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## HOST-RESPONSE OF CHICKPEA CULTIVARS TO *FUSARIUM OXYSPORUM* *F. SP. CICERIS*

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### ABSTRACT

Thirty one chickpea cultivars *viz.*, Sanyasi, D.G-89, Flip-04, D.G-92, ICCV-95469, Flip-03, NCSO-526, ICCV-92317, ICCV-73111, NCSO-521-A, NCSO-524, Flip-09, Flip-07, NCSO-521-B, ICCV-95419, Rabbat, ICCV-14, ICCV-89511, ICCV-03, ICCV-89512, ICCV-08, K-94-95, ICCV-6, NCSO-525, Flip-9037c, Flip-901576, L-550, M-30, Flip-90c, Flip-90167c and NCSO-521 were screened against *Fusarium oxysporum* f. sp. *ciceris*. No cultivar was completely immune against *F. oxysporum* f. sp. *ciceris*. One cultivar (NCSO-524) with minimum root infection (46.67%) ranges in susceptible while all the remaining cultivar ranges highly susceptible category according to used scale. Statistically there were no significant difference between root infection of NCSO-524 and NCSO-521-B, and these two cultivars caused less plant mortality i.e 50% and 36.67%, respectively. These cultivars with minimum root infection caused significantly less reduction (due to the inoculation of *F. oxysporum* f. sp. *ciceris*) in plant length and plant weight. While in most of the other cultivars there were no constant trend between the root infection and reduction in length and weight of inoculated plants lead to prove the variation in resistant and tolerance nature. Some of the cultivars which were severely infected with the test pathogen, but they showed tolerance to disease and produced better plant growth. While some cultivars badly affected and showed poor plant growth.

**Keywords:** *Cicer arietinum*, Screening, Fusarium wilt, Resistance, Tolerance.

### INTRODUCTION

Soil borne plant pathogens comprise of a very large group of devastating fungal species such as *Fusarium*, *Pythium*, *Gaeumannomyces*, *Phytophthora*, *Verticillium* and *Rhizoctonia*; all are responsible for causing numerous plant diseases of economic importance. Due to the typical survival capacity and their persistent nature, they are very difficult to control and thus become a major hindrance in achieving the goal of sustainable agriculture yields (Bonanomi *et al.*, 2007). These pathogens, once entered and established in the agro ecosystem, become well adapted and thus very problematic to control. A number of important plant diseases are caused by soil borne plant pathogens and they have remained as a major concern for more than a century of research. In developed countries like USA, they are responsible for about 90% of the major crop diseases (Lewis and Papavizas, 1991) causing losses of \$4 billion

per year (Lumsden *et al.*, 1995). In the past, methyl bromide was widely and effectively used as soil fumigation against soil borne pathogens, but a worldwide restriction on its use has further aggravated the situation (Martin, 2003). Moreover, these toxic chemicals not only increase the cost of crop production, but also harm the natural ecosystem and human, plant and animal life. Worldwide, a huge amount of about \$40 billion was spent on the purchase of 3 billion kg of pesticides in a year (PAN-Europe, 2003). In some places the indirect cost of these pesticides (resulting from indirect effect) is more than their direct cost. In USA alone the indirect cost or losses (losses in public health, crop losses, development of pesticide resistance, underground water contamination etc.) due to pesticide application are estimated to about \$9.6 billion (Pimentel and Burgess, 2014). All these drawbacks forced plant scientist to search some reliable and effective alternate control strategies of plant pathogens.

Chickpea (*Cicer arietinum* L.) is the third most important pulse crop after beans and peas (Vishwadhar and Gurha,

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1998; Saxena, 1990) as well as one of the chief sources of edible protein in both human and animal food throughout the world (Hulse, 1991; Hossain *et al.*, 2013). About 90% of the world chickpea production is concentrated in the Indian subcontinent (Juan *et al.*, 2000). Its production is badly limited by Fusarium wilt disease caused by *Fusarium oxysporum* (Schlechtend: Fr.) f. sp. *ciceris* (Padwick) Matuo & K. Sato (Jalali and Chand, 1992; Navas-Cortés *et al.*, 2000). Generally, production losses due to Fusarium wilt varied from 10-15%, depending upon the number of factors, including cultivar, cultural practices and pathogen race (Jalali and Chand, 1992, Trapero-Casas and Jiménez-Díaz, 1985), however under severe conditions complete crop loss occurred (Halila and Strange, 1996, Haware and Nene, 1980).

