

SUPPRESSION OF PHYTOPATHOGENIC FUNGI BY *PSEUDOMONAS* SSP. STRAINS

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Introduction

Among many modern and eco-friendly approaches in the plant health management, use of biopesticides, containing living microorganisms or their metabolites, seems a more sustainable and environmentally friendly way of plant disease control in crop production [Ram et al., 2018]. Among the various biocontrol agents, the *Pseudomonas* species are the promising candidates for agricultural application, due to the wide spectrum of enzyme activities and antimicrobial molecules production [Sheoran et al., 2015].

Materials and methods

Four *Pseudomonas* strains (16A1, 16B1, 23aP and 154), isolated from root nodules of wild leguminous plants growing in the southeastern part of Poland were characterized. Antagonism of bacterial isolates toward phytopathogenic fungi *Botrytis cinerea*, *Diaporthe rudis*, *Fusarium equiseti*, *Fusarium oxysporum* and *Sclerotinia sclerotiorum* was performed *in vitro* by the disc diffusion method using the potato dextrose agar (PDA) plates [Balouiri et al., 2016]. We also assessed their plant-growth promoting properties including production of siderophores, salicylic acid and hydrogen cyanide (HCN) [Arnow, 1937; Lorck, 1948].

Results

All the studied strains inhibited the growth of the phytopathogenic fungi on PDA agar plate assay (Fig. 1-4), but the degree of inhibition varied per endophyte. Strains 23aP and 16A1 were the most effective against phytopathogenic fungi, especially against the white mold disease agent, *Sclerotinia sclerotiorum*. Moreover, strain 16A1 inhibited the growth of all phytopathogenic fungi used in this assay (Fig. 5).



Fig.1



Fig.2



Fig.3



Fig.4

Fig. 1-4. The inhibition of phytopathogenic fungi growth after 14 days of incubation on PDA medium

Fig. 1. strain 16A1 and the growth inhibition zone of *S. sclerotiorum*; **Fig. 2.** strain 16A1 and the growth inhibition zone of *D. rudis*; **Fig. 3.** strain 23aP and the growth inhibition zone of *B. cinerea*; **Fig. 4.** strain 23aP and the growth inhibition zone of *D. rudis*

Suppression range of phytopathogenic fungi

	<i>Botrytis cinerea</i>	<i>Diaporthe rudis</i>	<i>Fusarium equiseti</i>	<i>Fusarium oxysporum</i>	<i>Sclerotinia sclerotiorum</i>
16A1	Green	Green	Green	Green	Green
16B1	White	Light Green	Light Green	White	White
23aP	Green	Dark Green	Green	White	Dark Green
154	White	Light Green	Light Green	Light Green	White

diameter [cm] of the fungi growth inhibition zone

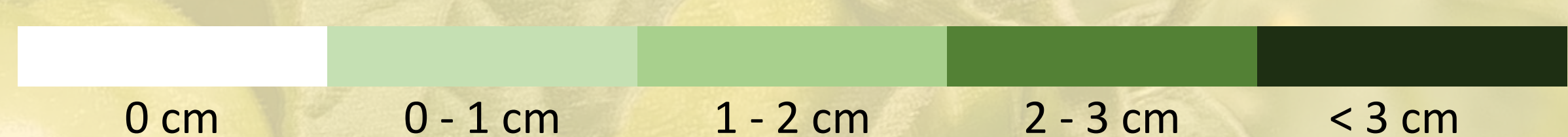


Fig. 5. Heatmap illustrating the antagonistic activity of *Pseudomonas* strains against phytopathogenic fungi

The results obtained showed that all tested strains were able to produce molecules with biocontrol potential as: hydroxamate siderophores, salicylic acid and hydrogen cyanide however, to a different degree. The strain 16A1 showed the highest level of hydroxamate siderophores production and a relatively high level of salicylic acid synthesis. While, strain 154 indicated the strongest reaction in the hydrogen cyanide test (Fig. 6).

	Siderophore [µg/ml]	Salicylic acid [µg/ml]	Hydrogen Cyanide + weak, ++ medium, +++ strong reaction
16A1	0,75	23	+
16B1	0,55	31	+
23aP	0,19	22	++
154	0,08	6	+++

Fig. 6. The production of hydroxamate siderophores, salicylic acid and hydrogen cyanide by tested *Pseudomonas* strains

Conclusions

Taking into account results of *in vitro* antagonistic assays observed as growth suppression of some phytopathogenic fungi, it can be concluded, that all studied strains has biocontrol potential and they seem to be a promising candidates to use in sustainable agriculture. However, due to no clear correlation was found between the production level of the tested metabolites and antifungal activity, it is not possible to indicate mechanisms underling these phenomena. It is likely that the synergistic action of these and other untested mechanisms contributed to observed results, therefore further investigations needs to be done.

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