



Occurrence of pea root rot/wilt complex disease in Himachal Pradesh

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Abstract

Incidence of pea root rot /wilt complex disease had been recorded in different agro climatic zone of pea growing areas of the Himachal Pradesh. In Zone IV, the disease had assumed severe form with incidence of 54.7% at HAREC, Kukumseri whereas, in Zone II and III, the status of the disease remained moderate with incidence of 17.7 to 35.3% at Bajaura and Palampur respectively. Two species of *Fusarium* viz., *Fusarium solani* f.sp. *pisi* and *Fusarium oxysporum* f.sp. *pisi* were found to be associated with pea root rot/wilt complex disease in Himachal Pradesh. Both species produced distinct symptoms when inoculated on pea seedlings in test tubes containing Hoagland's solution. *F. solani* f. sp. *pisi* was solely responsible to cause root rots (necrotic and macerated root tissues) of peas with yellowing of leaves from basal leaf to upward and then, ultimately collapse of infected pea plants whereas, *F. oxysporum* f. sp. *pisi* was responsible to cause wilting of peas (without root rots) and then, ultimately collapse of infected pea plants.

Key words: Pea root rot /wilt complex, *Fusarium solani* f.sp. *pisi* and *Fusarium oxysporum* f.sp. *pisi*.

Pea root rot/wilt complex disease is an emerging problem and poses serious threat to pea cultivation in the state. The pea cultivars grown in Himachal Pradesh are susceptible to the pathogens associated with the disease. The disease is occurring in all pea growing areas of the state at pre-flowering or flowering stage to affect the initial crop stand of pea which ultimately leads to enormous yield losses. The disease starts either with yellowing of basal leaves and then, proceeds up-ward or wilting of the infected plants and then, ultimately collapse of those. The disease syndrome like root rots, yellowing of leaves and wilting collectively represent a detrimental disease complex and hence, referred as root rot/wilt complex disease. The disease happens to be at initial stage of the pea crop which ultimately results in huge crop losses to incur heavy economic losses to the farming community. Under situation of monoculture and extensive cultivation of pea in agro-climate Zone IV of the state where it is grown as off- season crop, pea root rot/wilt complex disease have become the major constraints in pea productivity.

More than 20 different pathogens i.e. *Fusarium solani* f. sp. *pisi*, *F. oxysporum* f. sp. *pisi*, *Aphanomyces euteiches*, *Phoma medicaginis* var. *pinodella*, *Rhizoctonia solani*, *Hycosphaerella pinodes*, *Thielaviopsis basicola* and

Pythium spp. including *P. ultimum*, *P. vexans*, *P. splendans*, *P. debaryanum*, *P. aphanidermatum* and *P. irregulare* have been reported to be associated with the disease from different parts of the world (USDA 1960). In Himachal Pradesh, pathogens viz., *Fusarium oxysporum*, *Fusarium solani*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum* and *Phoma medicaginis* var. *pinodella* have been reported to be associated with root rot/wilt complex of pea (Sagar 2004, Dohroo *et al.* 1998, Kapoor *et al.* 2006). These reports lead to further investigation of pathogens associated with pea root rots/wilt complex disease in the state.

During 2014-2015, an extensive survey and surveillance of pea growing areas of Himachal Pradesh were conducted at initial stage of pea growing season to observe the disease status. The per cent disease incidence was recorded at each location by marking five spots of 2.0 m² area to assess the disease. Diseased samples were also collected in paper envelopes during the surveys to isolate pathogen (s) associated with the disease. Isolations were made on PDA to procure pure cultures of isolates by single hyphal tip method. Identification of pathogens was done on the basis of microscopic observations for cultural and morphological characteristics after conducting pathogenicity tests (Table 1).

Table 1. Identification of the pathogens on the basis of keys (Mycology Online)

Parameter	<i>Fusarium solani</i>	<i>Fusarium oxysporum</i>
Colour of colony on PDA	White to cream colonies growing rapidly with aerial mycelium becoming bluish-brown when sporodochia formed.	Cottony white colonies growing rapidly with aerial mycelium becoming purple to orange or dark blue to dark purple when sporodochia formed.
Growth on PDA	4.5 cm in four days	4.5 cm in four days
Macroconidia	Macroconidia of 28-42 x 4-6 μ m with three to five-septa and fusiform, cylindrical, often moderately curved, with an indistinct pedicellate at foot cell and a short blunt apical point.	Macroconidia of 23-54 x 3-4.5 μ m mostly with three septate are fusiform in shape and slightly curved with pointed end at the tip and basal cells pedicellate.
Microconidia	Cylindrical to oval microconidia of 8-16 x 2-4.5 μ m with one to two-celled are abundantly present.	Ellipsoidal to cylindrical (straight or often curved) microconidia of 5-12 x 2.3-3.5 μ m are mostly non-septate.
Chlamydospores	Smooth to rough-walled chlamydospores of 6-10 μ m borne singly or in pairs on short lateral hyphal branches or intercalary	Smooth to rough-walled chlamydospores of 5-13 μ m borne terminal or intercalary.

