

BACTERIAL PARASITES OF SUNFLOWERS (*Helianthus annuus* L.) IN YUGOSLAVIA

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SUMMARY

Bacterial isolates originating from sunflowers were examined for virulence, morphological, reproductive, biochemical, and physiological characteristics. The obtained results indicated them to belong to two different groups: *Erwinia carotovora* and *Pseudomonas syringae*.

INTRODUCTION

Bacterial parasites of sunflowers have generally been insufficiently studied. First bacteriosis to occur on sunflowers in Yugoslavia were observed in several locations in Vojvodina Province in the course of 1969 (Arsenijević, 1970). Since then, they occurred sporadically without causing extensive damage. In 1986, a bacteriosis causing rot was observed on some hybrids grown in the vicinity of Novi Sad. The pathogen has been identified as the bacterium *Erwinia carotovora* which is a well-known parasite of numerous plant species in all countries (Arsenijević and Maširević, 1987).

Continuing the investigation in 1987, the authors observed the occurrence of sunflower leaf spot caused by a bacterium that has not been registered before in this country.

MATERIALS AND METHODS

Isolation and virulence check. Bacteria were isolated on a mesopeptonic medium - nutrient agar (NA) - by the conventional procedure (Schaad, 1980; Fahy and Persley, 1983). The first virulence check of the obtained isolates was conducted by infiltration of tobacco and pelargonium (Klement, 1963; Arsenijević, 1987), using a suspension of bacteria at the concentration of 10^7 cells/ml.

Pathogenic characteristics of the first group of bacteria (*E. carotovora*) were additionally checked by inoculating sunflower stems with a suspension of bacteria at the concentration of 10^7 cells/ml. The suspension was applied with a brush, along a slit made in the epidermal tissue. The inoculated slits 3-4 cm long were then wrapped with wet cotton and covered with aluminum foil. Onion bulbs and parts of cabbage heads were inoculated with a needle, potato slices by pouring the bacterial suspension over them. The concentration of the bacterial suspension in the last two cases was 10^8 cells/ml.

The virulence of the second group (*Pseudomonas syringae*) was checked by spraying young sunflower plants with a bacterial suspension by means of a portable sprayer and an atomizer. The suspension of 10^8 cells/ml was used in the former case, the suspension of 10^6 in the latter. Peach annual shoots were inoculated by a needle (10^7 cells/ml) and bean pods by infiltration of a bacterial suspension (10^7 cells/ml).

Bacteriological characteristics. The fluorescence of both bacterial groups was determined on King's B substrate and the arrangement of cilia by means of an electronic microscope.

The development of oxidase, catalase, arginine dihydrolase, and other biochemical and physiological characteristics were examined by the conventional procedures (Schaad, 1980; Fahy and Pensley, 1933).

RESULTS

Virulence. Tobacco and pelargonium leaves, inoculated by the infiltration of the bacterial suspension of the carotovora group by means of a syringe, developed a hypersensitive reaction within 24 hours after the inoculation. However, only fresh isolates and reisolates were capable of inducing such a reaction. This capacity was lost after several sievings and the hypersensitive reaction on inoculated leaves did not occur. This has been observed earlier for isolates from this bacterial group (Arsenijević, 1970). Several days after the inoculation, brown-black to black spots appeared on sunflower stems. The spots quickly developed into a tissue rot which spread both lengthwise and sidewise, resembling closely the changes which occur in conditions of a spontaneous infection. The rotting and softening of the infected tissue lead to the breakage and wilting.

The inoculated potato slices, onion bulbs, and parts of cabbage heads displayed the typical symptoms of soft rot which kept developing and spreading the area of infected tissue.

The isolates from the *syringae* group caused a hypersensitive reaction on tobacco and pelargonium leaves but no reaction on potato slices.

The inoculated leaves of young sunflower plants developed moist and oily spots which kept increasing in size turning brown and dry. The spots merged, causing symptoms of mildew type on the leaves which went dry and died. The inoculated annual shoots of peach did not exhibit any changes except in the case of the check isolate (Pš-4), originating from wheat, which caused necrosis and wilting.

