing culture of *Foc* @ 1×10⁶ spores/ml of distill H₂O. 1.2 kg of this soil was added in each plastic pot (17×13 cm) size. Pots that contain inoculated soil and no organic matter were considered as control treatment. These pots were kept on greenhouse bench at 0.82 % RH and 28°C temperature of greenhouse under completely

randomized design (CRD) and data of disease incidence was recorded on weekly basis.

$$D.I. = \frac{\text{No. of infected plants}}{\text{Total no. of observed plants}} \times 100$$

Table 1. Different levels of organic matter adjusted for greenhouse experiment.

Different levels of organic manure (%)	Cow Manure		Poultry Manure	
	g/1kg soil	g/1.3kg soil	g/1kg soil	g/1.3kg soil
0.5	5	6.5	2	2.6
1	10	13	4	5.2
1.5	15	19.5	6	7.8
2	20	26	8	10.4

The quantity of organic matter was calculated for pot size of 17×13 cm containing 1300g (1.3kg) soil.

Data analysis: All the statistical tests were performed using SAS/STAT statistical software (SAS Institute, 1990). Means were separated by using Fisher's protected least significant difference (LSD) procedure by taking P = 0.05% probability level (Steel *et al.*, 1997). Analysis of variance (ANOVA), interaction of different treatments and their combinations were developed by using SAS/STAT software package

RESULTS

Evaluation of different levels of soil organic matter (OM) against Fusarium wilt of chilli under greenhouse conditions: Two varieties of chilli pepper (Maxi and Desi) were grown in different manures namely cow manure, poultry manure with four different concentrations viz. 0.5%, 1%, 1.5% and 2%. It was observed that both varieties showed disease incidence of 33.23, 25.20, 22.26 and 17.23 percent in cow manure having different concentrations (0.5, 1, 1.5 and 2%, respectively). Similarly, both varieties were grown in poultry manure with similar concentrations and expressed 38.23, 27.26, 23.30 and 19.30 percent disease incidence under green house conditions as compared to control. It was statistically observed that chilli pepper varieties expressed minimum disease incidence in cow manure (33.23, 25.20, 22.26 and 17.23%) as compared to poultry manure (38.23, 27.26, 23.30 and 19.30 percent) (Table 2, Fig. 1).

Table 2. Impact of organic matter on the development of Fusarium wilt of chilli under greenhouse conditions.

Treatments	Disease incidence (%)			
	Concentrations of OM			
	0.5%	1%	1.5%	2%
Cow manure	33.23f	25.20h	22.26j	17.23l
Poultry manure	38.23 f	27.26g	23.30i	19.30k
Control	68.70d	71.40c	75.50b	78.40a
LSD	0.2864			

Mean values in a column sharing similar letters do not differ significantly as determined by LSD test (P ≤ 0.05).

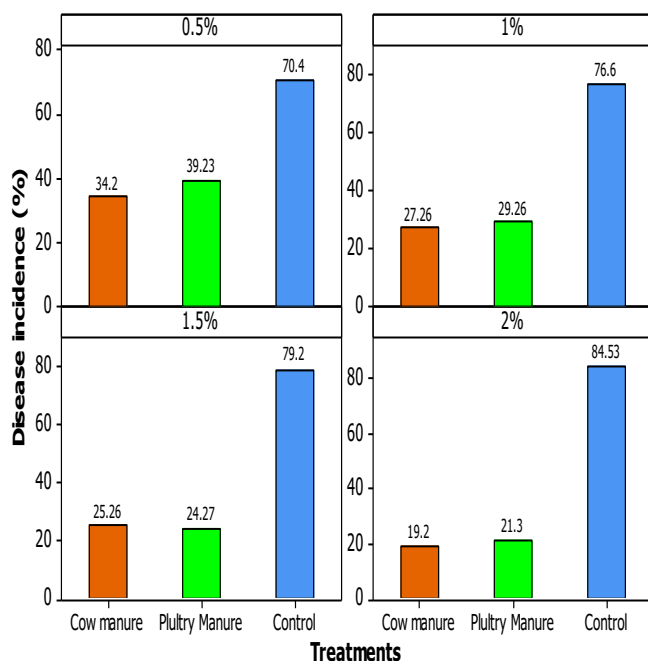


Figure 1. Impact of different organic manures on the development of Fusarium wilt of chilli .