Occurrence of disease

The per cent disease incidence varied considerably in different pea growing areas of the state. In agro climatic Zone IV, the highest disease incidence of 54.7% was recorded at HAREC, Kukumseri followed by Dalang (54.3%), Kishori (46.0%), Sissoo (32.0%) and Karpat (24%). In Zone III, the disease incidence varied from 17.7 (Bajaura) to 197% (Nagwain). In Zone II, the highest disease incidence of 35.3% was recorded at Departmental Farm of CSK HPKV, Palampur followed by Kachhiyari (30.7%) and Sagoor (22.0%). The data on per cent incidence clearly depicted that pea root rot /wilt complex disease was present in all pea growing areas of the state and had assumed the status of moderate to severe form. Sagar (1996) also noticed moderate to severe form of this disease in Himachal Pradesh and

highest incidence of 45.2% was reported from Lahaul valley with four fungal pathogens viz., *F. oxysporum*, *F. solani*, *R. solani* and *P. medicaginis* var. *pinodella* associated with the disease.

Isolation of Pathogen (s) and maintenance of pure cultures

Isolations were made from diseased samples of pea root rot/wilt complex collected from pea growing areas of the state during survey and surveillance. Pure cultures of the isolates were obtained through single hyphal tip method and were maintained on PDA medium at $26 \pm 1^\circ$ C for further studies. Fifteen isolates were obtained from different pea growing areas which were further subjected to pathogenicity tests and microscopic observations for their pathogenic confirmation and identification.

Pure cultures of fifteen isolates were observed for their morphological, cultural and pathogenic characteristics (Table 2). Pathogenicity tests of pure cultures of the isolates were carried out in the growth chamber of the department and two distinct types of symptoms were produced (Plate 1). One with root rots (necrotic and macerated root tissues) with yellowing of leaves from basal leaf to upward whereas, other one shows wilting without root rots and ultimately collapse of infected pea plants. Microscopic and pathogenic observations revealed that two different species of *Fusarium viz;* *F. solani* f.sp. *pisi* and *F. oxysporum* f.sp. *pisi* were associated with pea root rot / wilt complex disease. In case of *F. solani* f.sp. *pisi*, dull grey white mycelium was observed with moderately curved macro conidia of 30-40 x 4.5-6.0 μm having thick walls and 3-5 septa which were not sharply curved at the end point (Plate 2). Micro conidia of *F. solani* f.sp. *pisi* 8-15 x 2-4.0 μm were spherical to oval in shape. Rough walled chlamydospores of 6-9 μm on hyphal branches or intercalary. Pathogenicity test showed that this pathogen produced characteristic symptoms of root rots (necrotic and macerated root tissues) upto collar regions showing yellowing of basal

leaf to upward and then, ultimately collapse of pea seedlings when inoculated with spore suspension of *F. solani* f.sp. *pisi* in the test tubes containing Hoagland's solution (Plate 1). In case of *F. oxysporum* f.sp. *pisi*, cottony white mycelium with tints of the stroma coloring (commonly of purple red, bluish grey or yellowish tan) was observed with typically dorsoventrally curved sickle-shaped macro conidia of 25-35 μm x 3-5 μm with thin walls having 3-5 septations and were sharply curved with tapering ends (Plate 2). Oval to ellipsoidal micro conidia of *F. oxysporum* f. sp. *pisi* of 5-12 x 3-5 μm were present in abundance. Rough-walled chlamydospores of 5-12 μm borne terminal or intercalary. Pathogenicity test showed that the second species of *Fusarium* produced characteristic symptoms of wilting without root rots and then pea seedlings ultimately collapse due to clogging of xylem vessels when inoculated with spore suspension of *F. oxysporum* f.sp. *pisi* in the test tubes containing Hoagland's solution (Plate 1).

