BACTERIAL SOFT ROT OF SUNFLOWER STEM PITH

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Abstract

Several strains of bacteria were obtained from the infected sunflower stem pith and studied. It was concluded on the basis of pathogenic and bacteriological characteristics that all strains belong to the bacterium *Pseudomonas marginalis* (Brown) Stevens, which is sunflower pathogen studied insufficiently.

Key words: *Pseudomonas marginalis,* bacterium, rot of sunflower stem pith.

Introduction

As it is known (Arsenijević, 1988), the bacterium *Pseudomonas marginalis* has been studied as a causal agent of spot or rot disease of lettuce leaf margins.

Up to now, this pathogen variety has been scarcely studied both in the world (Hunter and Cigna, 1989; Shinde and Lukezic, 1974; Friedman, 1951; Kohn, 1973) and in Yugoslavia (Arsenijević, 1990).

In 1988 and 1989, we obtained several samples of sunflower diseased stem from the region of Banat. The symptoms manifested in form of rot of sunflower stem pith.

The study of the pathogenicity as well as of bacteriological characteristics showed that the pathogen belongs to the bacterium *Pseudomonas marginalis.*

Symptoms

The changes on the obtained sunflower samples were observed most often on stem in form of dark necrotic pith rot.

Material and methods

Isolation and Pathogenicity of the Bacterium

Several strains of the bacteria of white colonies were obtained according to the standard procedure using the macerate obtained from the infected sunflower stem and sowing on NA medium by streaking.

The pathogenicity of these strains were tested according to hypersensibility, infiltrating tobacco leaves, using the suspension of the bacteria of 10^7cuf/ml concentration.

The inoculation of sunflower was performed by pricking with a needle and spraying the plants using the suspension of the bacteria of 10^7cuf/ml concentration.

Unripped fruits of tomato, pepper and cherry were also inoculated with needle using the suspension of bacteria of the concentration 10^8 cuf/ml.

The slices of carrot, and onion, fragments of cabbage and lettuce leaves, were inoculated using the same bacterial concentration.

The tissue of French bean pods was inoculated by infiltration using the suspension concentration of 10^7cuf/ml.

The inoculated plants of sunflower, unripped fruits of tomato, pepper and cherry and the slices of carrot and onion, fragments of cabbage and lettuce leaves, were kept in a wet chamber for 2-3 days and after that in a greenhouse and laboratory.

The pods of French beans were placed in large Petri dishes. All control plants and fruits were inoculated with top water.

Bacteriological Characteristics of the Pathogen

Morphologic, cultural and biochemical characteristics were studied applying conventional (Šutić and Panić, 1969) and some modern methods (Arsenijević, 1988, 1992).

The color, size and shape of the colonies on NA medium, King's medium B and the medium enriched with saccharose (NAS) were studied.

The differentiation of bacteria according to Gram was performed applying 3% KOH (Suslow et al., 1982, Arsenijević, 1988).

Maximum temperature of development, tolerance to NaCl, catalase activity, gelatine hydrolysis, production of ammonium, hydrogen sulfide, indole, reduction of nitrates, production of phosphatase, and lecithinase

activity were studied according to Šutić and Panić (1969); Arsenijević (1988, 1992); Fahy and Persey (1983); Lelliott and Stead (1987).

Lopat tests were also studied: production of levane, oxidase activity, rot of potato slices and arginine metabolism (Lelliott et al., 1966; Arsenijević, 1988,1992).

Results

Pathogenicity of the Studied Bacteria

Tobacco. No one of the studied strains caused hypersensitive reaction on tobacco leaves.

Sunflower. All sunflower isolates and *P. marginalis* from parsnip incited necrosis on stem cortical tissue already after two days. Inner pith of stem changed also: there came to necrosis, rotting and softening losing the firmness.

Four days after inoculation, there came to plant wilting and bacteria spread throughout the vessels.

Fruits of Tomato, Pepper and Cherry. Dark spots occurred on the inoculated fruits of tomato and pepper on the injecting spot, while negative results were achieved with cherry fruits as well as with the control strain *Pseudomonas marginalis* (Pm-3).

