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Short Research Article

Occurrence of Fusarium Wilt of Tomato under Protected Conditions in Himachal Pradesh, India

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Abstract

A survey of different polyhouses in tomato growing areas in Solan, Sirmour, Hamirpur and Bilaspur districts of Himachal Pradesh, India was carried out to record the incidence of Fusarium wilt during September, 2012. Overall incidence of Fusarium wilt in four districts surveyed was 19.50%. Amongst various localities surveyed highest incidence of the wilt was recorded at Kadriyana (27.65%) in district Hamirpur followed by Nauni (25.56%) in district Solan, whereas lowest incidence was recorded at Dharot (9.90%) in district Hamirpur followed by Gassour (12.50%) in district Bilaspur. Locality wise data indicated that the incidence of wilt was higher in comparatively warmer areas and also in the areas where tomato cultivation was continuously done under polyhouses. The wilt incidence at locations in warmer regions in all the districts surveyed was 20% and above. Previously no survey of the polyhouses in Himachal Pradesh has been done to know the status of Fusarium wilt, because of non occurrence or less incidence of the disease in the region. Hence, the present study is new in this regard. Isolation of the pathogen associated with tomato wilt was made from the diseased tissues in roots and collar region of the plant on the potato dextrose agar (PDA) medium. The pathogen thus isolated was identified to be *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W.C. Snyder and H.N. Hans on the basis of its colony morphology and microscopic features. The pathogenecity of the fungal pathogen was also proved after artificial inoculation of the tomato seedlings.

1. Introduction

In India, total area under protected cultivation is around 25,000 ha of which 2,000 ha area is under greenhouse vegetable cultivation (Sabir and Singh, 2013). Greenhouse technology has made the farmers in hilly states like Himachal Pradesh to earn additional remuneration even during off-season with in a small piece of land and. Tomato (*Solanum lycopersicum* L.) is the main vegetable crop grown under protected cultivation round the year. However, prevalence of changed microclimatic conditions and intensive cultivation of a single crop in polyhouses have led to the emergence of soil-borne plant diseases. In some areas this situation has resulted in huge economic losses to the growers. Fusarium wilt caused by *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W.C. Snyder and H.N. Hans, is a serious disease of tomato especially in warmer areas because the pathogen requires a soil temperature around 28^{0}C for its growth and development (Gupta and Thind, 2006). In Himachal Pradesh this disease was not so serious in recent past, but now it has started appearing in many polyhouses. The disease may cause crop losses to the tune of 10 to 80% (Kapoor, 1988). The fungal pathogen is soil-borne and known to persist in soil for many years. Infection of the disease mostly originates from the pathogen propagules present in soil or infected plant debris. Therefore, healthy plants which are transplanted in infested soil also become infected (Ignjatov et al., 2012). As the disease has started to make its impact on tomato cultivation in polyhouses, the present investigation on the survey of various localities in the state to know the status of Fusarium wilt and its identification was conducted.

2. Materials and Methods

2.1. Disease survey

A survey of different polyhouses in tomato growing areas in Solan, Sirmour, Hamirpur and Bilaspur districts of Himachal Pradesh was carried out to record the incidence of Fusarium wilt. The survey was conducted during September, 2012. The areas surveyed were Nauni, Ghatti, Kandaghat and Solan Brewary in district Solan; Maryog, Dilman and Yashwantnagar in district Sirmaur; Dharot, Kadriyana and Tikker in district Hamirpur; and Parnali, Gassour and Jukhala in district Bilaspur.

At each location the observations were recorded in 3 to 4 polyhouses with a sample size of 100 plants in each polyhouse. Disease incidence (%) was calculated by the formula given below:

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased plants}}{\text{Total number of plants observed}} \times 100$$

2.2. Isolation and identification of the pathogen

The isolation of wilt pathogen was taken from roots, and collar region of those plants which were showing the characteristic wilt symptoms. The infected plant material was washed with tap water to remove the surface soil. Small bits of 1 to 2 mm size were cut from the juncture of diseased and healthy portion of roots, and collar region of the plant with the help of a sterilized blade. These bits were surface sterilized with mercuric chloride (0.1%) for 10 to 20 seconds and were washed thrice with sterilized distilled water under aseptic conditions. The bits were then placed on the sterilized filter paper to remove the excess moisture and were subsequently transferred to sterilized Petri plates containing potato dextrose agar (PDA) medium. The medium was supplemented with streptocyclin (30 mg l^{-1}) while pouring it in petriplates after sterilization (autoclaved at 1.05 kg cm^{-2} for 20 minutes). The inoculated Petri plates were incubated at 27±1°C in BOD incubator and examined daily for mycelial growth, if any. The fungal growth developed in Petri plates was purified by hyphal tip technique and was further cultured on slants containing PDA. The morphological characters of the fungus were studied on host as well as in the culture grown on PDA.

2.3. Pathogenecity test

For pathogenicity test, conidial suspension of the pathogen was prepared (1×10^6 cfu ml^{-1}) in a conical flask from cultures grown in potato dextrose broth with constant agitation (110 rpm) at 28°C for 7 days. Roots of four weeks old seedlings (three fully expanded true leaves) of susceptible cultivar Himsohna were immersed in the appropriate conidial suspension for 10 min, individually. Inoculated seedlings were transplanted into a pot containing 1 kg of sterilized soil, and grown in a green house for 5 weeks. In control plants roots were dipped for 10 min in sterile water and were grown similarly. The inoculated seedlings showed typical symptoms of Fusarium wilt including yellowing, vascular necrosis and wilting after three weeks of inoculation. None of the inoculated plants survived 40 days after inoculation. The control plants remained without symptomatic.