Two species of *Fusarium viz.*, *F. solani* f.sp. *pisi* and *F. oxysporum* f.sp. *pisi* were found associated with pea root rot/wilt complex in Himachal Pradesh (Table 2 & Plate 2).

Table 2. Morphological, cultural and pathogenic characteristics of isolates

	Morphology	Culture	Pathogenicity	Symptom	Pathogen
Type-I (Fs1- Fs9)	Dull white mycelium was observed with sickle to oval shaped macro conidia of 30-40 x 4.5-6.0 μm having 3-5 septation with thick walls, not sharply curved at the end. Micro conidia of 8-15 x 2-4.0 μm are spherical to oval in shape. Rough walled chlamydospores of 6-9 μm on hyphal branches or intercalary	Dull cottony mycelium white colour	+	Necrotic and macerated root tissues up to collar regions (root rots) with yellowing starts from basal leaf to upper leaves	<i>Fusarium solani</i> f.sp. <i>pisi</i>
Type-II (Fo1-Fo6)	Dorsa-ventrally curved sickle- shaped macro conidia of 25-35 μm x 3-5 μm with 3-5 septation with thin walls and sharply curved or tapering toward the ends. Micro conidia of 5-12 x 3-5 μm , oval to ellipsoidal in shape, were present in abundance. Rough-walled chlamydospores of 5-12 μm borne terminal or intercalary	Light white mycelium with tints of the stroma coloring, commonly of purple-red, bluish-grey or yellowish-tan	+	Wilting with vascular discolouration and then, collapse of pea seedlings	<i>Fusarium oxysporum</i> f.sp. <i>pisi</i>

+=Pathogenic

Both species of *Fusarium* viz., *F. solani* f.sp. *pisi* and *F. oxysporum* f.sp. *pisi* produced distinct symptoms (Plate 1) in the pathogenicity tests. One with root rots (necrotic and macerated root tissues) resulting in yellowing of leaves from basal leaf to upward whereas, other one shows wilting without root rots and then, ultimately collapse of infected pea plants. However, it has been observed that *F. solani* f.sp. *pisi* was solely responsible to cause root rots of peas whereas, *F. oxysporum* f.sp. *pisi* was responsible to cause wilt of the same.

Further it has been ascertained that mixed infection of both *F. solani* f.sp. *pisi* and *F. oxysporum* f.sp. *pisi* can produce disease syndrome like root rots, yellowing of leaves and ultimately wilting of infected pea plants in the state.

Hence, the predominance of both the species of *Fusarium* viz; *F. solani* f. sp. *pisi* and *F. oxysporum* f. sp. *pisi* has been ascertained to be associated with root rots and wilt of peas and other pathogens reported by workers might be present rarely in some pockets of pea growing areas in the state.