Due to the dynamic nature of this soil borne pathogen, it can effectively be controlled by the exploration of host plant resistance (Jalali and Chand, 1992). The use of resistant varieties is the most effective and reliable control tactics to combat the threats poses by the devastating plant pathogens. It considered a direct control tactic in integrated plant disease management. In present study chickpea germplasm are evaluated against *F. oxysporum* f. sp. *ciceris* to find out resistant cultivars as potential alternate of fungicides.

#### MATERIALS AND METHODS

In order to evaluate the varietal response of different chickpea cultivars to *F. oxysporum* f. sp. *ciceris* (*Foc*), a pot experiment was conducted at Department of Plant Pathology, Sindh Agriculture University Tandojam, in the month of November 2010. Thirty one chickpea cultivars *viz.*, Sanyasi, D.G-89, Flip-04, D.G-92, ICCV-95469, Flip-03, NCSO-526, ICCV-92317, ICCV-73111, NCSO-521-A, NCSO-524, Flip-09, Flip-07, NCSO-521-B, ICCV-95419, Rabbat, ICCV-14, ICCV-89511, ICCV-03, ICCV-89512, ICCV-08, K-94-95, ICCV-6, NCSO-525, Flip-9037c, Flip-901576, L-550, M-30, Flip-90c, Flip-90167c and NCSO-521 were used in this experiment. Seeds of each variety were surface sterilized and ten seeds were sown at 1cm depth in earthen pots containing 2kg steam sterilized soil.

Before sowing, the soil was artificially infested with the test pathogen inoculum at  $10^5$  conidia per gram of soil. The un-inoculated pots served as control. The experiment was arranged as RCBD with three replications. The data were recorded on plant mortality, plant length and plant weight as well as reduction

percent in plants growth and weight as compared to un-inoculated plants after 45 days of sowing. The following formula was used to determine reduction percent.

$$\text{Reduction (\%)} = \frac{\text{Un - inoculated plants} - \text{inoculated plants}}{\text{Un - inoculated plants}} \times 100$$

The response of chickpea varieties to pathogen infection was determined by recording root infection percentage. For this purpose roots of uprooted chickpea plants were washed, cut into small pieces, surface sterilized and placed in petri dishes containing PDA medium. The root infection percentage was calculated by following formula:

$$\text{Infection (\%)} = \frac{\text{Number of pieces colonized by the fungus}}{\text{Total number of pieces studied}} \times 100$$

The disease severity was observed on following scale:

<u>Root infection%</u>	<u>Host Response</u>
1-10%	Resistance (R)
11-20%	Moderately Resistant (MR)
21-30%	Moderately Susceptible (MS)
31-50%	Susceptible (S)
51-100%	Highly Susceptible (HS)

**Statistical Analysis:** The data were analyzed by ANOVA using Statistix 8.1 software. Least significant differences (LSD) were calculated using significant level at  $P = 0.05$ .

#### RESULTS

**Plant Mortality:** All chickpea cultivars were severely affected by the causal fungus. Among thirty one cultivars, no one found to be resistant to Fusarium wilt pathogen. Only two cultivars showed 50% or less plant mortality *i.e.* NCSO-521-B (36.67%) and NCSO-524 (50%), in remaining cultivars plant mortality were very high ranging from 60-96%. The inoculation of *Foc* caused highest plant mortality in cv. Flip-03 and ICCV-92317 followed by NCSO-521, Flip-07, NCSO-526, ICCV-95469, D.G-92 and Sanyasi (Fig. 1).

**Root Infection:** The data on root infection revealed that no chickpea cultivar evaluated during the present study has a strong resistance against the *Foc*, as all genotypes more or less susceptible to the test fungus. The significantly lowest root infection was recorded in cultivar NCSO-524 (46.67%) and NCSO-521-B (53%) followed by Flip-04, ICCV-95469 and NCSO-525 as compared to the other cultivars. The maximum root infection by *Foc* was recorded from the roots of cv. ICCV-92317, ICCV-14, Flip-9037c, L-550 and Flip-90c (Fig. 2). Except two cultivars (NCSO-524 and NCSO-521-B), all other chickpea cultivars severely infected with *Foc* and showed 76-100% root infection.

