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The survival of *Fusarium oxysporum* f. sp. *ciceri* in the soil in the absence of chickpea (*)

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Summary. Fusarium wilt of chickpea (*Cicer arietinum*) - caused by *Fusarium oxysporum* f. sp. *ciceri*, is a seed- and soil-borne disease of economic importance. The fungus is systemic and can be isolated from all parts of the infected plant including seed. The survival of the fungus in crop residues was studied from 1978 to 1984. Wilted plants were collected in March 1978 to study fungus survival in leaflets, terminal branches, stems, and roots. Infected plant parts were buried in the soil (Vertisol). The fungus could not survive in the leaflets stored in the laboratory or soil for more than 2 months, but it could survive in terminal branches for 6 months in the laboratory and 9 months in the soil. The fungus could survive on crop residues (root and stem portions) buried in the soil for at least 72 months. The pathogen within the host tissue could not survive in flooded soil for more than 65 days; however, it could survive for over 200 days in the soil which was kept continually wet. Influence of crop rotation and intercropping on wilt incidence in chickpea was studied from 1980 to 1983. The crops included were sorghum, maize, and wheat. The study was conducted in a portion of a wilt sick plot, developed at ICRISAT Center, Patancheru, using a highly susceptible cultivar JG 62. At the end of a 3-year study, no significant reduction in wilt incidence was observed.

Introduction

Chickpea (*Cicer arietinum* L.) wilt, caused by *Fusarium oxysporum* Schlecht. emend. Snyd. *et* Hans. f. sp. *ciceri* (Padwick) Snyd. and Hans is soil-borne disease of economic importance. The fungus is both soil-borne and seed-borne (Haware *et al.*, 1978). Chlamydospore-like structures were seen in the hilum region of seeds from diseased chickpea. *F.o.* f. sp. *ciceri* produces wilt symptoms only in *Cicer* spp., but it can colonize the roots of pea, pigeonpea, and lentil. These three crop species are considered symptomless carriers (Haware and Nene, 1982). Pathogenic formae of *F.o.* are known to persist in the soil long after the susceptible crop has been removed from the field. The chickpea wilt fungus is systemic and can be isolated from all plant parts. The fungus survives in decaying host tissues. The present studies were conducted to investigate the ability of the fungus to survive in decaying host tissues in soil in the absence of a host.

Little information is available on the application of irrigation or flooding the soil with water to control soil-borne diseases. Flooding was extensively used in Central America for the control of *F. oxysporum* Schl. f. sp. *cubense* Snyder *et* Hansen on banana (Stover, 1962). An experiment was conducted to determine the ability of the fungus to survive in wet and flooded soils.

The population of resting structures of vascular pathogens in soil can be reduced by withholding their host through crop rotation. Crop rotation is man's oldest control measure against soil-borne plant pathogens and is effective against

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many diseases (Baker, 1981). A three-year study was conducted at ICRISAT Center to examine the effects of crop rotation and intercropping on wilt in chickpea.

Materials and methods

Survival in crop residues. An experiment was initiated in March 1978 to study the survival of *F.o.* f. sp. *ciceri* in crop residues. Wilted chickpea plants were collected and leaves, terminal branches, the main stem and root, along with the 5-cm stem base, were separated and air-dried. Host tissues (2 cm long) weighing 50 g were placed in a nylon mesh and buried in pot soil (Vertisol) at depth of 7 cm. Part of the infected material in paper bags was kept in the laboratory. There were three replications. Isolations were attempted each month from 15 stem and root pieces and 30 leaflets, for 15 months.

In another experiment, chickpea roots along with the 5-cm stem base were buried in 45-cm earthen pots (bottom removed) containing field soil (Vertisol). The pots were buried in the soil in such a way that the tops of the pots were level with the soil surface. Two roots were buried in each pot. Four roots were removed from two pots after every 3 months. After washing in running water, the tissues were surface-sterilized in 2.5% sodium hypochlorite for 2-3 min. and plated on Czapek-Dox agar, which contained, in addition to normal ingredients, 500 mg PCNB, 25 mg malachite green, 750 mg Dicrysticin-S, and 2 g yeast extract per liter (Haware *et al.*, 1978). The plates were incubated at 20°C for 8 days. Identity of the fungus was verified and pathogenicity was proved. The fungus was multiplied on sand-maize meal medium for 14 days and mixed thoroughly with autoclaved soil at the rate of 100 g inoculum/2 kg soil in a 15-cm plastic pot. The seed of 'JG 62', a highly susceptible cultivar was sown. The seedlings wilted within 20 days (Haware and Nene, 1982). The pathogen was isolated from wilted plants. The isolations were continued from disintegrating root-tissues up to 33 months; after that it was impossible to take out stubble-remnants from the soil. Therefore, soil from each of two pots was transferred into new pots separately and carried into the net house. Twenty seeds of a highly wilt-susceptible cv. JG 62, collected from healthy plants and surface-sterilized with 2.5% sodium hypochlorite, were sown in two pots. A new set of two pots was used after every 3 months. Seedlings were observed for wilt symptoms. The experiment continued for 6 years and the last two pots were used in March 1984.

Survival in wet soil. Root tissues of 2-3 cm length from a wilted chickpea plant were mixed in soil (Vertisol) in four 30-cm earthen pots at the rate of 250 g air-dried root pieces per pot. The soil in pots was always flooded with water (4-5 cm above the soil surface). In another set of pots the soil was kept constantly wet. The root pieces were removed from these pots every week, washed in water and air dried. The infected tissues were surface sterilized with sodium hypochlorite and plated on modified Czapek-Dox agar.