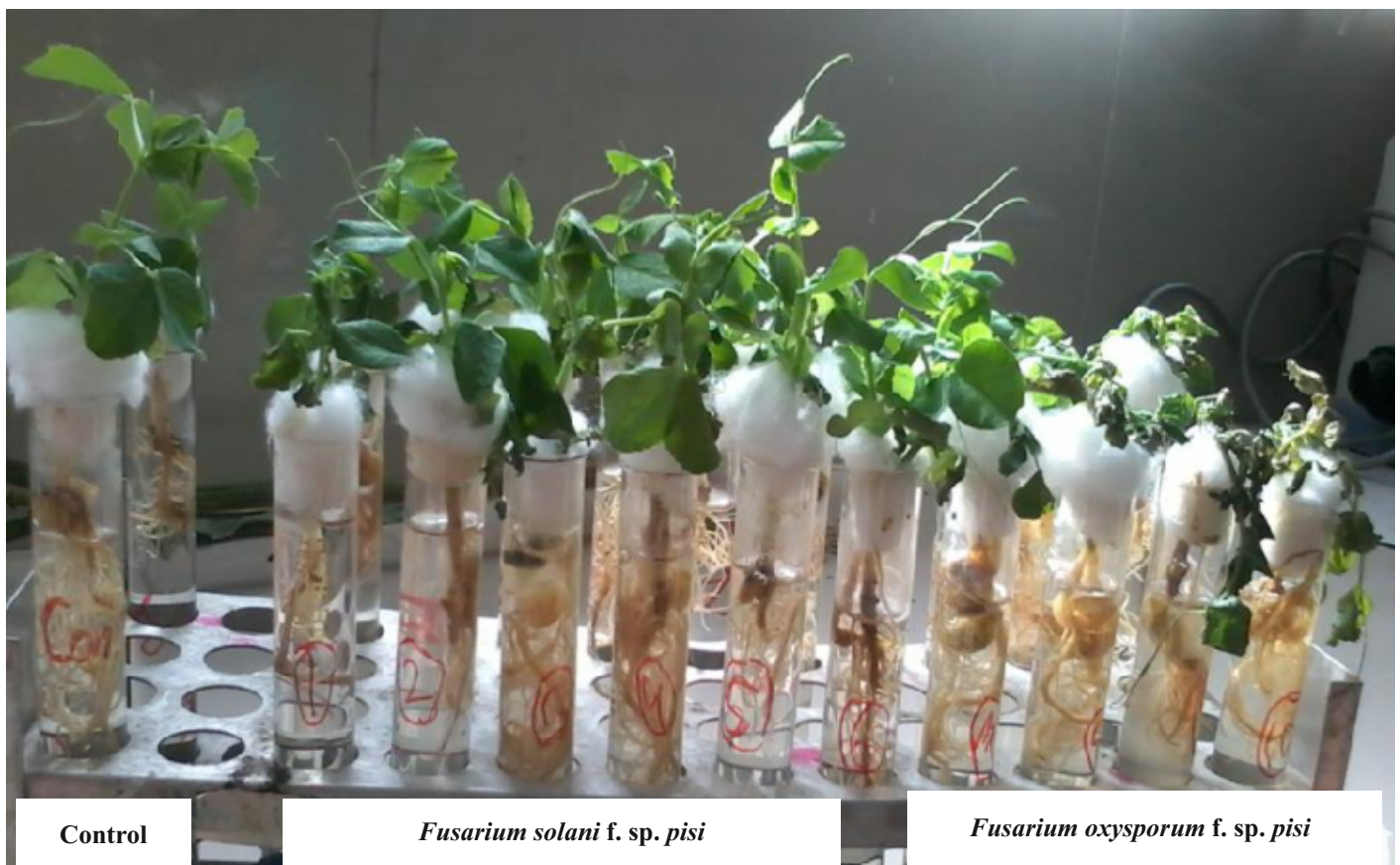
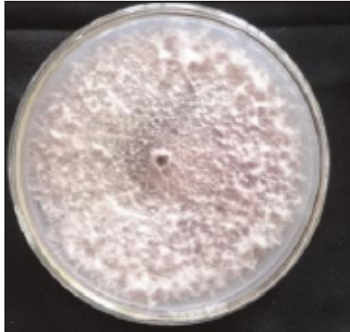
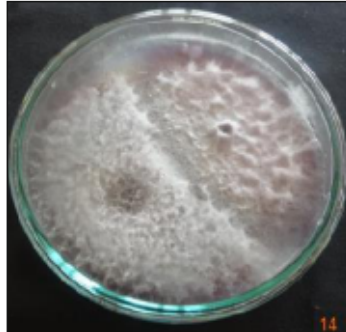


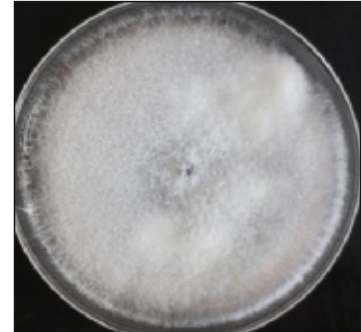
Plate 1. Pathogenicity tests of *Fusarium oxysporum* f.sp. *pisi* and *Fusarium solani* f.sp. *pisi*



Culture of *Fusarium oxysporum* f.sp. *pisi*



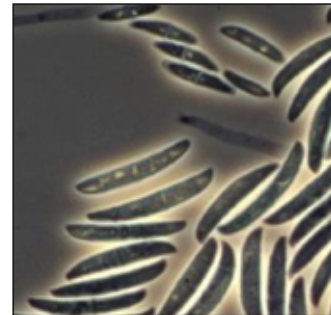
Culture of *Fusarium oxysporum* f.sp. *pisi* (Fo) and *Fusarium solani* f.sp. *pisi* (Fs)



Culture of *Fusarium solani* f.sp. *pisi*



Culture of *Fusarium oxysporum* f.sp. *pisi*



Culture of *Fusarium oxysporum* f.sp. *pisi* (Fo) and *Fusarium solani* f.sp. *pisi* (Fs)

Plate 2. Culture and spores of Fusarium species

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