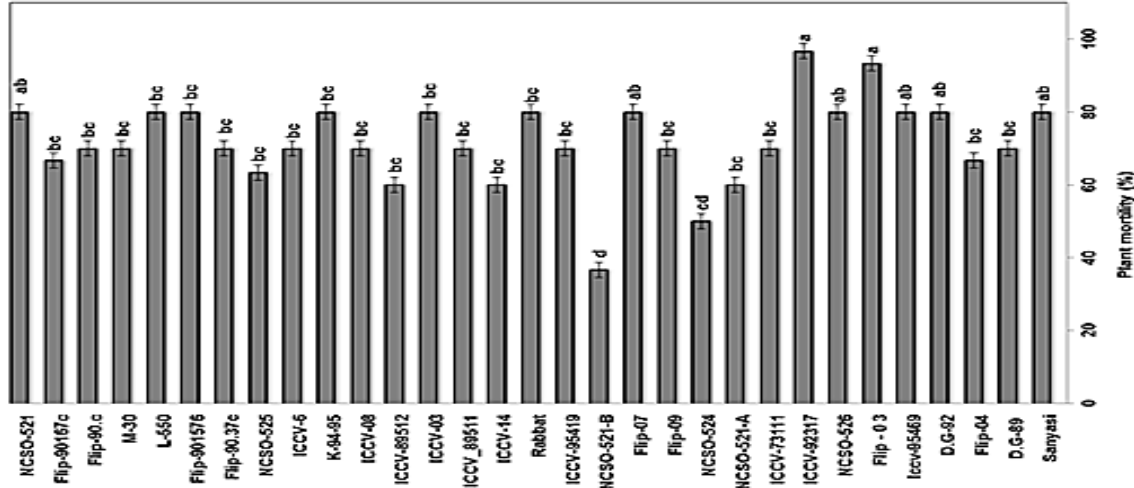


Figure 1. Plant mortality in different chickpea cultivars grown in soil artificially infested with *F. oxysporum* f. sp. *ciceris*. Means followed by different letters in respective bar are significantly different at P= 0.05.

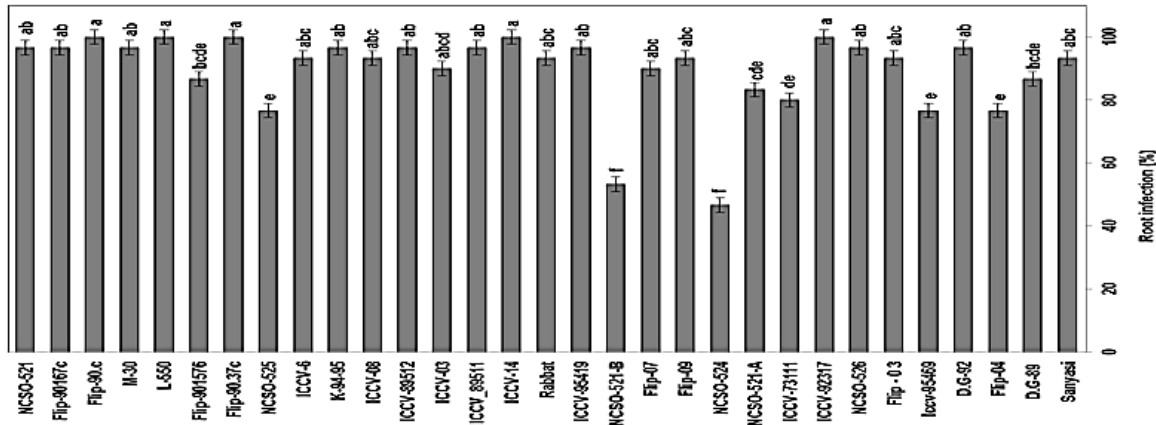


Figure 2. Response of different chickpea cultivars on root infection (%) of chickpea plants against *F. oxysporum* f. sp. *ciceris*. Means followed by different letters in respective bar are significantly different at P= 0.05.