Carrot and onion slices and cabbage and lettuce leaf fragments. The scoring of symptoms on carrot slices was made 24, 48 and 72 hours after the inoculation. The soft rot appeared 3 days after, developing very slowly.

Considering the inoculation of cabbage and lettuce leaves, the studied strains brought to light brown, soft rot that covers a large part of the leaf. Similar changes were observed by Arsenijević (1990). The control strain *Pseudomonas marginalis* (Pm-3), also caused the soft rot on the slices, onion bulbs and leaves.

French Bean Pods. The studied strains of *Pseudomonas marginalis* from the infected sunflower stem caused light-brown, surface spots on the inoculated French bean pods that are characteristic for an incompatible parasite -host relationship (Klement, 1968) which is typical for hypersensitive reaction.

Bacteriological characteristics

Morphological characteristics.

The bacteria are rod-shaped, gramnegative with polar flagellation.

Cultural characteristics.

All strains of the investigated bacterium formed white colonies on AN medium.

The colonies on King's medium B are large and shiny while on a medium enriched with saccharose (NAS), the colonies are large, white, convex and shiny.

All strains on King's medium B produced fluorescent pigment which in agreement with the literature data (Arsenijević, 1989, 1990).

Biochemical characteristics.

Observing the standard biochemical and physiological characteristics, the studied strains showed unified results: the bacteria did not develop at 37°C, grew well in a medium containing 5% NaCl, produced catalase (Arsenijević, 1990), reduced nitrates lignified gelatine, produced ammonium, did not produced hydrogen sulfide and indole (Friedman, 1951), but produced phosphatase and lecithinase.

Considering Lopat tests and bacteriological characteristics of the studied strains, it can be concluded that they belong to the group IV of fluorescent bacteria from the genus *Pseudomonas* (Lelliott et al., 1966).

Discussion

The bacterium *Pseudomonas marginalis* has been considered as the causal agent of spot or rot disease of lettuce leaf margins (Arsenijević, 1988).

Additionally to lettuce, this bacteria can bring to changes on cauliflower (Arsenijević, 1990), chicory (Friedman, 1951), and parsnip (Hunter and Cigna, 1989).

In 1988 and 1989, we received the samples of sunflower with characteristic symptoms in the form of dark rot of stem.

The complete analysis of the separated isolates showed that the causal agent of sunflower stem pith rot is the bacterium *Pseudomonas marginalis.*

The inoculation of various host plants showed the differences with respect to symptom appearance. Necrotic spots occurred on sunflower stem.

The inoculation of carrot slices, onion bulbs, cabbage and lettuce leaf fragments with the studied strains caused light- brown, soft rot that continually spreads.

Light-brown spots that are characteristic for parasite-host incompatibility relationship occurred on French bean spots.

Considering biochemical and physiological characteristics of *Pseudomonas marginalis*, our results agree with the data from literature (Friedman, 1951; Arsenijević, 1990).

The results of Lopat tests showed that the studied bacteria correspond to the group IV of the genus *Pseudomonas* , i.e. to the bacterium *Pseudomonas marginalis* (Lelliot et al., 1966; Arsenijević, 1988, 1992).

Conclusion

All studied strains from the infected stem of sunflower showed several common characteristics of pathogenicity, cultural, biochemical and physiological nature.

These strains brought to the reaction similar to hypersensitive reaction on the inoculated French bean pods opposite to tobacco as there occurred negative reaction. The studied strains caused necrotic spots on sunflower stem.

The inoculation of carrot and onion bulbs slices, fragments of cabbage and lettuce leaves brought to the occurrence of light brown soft spots that gradually spread.

The strains of the studied bacteria are rod-shaped with polar flagellation, gramnegative and asporogenic. The colonies on NA medium are white.

All strains studied produce fluorescent pigment on King's medium B, do not develop at 37°C, grew well in the medium with 5% NaCl, produce catalase, lignify gelatine, produce ammonium, do not produce hydrogen sulfide and indole, produce phosphatase and lecithinase, reduce nitrates.

According to pathogenicity, morphological, cultural and biochemical characteristics, it can be concluded that the strains that we studied, isolated from the infected sunflower belong to the bacterium *Pseudomonas marginalis.*

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