STUDIES ON FUSARIUM OXYSPORUM ATTACKING THE ROOTS OF BEAN PLANTS

By

[illegible] Hajoush

M.Sc. Ain Shams University [illegible]

THESIS

Submitted in Partial Fulfilment of the Requirements for the Degree

Of

DOCTOR OF PHYLOSOPHY

in

PLANT PATHOLOGY

Plant Pathology Department

Faculty of Agriculture

Ain-Shams University

Cairo

1975

APPROVAL SHEET

Thesis for Ph.D. Degree entitled Studies on *Fusarium oxysporum* attacking the roots of bean plants.

Approved by:

....

....

....

Date: 21/4/1975.

بسم الله الرحمن الرحيم

C O N T E N T S

INTRODUCTION

Snap bean (*Phaseolus vulgaris*) is considered to be one of the most important vegetable crop for its valuable as a source of protein, calcium, iron, riboflavin, and vitamin B_1. It is clear also important for exportation where the export amount in 1972 reached 300 tons.

The bean yield is greatly affected by wilt disease caused by *Fusarium oxysporum* f. sp. *phaseoli*. The disease has long bean known as a deleterious disease of bean in A.R.E. and among parts of the world. Harter (1929) was the first who recorded this disease in Sacaramento Valley of California. Its damage reached 50 % in a field of approximatly 50 areas. Kendrick (1942) observed the disease in the same Valley. Cardoso (1966) recorded this disease for the first time in Brazil.

The present investigation was carried out to study the relation between nitrogen and phosphorus (in sand culture) and the resistance to this disease in bean varieties. The effect of nutrition on the component of susceptible and less susceptible bean varieties and also on the fungal growth as spore production on media, was concerned.

Another study was played on the effect of systemic fungicides on controlling the disease, fungal growth, sporulation, spore germination and pectinase activity.

Histopathological studies were tried with nutrition and fungicides treatments. Their effect was studied on the tissues area of bean plant, mode of the fungus pentration and finaly the role of pectinase in induction fusarium wilt.

REVIEW OF LITERATURE

Enzyme activity :

The role of pectin in plugging is still debated. Dimond and Waggoner (1953) mentioned that pectins have the wilt including properties of other polysaccharides. It has been reported by different authors that the wilt inducing pathogens produce pectolytic enzymes which could partially hydrolyze the pectins in vascular tissues. Gothoskar et al.(1955);Waggoner and Dimond(1955); Wood (1961) and Dees and Stahmann (1962) found such enzymes in juice from crushed infected stems of tomato.

Scheffer and Walker (1953); Husain and Kelman (1958) attributed wilting of diseased plants to altered viscosity of tracheal fluid. Husain and Kelman (1958) found that Pseudomonas solanacearum produces an extracellular polysaccharide. They added that pathogenicity of different strains of the organisms was correlated with their ability to produce this polysaccharide. Pathogenicity of strains was also correlated with viscosity of culture filtrates, which is considered a measure of polysaccharides concentration. Vascular pathogen produces an array of hydrolytic enzymes which depolymerize pectic and cellulosic substances. Gothoskar et al. (1955) reported that culture filtrates of *Fusarium oxysporum* f. *lycopersisi*, grown on moist wheat bran caused vascular browning and wilt in tomato plants. The active factor in these filtrates was heatlabile and non-dialyzable, indicating an enzymatic nature of this filtrate.

Similar results were reported by Pierson et al. (1955) who presented evidence on the role of pectic enzymes in vascular plugging and discoloration in tomato wilt.

Waggoner and Dimond (1955) reported that *Fusarium oxysporum* f. *lycopersici* produced pectin methylectarase and polygalacturonase, when it was grown on a pectin-containing medium.

Dees and Stahmann (1960, 1962 a, 1962 b) found that juice extracted from stems of infected plants with *Fusarium oxysporum* f. *lycopersici* and banana plants infected by *Fusarium oxysperum* f. *cubense*, were found to contain higher amounts of pectin methylestrase and chain-splitting pectolytic activity than healthy plants do. Since pectic enzymes play an important role in producing wilt, the authors suggested that resistance to fusaria may be due to a suppression of the formation of fungal pectic enzymes by factors in the resistant tissue and that susceptibility may be assosiated with great pectic enzyme formation.

Husain and Dimond (1955) noticed that culture filtrates of F. oxysporum f. lycopersici showed high cellulase (Cx) activity when the fungus was grown on Richard's solution containing cellulose as carbon source but not when glucose was the carbon source. When the fungus was grown on sterilized living tomato stem sections, the Cx enzymes was produced but no enzyme was detected in non-inoculated stem pieces. A solution of partially purified enzyme caused wilting of tomato cuttings when the enzyme solution was introduced through cut ends of the stems for 16 - 24 hours. No injury was caused by a heat inactivated enzyme sample. The authors indicated that the fungus is a cellulolytic organism and suggested that cellulase produced by this pathogen may play a role in the disease syndrome.