**Plant Growth:** In contrast to the plant mortality and pathogen infection, the chickpea cultivars showed responses that are more variable in term of plant growth. Some of the cultivars, which were severely infected with the test fungus, showed some tolerance to disease and produced better plant growth than cultivars having lower infection rate. While some cultivars were badly affected and showed poor plant growth. The inoculation of *Foc* severely affected the shoot length of some chickpea cultivars, as its inoculation brought a significant maximum reduction in plant length of cv. M-30 (55.7%) followed by NCSO-526 (52.6%), Flip-03 (51.9%) as compared to the other cultivars. On the other hand pathogen failed to cause noticeable negative effects on shoot length of some cultivars including NCSO-524, Flip-09, NCSO 521-A, D.G-92 and ICCV-14 in which *Foc* caused 11.28-13.61% reduction in shoot length (Fig. 3a). In regard

of shoot weight the pathogen remarkably reduced shoot weight in most of the chickpea varieties. The highest reduction in shoot weight was observed in ICCV-98512, ICCV-98511 followed by Flip-03, NCSO-521 and ICCV-08 ranging from 86.59-88.72%, while in cv. NCSO-524 pathogen inoculation caused very low impact on shoot weight, as significant minimum reduction was observed in it i.e. 18.29% (Fig. 3b). The chickpea cultivar NCSO-524 performed well in the presence of the *Foc*, as a minimum reduction in root length and weight was observed in it. The test pathogen brought a maximum reduction in root length of cv. Flip-9037c (69.13%) followed by ICCV-73111 (66.8%), Flip-03 (65.16%) and L-550 (64.9%) (Fig. 3c). Similarly, highest reduction in root weight was recorded in cv. Flip-9037c (94.26%) followed by NCSO-526 (93.88%), Flip-03 (92.15) and ICCV-08 (92.23%) (Fig. 3d).