Evaluation of soil types against Fusarium wilt of chilli under greenhouse conditions: In current experiment, different soil types i.e. sandy soil, clay soil and sandy loam soils were used as treatments. Two chilli pepper varieties viz. Desi and Maxi were grown under greenhouse conditions and the impact of treatments (sandy soil, clay soil and sandy loam soils) were statistically observed. Both varieties expressed maximum disease incidence of 63.27 and 49.27 percent sandy whereas in clay soil the disease incidence of 36.27 and 29.57 percent was seen on similar varieties of chilli pepper. Likewise, both varieties showed minimum disease incidence of 25.23 and 23.43 percent in sandy loam soil. Therefore, it was observed that sandy loam soil is statistically significant to diminish the disease incidence as compared to other treatments viz. clay soil and sandy soil (Table 3, Fig. 2).

Table 3. Impact of soil types on the development of Fusarium wilt of chilli under greenhouse conditions.

Treatments	Disease incidence (%)	
	Varieties	
	Maxi	Desi
Sandy soil	63.27a	49.27b
Clay soil	36.27c	29.57d
Sandy loam (Control)	25.23e	23.43f
LSD	0.3753	

Mean values in a column sharing similar letters do not differ significantly as determined by LSD test ($P \leq 0.05$).

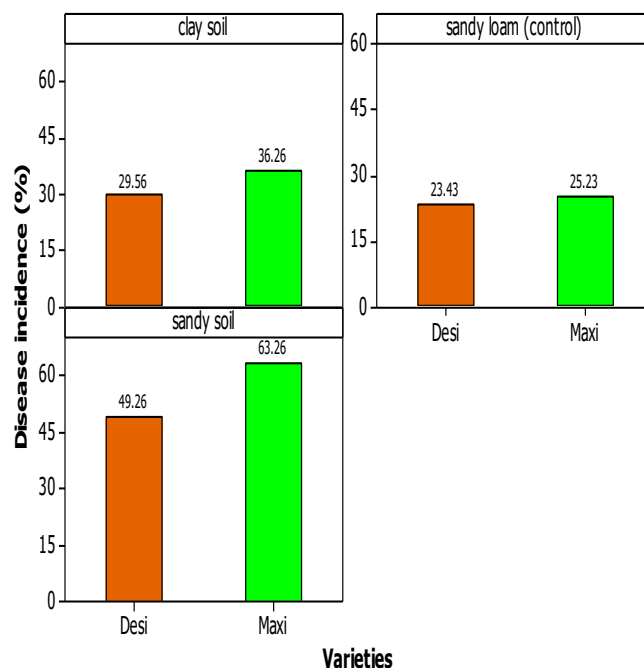


Figure 2. Impact of soil types on the development of Fusarium wilt of chilli.

DISCUSSION

Fusarium wilt caused by *Fusarium oxysporum* f. sp. *capsici* is a potential threat in chilli growing areas of Pakistan, causing 70-100% yield losses under conducive environmental conditions (Ashfaq *et al.*, 2014). Chlorosis, vascular discoloration and wilting are the characteristic symptoms of this disease (Matthew *et al.*, 2006).

Soil plays a crucial role for growth, survival, multiplication and dissemination of soil borne microorganisms because it provides a habitat for their growth and development (Chuankun *et al.*, 2004). Among these microorganisms, *Fusarium oxysporum* f. sp. *capsici* is thermophilic, ubiquitous soil borne plant pathogen which is abruptly affected by different types of soils (Ozer *et al.*, 2009) like sandy, clay and sandy loam soils. These soils possess different electric conductivity (EC), pH (alkaline and acid character), organic manure, soil texture (ratio of sand, silt and clay particles) and inhibitory volatile fungistatic compounds which consequently alters the activity of soil borne plant pathogens (Ma *et al.*, 2001).

In the current studies, sandy loam soil exhibited significant results with minimum disease incidence due to the presence of adequate proportion of soil particles (sand, silt and clay) which have specific acid and alkaline nature as compared to sandy and clay soils reduced the growth of chlamydospores, microconidia and macroconidia of *Fusarium oxysporum* f. sp. *capsici* (Attitalla *et al.*, 2011) by disturbing genetic properties like aggressiveness and virulence of *Fusarium oxysporum* f.