The isolates originating from sunflower induced the occurrence of light brown spots, as a sign of a hypersensitive reaction, on the inoculated bean pods. The isolate Pá-1 (from wheat) caused brown, concave spots typical of the bacterium Ps. s. pv. syringae van Hall. Reisolations were achieved easily, no matter if sunflowers were inoculated by isolates from the first or the second group of bacteria.

Bacteriological characteristics. The obtained results show that the two groups of bacteria differed considerably. The strains from the first group (*carotovora*) did not develop fluorescent pigment on King's E substrate, the strains from the second group (*syringae*) did. The former group had the peritrichous arrangement of the cilia, the latter group the polar arrangement. Neither group formed oxidase, arginine-dehydrolase, and indole, hydrolised starch, and proteolysed milk. Regarding the rot in potato slices, the two groups differed in the formation of catalase, H₂S, nitrites from nitrates, and the development at 36°C.

DISCUSSION

Judging from the available literature data, sunflower bacteriosis attract increasingly the attention of researchers. Bacteriosis are encountered in several countries which are major sunflower producers (Arsenijević, 1970; Fucikovskiy et al., 1978; Zurini, 1977; Mazzucchi and Bazzi, 1979; Richeson, 1981; Gudmestad et al., 1984; Piening, 1976; Stancescu and Severin, 1983).

These above data indicate two groups of bacteria, *E. carotovora* and *Ps. syringae*, as predominant bacterial parasites of sunflowers. This was confirmed for Yugoslavia by our results (Arsenijević and Maširević, 1987).

It is difficult to forecast which bacteria will occur on sunflowers and what their intensity will be. In any case, these parasites are potentially detrimental for sunflower production. It is an important task for phytopathologists and breeders to register their presence on time and study them in sufficient detail, in order to preclude extensive damage that may result from the destructive action of these pathogens on susceptible sunflower hybrids.

The studied isolates of the two bacterial groups exhibited considerable aggressiveness on the inoculated sunflowers, resulting either in bacterial rot or mildew. Discriminative reactions observed among the hybrids in respect to the former disease may be useful to breeders in developing resistant genotypes.

Regarding some physiological characteristics, the representatives of the *carotovora* group seem to invite also a taxonomic interest (Fucikowsky et al., 1978; Gudmestad et al., 1984; Arsenijević and Maširević, 1987). Namely, the formation of reductive substances from sucrose, exhibited by the studied isolates from sunflowers, has so far been considered specific for the bacterial population of *atroseptica* type.

CONCLUSIONS

In the course of 1986 and 1987, a number of bacterial isolates were made from samples taken from infected sunflower plants. The isolates were divided in two groups according to distinct characteristics: one group of isolates came from plants with the symptoms of bacterial rot, another from plants with the symptoms of leaf spot.

The representatives of both groups induced hypersensitive reactions on inoculated tobacco and pelargonium leaves. However, the members of the first group belonged to non-fluorescent bacteria, and the members of the second group to fluorescent bacteria. The former group had the peritrichous arrangement of the cilia, the latter the polar arrangement. Both groups were asporogenic, gram negative bacteria.

In addition to sunflower, the first group, the agent of soft rot, parasitized on onion bulbs and cabbage heads. It has different biochemical and physiological characteristics in relation to the second group.

Therefore, the first group belongs to *Erwinia* genus, *carotovora* group, being most similar to *E.c.ssp. carotovora* /Jones, 1901/ Bergey, Harrison, Breed, Hammer et Huntton, 1923 and *E.c.ssp.atroseptica* van Hall 1902/Dye 1969.

According to LOPAT tests and other bacteriological and pathogenic characteristics, the second group, the agent of bacterial spot and mildew of sunflower leaves, belongs to *Pseudomonas syringae* group, being most similar to *Ps.s.pv.helianthi* /Kawamura 1934/Young, Dye et Wilkie 1978.

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