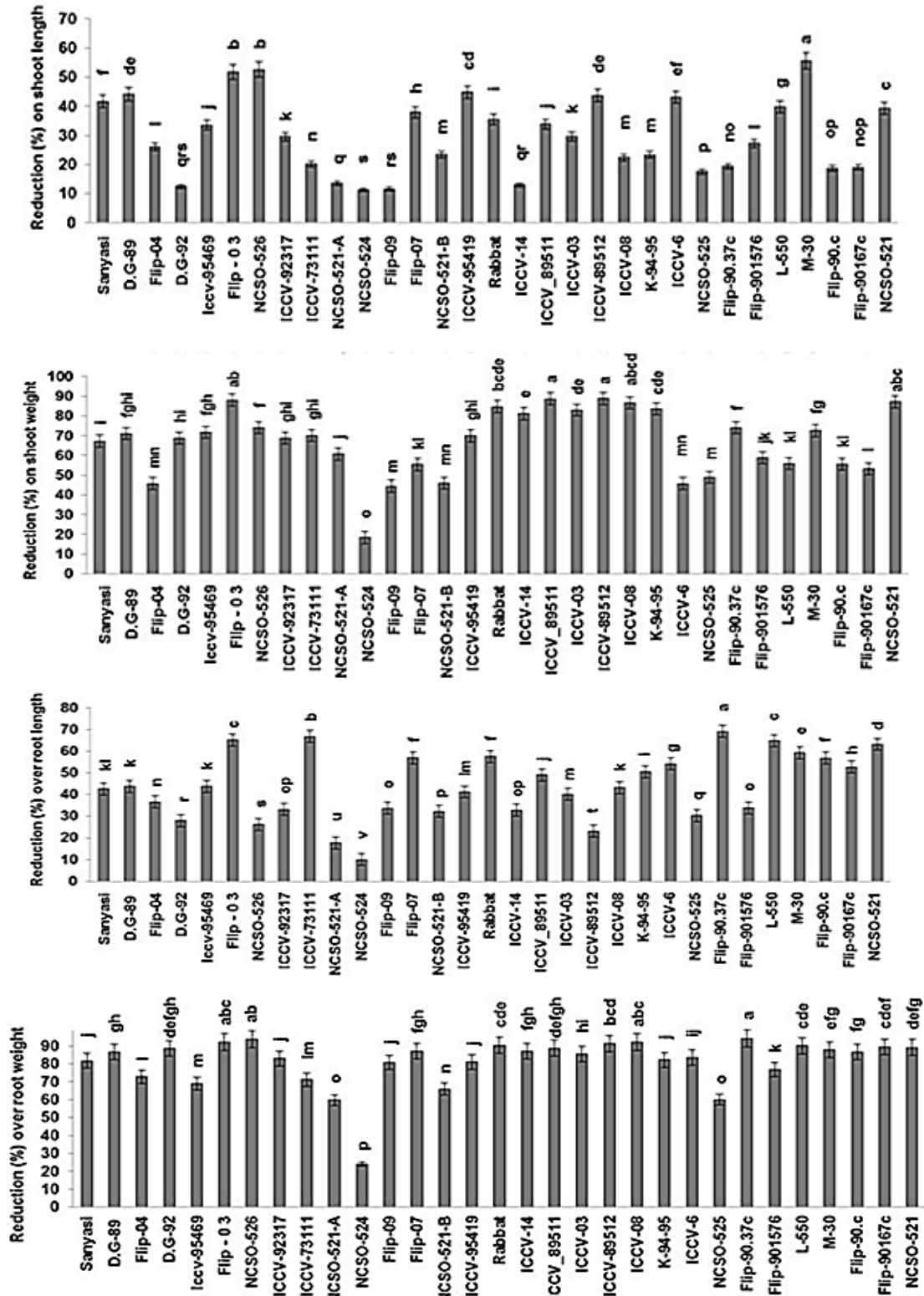


Figure 3. Response of different chickpea cultivars to *F. oxysporum* f. sp. *ciceris* (a) reduction in shoot length (b) reduction in shoot weight (c) reduction in root length and (d) reduction in root weight as compared to un-inoculated chickpea plants. Means followed by different letters in respective bar are significantly different at P= 0.05.

Correlation of infection percent was examined with reduction percentage of length and weight of root and shoot of inoculated cultivars. A positive and strong correlation (0.75%) was observed between infection

percent and reduction in root weight while correlation of infection percent with root length, shoot length and shoot weight was very weak i.e. 0.20, 0.09 and 0.37, respectively (Fig. 4).