sp. *capsici* (Cramer, 2000). The other reasons for less disease incidence is moderate water holding capacity of sandy loam soil which is inappropriate for fungal growth (Latiffah *et al.*, 2009). Similar results were reported by Chellemi (2002), Noble and Coventry (2005) and Darby *et al.* (2006). Research revealed that application of organic matter in soil helps in improving physical (water holding capacity, aeration and nutrient uptake) and chemical properties of soil which not only enhanced crop growth but also suppress soil borne pathogens like *Fusarium oxysporum* (Bilgrami and Dube, 1976; Zhang *et al.*, 2006; Bonanomi *et al.*, 2010; Gotor *et al.*, 2014). In the contemporary studies, cow and poultry manures were used as a source of OM for management of Fusarium wilt of chilli pepper. Both sources expressed significant results but cow manure showed pronounced results in suppressing *Fusarium oxysporum* f. sp. *capsici*. Because OM not only boosts up growth rate but also increases rigidity of cell wall and resistance in plants against soil borne pathogens (Basak *et al.*, 2002). Outcomes of the present study are supported by the work of Randall *et al.* (2000) who used OM against Fusarium wilt disease and found promising results but, rate of disease inhibition depends on the source of organic matter, application method, rate, timing and weather conditions (Mattila and Joki, 2003; Rochette *et al.*, 2006; Reijs *et al.*, 2007). It has been observed that incidence of Fusarium was reduced from 50-80% through application of OM which suppressed Fusarium wilt disease by changing soil pH (Basak *et al.*, 2002; Darby *et al.*, 2006; Ha and Huang, 2007; Bustamante *et al.*, 2008).

Conclusion: Cow manure at 2% concentration is statistically significant to reduce disease incidence (17.23%) as compared to poultry manure at 0.5, 1, 1.5 and 5% concentrations. Likewise, comparatively minimum disease incidence (23.43%) was observed on sandy loam soil than sandy and clay soil.

REFERENCES

- Ashfaq, M., S. Iqbal, T. Mukhtar and H. Shah. 2014. Screening for resistance to cucumber mosaic cucurbit virus in chilli pepper. *J. Anim. Plant Sci.* 24:791-795.
- Attitalla, I.H., Z. Latiffah, B. Salleh and S. Brishammar. 2011. Biology and partial sequencing of an endophytic *Fusarium oxysporum* and plant defense complex. *Amer. J. Biochem. Mol. Biol.* 1:121-44.
- Basak, A.B., M.W. Lee and L.S. Tae. 2002. *In-vitro* inhibitory activity of cow urine and dung to *Fusarium solani* f. sp. *cucurbitae*. *Can. J. Microbiol.* 30:51-54.
- Bashir, M., N. Javed, M. Atiq and W. Wakil. 2016. Mineral profiling of chilli pepper (*Capsicum annum* L.) inoculated with *Colletotrichum capsici* (sydow), Butler and Bisby. *J. Anim. Plant Sci.* 26:1304-1312.
- Bilgrami, K.S. and H.C. Dube. 1976. A text book of modern Plant Pathology. Vikas Publishing House, New Dehli, India; pp.344.
- Bonanomi, G., V. Antignani, M. Capodilupo and F. Scala. 2010. Identifying the characteristics of organic soil amendments that suppress soil borne plant diseases. *Soil Biol. Biochem.* 42:136-144.
- Bosland, P.W. and E.J. Votava. 2000. Peppers: Vegetable and Spice Capsicums. Crop production science in horticulture series No. 12. CABI Publishing, United Kingdom.
- Bustamante, M.A., R. Moral, C. Paredes, A. Perez-Espinosa, J. Moreno-Caselles and M.D. Perez-Murcia. 2008. Agrochemical characterization of the solid byproducts and residues from the winery and distillery industry. *J. Waste Manage.* 28:372-380.
- Chellemi, D.O. 2002. Non-chemical management of soil borne pests in fresh market vegetable production systems. *Can. J. Phytopathol.* 92:1367-1372.
- Chuankun, X., M. Minghe, Z. Leming and Z. Keqin. 2004. Soil volatile fungistasis and volatile fungistatic compounds. *Soil Biol. Biochem.* 36:1997-2004.
- Cramer, C.S. 2000. Breeding and genetics of *Fusarium* basal rot resistance in onion. *Euphytica* 11:159-166.
- Darby, H.M., A.G. Stone and R.P. Dick. 2006. Compost and manure mediated impacts on soil borne pathogens and soil quality. *Soil Sci. Soc. Amer. J.* 70:347-358.
- El-Kazzaz, M.K., G.B. El-Fadly, M.A.A. Hassan and G.A.N. El-Kot. 2008. Identification of some *Fusarium* spp. using molecular biology techniques. *Egypt. J. Phytopath.* 36:57-69.
- Estrella, F.S., M.C. Vargas, M.A. Elorrieta, M.J. Lopez and J. Moreno. 2003. Temperature effect on *Fusarium oxysporum* f. sp. *melonis* survival during horticultural waste composting. *J. Appl. Microbiol.* 94:475-482.
- Gotor, T., L. Masaka and M. Sungirai. 2014. Effect of cow urine on the growth characteristics of *Fusarium lateritium*, an important coffee fungus in Zimbabwe. *Int. J. Agron.* 4:231-235.
- Ha, M.T. and J.W. Huang. 2007. Control of Fusarium wilt of asparagus bean by organic soil amendment and microorganisms. *Plant Pathol. Bull.* 16:169-180.
- Hussain, F. and M. Abid. 2011. Pests and diseases of chilli crop in Pakistan: A review. *Int. J. Biol. Biotechnol.* 8:325-332.
- Joshi, M., S. Rashmi, K.S. Anil and P. Anil. 2012. Screening of resistant varieties and antagonistic *Fusarium oxysporum* for biocontrol of Fusarium wilt of chilli. *J. Plant Pathol. Microb.* 3:5-11.
- Khan, S.A., N. Javed, S.A. Anwar, I.U. Haq, K. Naveed, Z. Ullah and A. Safdar. 2016. Survival of entomopathogenic nematodes in sterilized vs non-sterilized soil. *Pak. J. Zool.* 48:1349-1352.