Influence of crop rotation and intercropping. This study was initiated in 1980-81 and was conducted for 3 years on a deep black soil (Vertisol). The experiment site was a uniformly wilt-sick area at ICRISAT Center, developed by incorporating wilted plant material and growing a susceptible cultivar for several years. The experiment consisted of 12 treatments in four randomized blocks (Table 1). Treatment 1 was a basic reference treatment giving the level of wilt incidence in a continuous fallow-chickpea rotation. The second and third treatments were similar to the first with chickpea grown in all the years; but, instead of fallow, sorghum and maize were sown in each rainy season. The next four treatments tested the effect of substituting chickpea in alternate years with two other post-rainy season crops, sorghum and wheat. Treatments 8 and 9 were aimed at studying the effect of growing three and five sorghum crops before chickpea. The effect of intercropping chickpea with either sorghum or wheat on the wilt incidence was studied in the last 3 treatments.

The cultivars used were: Chickpea; 'JG 62', rainy-season sorghum; 'CSH 6', post-rainy season sorghum 'M 35-1', maize 'SB 23', and local wheat. All the crops were grown on ridges 75 cm apart. Each plot consisted of nine ridges of 9 m length. The chickpea received a basal dose of 18 kg N and 20 kg P ha^{-1} and cereal crops were top-dressed with an additional 62 kg N ha^{-1}. All operations were carried out by using bullock-drawn implements to avoid interplot contamination.

TABLE I. - Influence of crop rotation in intercropping on chickpea wilt at ICRISAT Center, Patancheru.

Sample No.	Treatments					
	1980-81		1981-82		1982-83	
	A	B	A	B	A	B
1.	Fallow	Chickpea	Fallow	Chickpea	Fallow	Chickpea
2.	Sorghum	Chickpea	Sorghum	Chickpea	Sorghum	Chickpea
3.	Maize	Chickpea	Maize	Chickpea	Maize	Chickpea
4.	Fallow	Sorghum	Fallow	Chickpea	Fallow	Chickpea
5.	Fallow	Chickpea	Fallow	Sorghum	Fallow	Chickpea
6.	Fallow	Wheat	Fallow	Chickpea	Fallow	Chickpea
7.	Fallow	Chickpea	Fallow	Wheat	Fallow	Chickpea
8.	Sorghum	Sorghum	Sorghum	Chickpea	Fallow	Chickpea
9.	Sorghum	Sorghum	Sorghum	Sorghum	Sorghum	Chickpea
10.	Fallow	Sorghum /Chickpea	Fallow	Sorghum/ Chickpea	Fallow	Sorghum/ Chickpea
11.	Fallow	Wheat/ Chickpea	Fallow	Wheat/ Chickpea	Fallow	Chickpea
12.	Sorghum	Sorghum	Fallow	Sorghum/ Chickpea	Fallow	Sorghum/ Chickpea

A: Rainy season; B. Postrainy season.

Results and discussion

F.o. f. sp. *ciceri* could not be isolated from chickpea leaflets stored in the laboratory or in soil after 2 months. Fungus could not be isolated after 6 months from terminal branches kept at room temperature and after 9 months from those buried in the soil. After 12 months, the fungus could not be isolated from the stem and root tissues stored at room temperature, but could be isolated from stem and root tissues buried in the soil for 15 months.

The fungus could survive on crop residues (root and stem portions) buried in the soil for at least 72 months. This was indicated by isolating *Fusarium* colonies from host tissues on a modified Czapek-Dox agar medium for 33 months at 3-month intervals. Each time 30 small pieces (2-5 mm) were used for isolation in 3 Petri plates. Fusarium colonies were then multiplied on sand-maize meal medium for multiplication and pathogenicity. Pathogenicity of *F. oxysporum* was proved. After 33 months, 'JG 62', a wilt-susceptible cultivar, was planted in two sets of pots. The chickpea seedlings wilted within 30 to 60 days after planting. *F. oxysporum* was isolated from the roots of wilted plants and its identity confirmed. Pathogenicity of the reisolated fungus was proved. The last two pots were used in the month of March 1984. The results indicated that *F.o.* f. sp. *ciceri* could survive in the soil for more than 72 months. The soil pathogen may survive for some time in wet soil as well as in rainwater, which may get contaminated with the pathogen present in field soil. Infected tissues harbour the pathogen and rainwater may carry such tissues to contaminate other fields. Chickpea wilt fungus within the host tissues could survive in flooded soil for 65 days and in wet soil for 200 days.

Flooding the soil has been suggested as a means of eliminating *F.o.* f. sp. *cubense* (Stover, 1962).

The influence of crop rotation and intercropping on wilt of chickpea was studied for 3 years from 1980 to 1983. In the first year (1980-81), wilt incidence in chickpea was 100%. There was no difference among the treatments. More than 90% plants wilted at 80 days after planting. In the year of 2nd experiment (1981-82) the wilt symptoms appeared late in treatments where sorghum was in crop rotation. However, more than 90% of chickpea plants wilted in all the treatments at 80 days after planting. In the year of 3rd experiment (1982-83), the wilt incidence in chickpea in treatment was more than 90% at 80 days after planting. However, different results were obtained in similar experiments on pigeonpea wilt (*F. udum* Butler). It was reduced effectively by intercropping or rotating pigeonpea with sorghum. The reduced wilt incidence was found to be consistent across 14 susceptible pigeonpea cultivars (Natarajan *et al.*, 1984).

The present studies indicate that Fusarium chickpea wilt has the ability to survive in the soil for more than 6 years and that 3 years of crop rotation is not effective in reducing wilt incidence. In view of these findings, host resistance seems to be the only possibility to control chickpea wilt. Fortunately, resistance to Fusarium wilt is available in chickpea germplasm (Nene and Haware, 1980; Haware *et al.*, 1992).

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