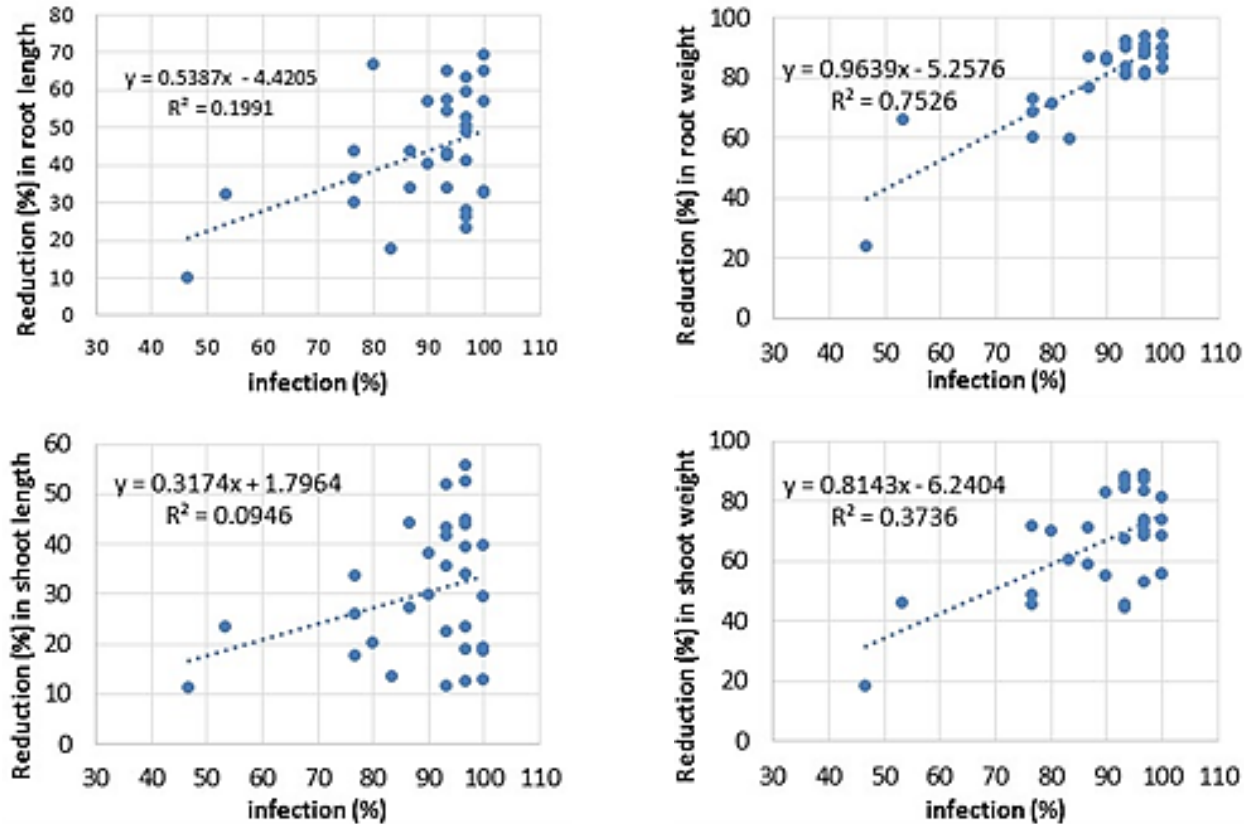


Figure 4. Correlation of infection percent with reduction percent in length and weight of 31 cultivars due to inoculation of *F. oxysporum* f. sp. *ciceris*.

## DISCUSSION

Due to soil borne and persistent nature of *Fusarium oxysporum*, its control through fungicidal spray is unfeasible. The exploitation of resistant varieties is the best option which provides not only most effective and economical control, but it is eco-friendly too. Considering the importance of resistant varieties against Fusarium wilt of chickpea, scientists throughout the world screened germplasm collection against this disease (Arvayo-Ortiz *et al.*, 2012; Iqbal *et al.*, 2010; Shah *et al.*, 2009; Nene and Haware, 1980; Halilal and Strange, 1997; Sharma *et al.*, 2005). No cultivar out of 31 was found to be completely immune to *Foc* and showed significantly higher plant mortality and pathogen infection. Lack of strong genetic resistance in chickpea cultivars against Fusarium wilt is general phenomenon throughout the world. Nene and Haware (1980) reported that only 14 varieties were found resistant out

of 7000 chickpea accessions. Similarly, Iqbal *et al.* (2005) screened 145 chickpea genotypes against *Foc* and found that no one was resistant at reproductive stage, but 14 were resistant at seedling stage. Sarwar *et al.* (2012) evaluated 41 chickpea cultivars and observed that only 2 were highly resistant and 8 were resistant. Our results were also in close confirmation to those reported by Nazir *et al.* (2012) who screened 178 chickpea lines against *Foc* and observed that none of the test lines is immune. Chaudhry *et al.* (2006) evaluated 414 cultivars and found that only 5 lines were resistant. The varieties which appeared highly resistant in small scale screening studies are usually evaluated for their field performance with multi locational trials and if found satisfactory, allowed to be included in the present cropping system. Plant defend themselves against pathogen, their strategies to cope with pathogen may be encounter as resistant and tolerance. Resistant is the ability of host to

limit pathogen infection while tolerance is the ability of host plant to limit the effect of pathogen infection on plant fitness (Kover and Schaal, 2002). There were no associations between the root infection and reduction in length and weight (except positive and strong association between infection percent and reduction in root weight) of inoculated plants lead to prove the variation in resistant and tolerance nature. Some of the cultivars which were severely infected with the test pathogen, but they showed tolerance to disease and produced better plant growth. While some cultivars badly affected and showed poor plant growth. The study and review of literature indicate that resistant to chickpea Fusarium wilt is not very common. An extensive screening and breeding program should be initiated to explore resistant and tolerant varieties.

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