- Latiffah, Z., M.I. Padzilah, S. Baharuddin and Z. Maziah. 2009. Short communication *Fusarium* species in forest soil of bird valley. Malays. J. Microbiol. 5:132-133.
- Ma, Q., J. Gan, J.O. Becker, S.K. Papiernik and S.R. Yates. 2001. Evaluation of propargyl bromide for control of barnyardgrass and *Fusarium oxysporum* in three soils. Pest Manag. Sci. 57:781-786.
- Martin, F.N. 2003. Development of alternative strategies for management of soil borne pathogens currently controlled with methyl bromide. Ann. Rev. Phytopathol. 41: 325-350.
- Matthew, A., A. Lawal, P. Chindo and B. Olalekan. 2006. Outbreak of basal stem rot and wilt disease of pepper in northern Nigeria. J. Plant Prot. Res. 46:7-14.
- Mattila, P.K. and E.T. Joki. 2003. Effect of treatment and application technique of cattle slurry on its utilization by ley: I. Slurry properties and ammonia volatilization. Nutr. Cycl. Agroecosyst. 65:221-230.
- Nikoli, T.H. and T.H. Matsi. 2011. Influence of liquid cattle manure on micronutrients content and uptake by corn and their availability in a calcareous soil. Agron. J. 103:113-118.
- Noble, R. and E. Coventry. 2005. Suppression of soil borne plant diseases with composts: A review. Biocontrol Sci. Technol. 15:3-20.
- Ozer, N., M. Koç and B. Der. 2009. The sensitivity of *Aspergillus niger* and *Fusarium oxysporum* f. sp. *cepae* to fungistasis in onion growing soils. J. Plant Pathol. 91:401-410.
- Randall, G.W., T.K. Iragavarapu and M.A. Schmitt. 2000. Nutrient losses in surface drainage water from dairy manure and urea applied for corn. J. Environ. Qual. 4:1244-1252.
- Rauf, C.A., M. Ashfaq and I. Ahmad. 2007. Management of black scurf disease of potato. Pak. J. Bot. 39:1353-1357.
- Reijs, J.W., M.P.W. Sonneveld, P. Sorensen, R.L.M. Schils, J.C.J. Groot and E.A. Lantinga. 2007. Effects of different diets on utilization of nitrogen from cattle slurry applied to grassland on a sandy soil in The Netherlands. Agric. Ecosyst. Environ. 118: 65-79.
- Rochette, P., D.A. Angers, M.H. Chantigny, B. Gagnon and N. Bertrand. 2006. In situ mineralization of dairy cattle manures as determined using soil-surface carbon dioxide fluxes. Soil Sci. Soc. Amer. J. 70:744-752.
- Steel, R.G.D., J.H. Torrie and D.A. Dickey. 1997. Principles and Procedures of Statistics: A biometrical approach, 3rd Ed. McGraw Hill Pub. Co., New York.
- Wong, M.H., Y.H. Cheung and C.L. Cheung. 1983. The effects of ammonia and ethylene oxide in animal manure and sludge on seed germination and root elongation of *Brassica parachinensis* (flowering Chinese cabbage). Environ. Pol. Ser. A. Ecol. Biol. 30:109-123.
- Zhang, M., R. Gavlak, A. Mitchell and S. Sparrow. 2006. Solid and liquid cattle manure application in a subarctic soil: Bromegrass and oat production and soil properties. Agron. J. 98:1551-1558.
- Ziino, M., C. Concetta, R. Vincenza, T. Gianluca and V. Antonella. 2009. Volatile compounds and capsaicinoid content of fresh hot peppers (*Capsicum annuum* L.) of different Calabrian varieties. J. Food Sci. Agric. 89:774-780.