3. Results and Discussion

3.1. Incidence of Fusarium wilt of tomato under polyhouses in of Himachal Pradesh

The incidence of Fusarium wilt of tomato caused by *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W.C. Snyder and H.N. Hans was recorded during the cropping season of 2012 in different tomato growing areas of four districts of Himachal Pradesh (Table 1). Incidence of wilt was observed in almost all the locations surveyed and it ranged between 9.90 to 27.65%. Overall incidence of Fusarium wilt in four districts surveyed was 19.50%. Disease incidence was maximum (22.19%) in district Solan out of four districts surveyed followed by district Sirmaur (20.07%). Amongst various localities surveyed highest incidence of the wilt was recorded at Kadriyana (27.65%) in district Hamirpur followed by Nauni (25.56%) in district Solan, whereas lowest incidence was recorded at Dharot (9.90%) in district Hamirpur followed by Gassour (12.50%) in district Bilaspur. Locality wise data indicated that the incidence of wilt was higher in comparatively warmer areas and also in the areas where tomato cultivation was continuously done under polyhouses. The wilt incidence at locations in warmer regions in all the districts surveyed was 20% and above. Previously no survey of the polyhouses in Himachal Pradesh has been done to know the status of Fusarium wilt, because of non occurrence or less incidence of the disease in this area. Hence, the present study is new in this regard. However, if we see within country basis Kapoor (1988) has reported wide spread occurrence of this disease in different tomato growing states like Maharashtra, Tamilnadu, Bihar, Dehli etc. with losses between 10 to 80% in different regions of the country. Under present investigation, wilt incidence was higher in comparatively warmer areas.

Table 1: Incidence of Fusarium wilt of tomato grown under protected conditions in mid hills of Himachal Pradesh, India during 2012

District	Locality	Disease incidence (%)
Solan	Nauni	25.56
	Ghatti	23.59
	Kandaghat	18.40
	Solan Brewary	21.24
	Mean	22.19
Sirmaur	Maryog	15.86
	Dilman	22.55
	Yashwantnagar	21.80
	Mean	20.07
Hamirpur	Dharot	9.90
	Kadriyana	27.65
	Tikker	18.42
	Mean	18.65
Bilaspur	Parnali	20.90
	Gassour	12.50
	Jukhala	17.93
	Mean	17.11
Overall mean		19.50

Fusarium wilt is known to become serious in warmer areas because of the fact that this pathogen requires 25-31 ^{0}C soil temperature for its development (Gupta and Thind, 2006). During summer the temperature of polyhouses in mid hills of Himachal Pradesh reach up to 30 ^{0}C and above, as a 20% increase in temperature inside the polyhouse is reported than outside temperature (Spehia, 2011). Prevalence of higher temperature is favorable for the development of wilt pathogen. Continuous cropping of tomato round the year under polyhouses might be another reason for increased incidence of this disease under protected cultivation. Multiplication and spread of soil borne diseases especially Fusarium wilt is more when a specific crop is grown continuously (Charoenporn et al*.,* 2010). As the disease is emerging as a serious disease under polyhouses of mid hills of Himachal Pradesh, there is a need to find out suitable control measures for this disease so that it may not cause economic losses to the growers.

3.2. Isolation and identification of the pathogen

During the course of survey, typical symptoms of the disease were observed in different polyhouses. The initial symptoms were typical yellowing of lower leaves, vein clearing, stunting, necrosis and wilting. The symptoms of the wilt moved upwards with the gradual upward extension of the pathogen and the discoloration of the vascular tissue of roots and stems and finally the entire plant got affected and ultimately killed. Isolations of the pathogen associated with tomato wilt were made from the diseased roots and collar region of the plant on the potato dextrose agar (PDA) medium. Culture of *Fusarium oxysporum* f.sp. *lycopersici* (FOL) was pinkish white to pinkish in colour on PDA and took 7 days to fully cover the petriplate. Under microscope two types of conidia i.e. micro and macro conidia were observed along with thin septate hyaline mycelium. The microconidia were aseptate, hyaline, oval to round in shape and measured 2.2-4.8×1.4-2.4 μm^2 in size. Macroconidia were 3 to 5 septate, often hooked at tapered tip and somewhat thicker at upper third portion than in central portion and measured 23.0-38.0×2.8-5.4 μm^2 in size. The isolated fungus was identified as *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W. C. Snyder and H.N. Hans on the basis of its colony morphology and microscopic features and their analogy with that given by Booth (1971) and Brayford (1992).

4. Conclusion

The tomato cultivation under protected conditions is increasing day by day in hilly areas of Himachal Pradesh and the monoculture of the crop under such conditions has resulted in appearance of various soil borne diseases. The Fusarium wilt is identified to be the main soil borne diseases in mid hills of Himachal Pradesh under protected conditions. The pathogen associated with this disease is identified to be *Fusarium oxysporum* f. sp. *lycopersici* (Sacc.) W.C. Snyder and H.N. Hans. The highest disease incidence among the four districts was recorded in Solan district followed by Sirmour, Hamirpur and Bilaspur.

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