Wood (1960) evaluated the role played by pectic enzymes in disease. He stated that pectin enzymes may play a part in the disease development. It is based on the fact that the pathogen often produces them in culture; and that preparations with pectic enzyme activity can produce, in cutting, some of the natural disease symptoms. In addition, there is evidence for the action of pectic enzymes particularly in the final stages of some diseases. More pectinesterase and, in some cases, depolymerase activities were found in the diseased tissues than in healthy plants.

Mc Donnell (1962) examined the pathogenicity of a series of ultraviolet-induced mutants of Fusarium oxysporum f. lycopersici in relation to their pectolytic enzyme production. All the strains produced pectin methylestrase and all but one produced pectolytic enzymes. No correlation was found between the ability to produce the enzymes, in vitro, and pathogenicity. The absence of pectolytic activity in one strain suggested that this type of enzyme is not concerned with the production of moderate disease symptoms in tomato plants.

Nutrition :

Yu and Fang (1948) found that poor soil was more favourable for the development of wilt disease than fertile soil.

Menon (1957) found that the total phenol content of the leaves and petioles in the resistant tomato was higher in the infected plants than in healthy ones. This was not true in susceptible varieties. The polyphenol oxidase level varied according to the total content, which was proportional to resistance. It was concluded therefore, that phenols cannot be responsible for wilting.

Pleskov et al. (1957) detected 23 amino acids from leaves of beans grown in fertilized soil, sand culture, with NPK, NK, NP and PK. They reported that arginine was most seriously affected by nutritional conditions showing with NP a 150 % increase and with PK a 85 % a decrease on the NPK figure. The total amount of amino acids was greatest with NP and smallest with PK.

Benedict and Hildebrand (1958) noted an increase in concentration of all amino acids as soybean stem tissue matured and became more resistant to infection by stem canker. Total nitrogen of the tissues studied also increased with plant maturity.

Nishimura (1959) reported that browning of infected water melon tissues with <u>Fusarium</u> <u>oxysporum</u> f.

niveum was limited in early stages of infection in the xylem parenchyma, where also an increase of phenolic substances and abnormal activation of phenoloxidase occurred. Although tissue browning proceeded the invading hyphae, yet it did not form a chemical barrier to the advancing fungus, mycelium which was later found growing in the discoloured cells.

Toussoun et al. (1959) stated that pathogenicity of Fusarium solani f. phaseali to bean stem was governed by the nutrition of the pathogen. In addition, disease ~~deve~~lopment was enhanced by nitrogen nutrition of the fungus and delayed by glucose nutrition of it.

Bains (1961, 1967 and 1969) found that the additional increments of N, P_2O_5 and K_2O increased the concentration of N, P and K respectively, in bean tissues. This increase was significantly and positively related to NPK in the plants.

Chi and Hanson (1962) reported that the concentration of nitrogen, phosphorus, or potassium in a balanced nutrient solution had a profound effect on host (Trifolium protense) and Fusarium disease development. Least plant growth and most disease was developed at the lowest concentration of nutrition. In general, least vigorous plants had most disease.

Černavskaja; Černavskaja and Niciporovič (1963) noticed that the minimum dry weight of seedlings tended to occur later and to be lower with heigher levles of N in the substrate. The total N and protein N contents of beans were increased and the carbohydrate content was reduced by raising the level of N in culture solution.

Cochrane et al. (1963) found that nitrogen and carbon sources were essentially for macroconidia growth and germination of _Fusarium_ _solani_ f. _phaseoli_.

Matta and Dimond (1963) indicated that polyphenol oxidase activity in wilted stems of tomato was 3 - 10 times higher than that in the healthy stems. In diseased stems, the polyphenol oxidase activity associated with infection, raised gradually until the ninth day and increased sharply thereafter. Polyphenol oxidase activity was well correlated with the severity of foliar symptoms and quantity of mycelium. Vascular browning appeared after polyphenol oxidase activity became high.

Khaj and Pleskov (1964) reported that amino plant growth and reduced disease incidence.

Maclean and Byers (1968) found in field peas that percentage of N decreased as the season progressed while P remained relatively constant.

Maier (1968) stated that bean plants grown in a nitrogen-poor sand medium were more susceptible to infection by Fusarium solani f. phaseoli than vigorous plants produced in a nitrogen-enriched medium.

Osawa and Lorenz (1968) reported that bean plants with low P levels produced small plants with dull green leaves.

Bezljudnyj (1969) found that peas well provided with P showed a high total, protein N content and a greater concentration of neuclo-protein P in the proteins than P- deficient plants. He also added that the higher level of protein synthesis the more rapid was the dry matter accumulation.

Igue (1969) in his trials on the effect of N, P and K fertilizers on bean plants, found that significant interactions were most frequent where the effect of the separate nutrients were significant. He also found that responses were greater to P than N or K. At low levels