

#### The Influence of Carbon Source on the Antagonistic Activity of Bacillus sp. Against the Aflatoxigenic Aspergillus flavus



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#### Introduction

Besides yield losses, considered a minor problem, mycotoxigenic fungi belonging to the Aspergillus flavus are recognized as a severe threat to food safety. Economic losses caused by crop contamination and harmful effects on human health due to aflatoxins entering the food chain indicate the high priority for developing the strategy to suppress those fungal pathogens. Global climate changes enhance conditions suitable for the development of mycotoxigenic fungi, even in the geographical regions characterized by a low incidence of their appearance. This kind of scenario makes the task mentioned above even more urgent and attracts the attention of scientific society, resulting in much research conducted to find a promising solution to address the existing problem. The growing trend of biopesticides in the world market proves the changes occurring in modern farming regarding plant disease management. The constantly rising popularity of biocontrol agents confirms it is just a matter of time when their domination over synthetic products will occur. Bacterial antagonists, with the emphasis on the Bacillus genus members, are considered leaders in the category of microbial biopesticides, highly effective in the suppression of phytopathogenic species. Even though biocontrol agents are defined as a versatile weapon in plant diseases control, there is a significant gap between the potential of microbial-based biopesticides and the number of products registered and utilized in agricultural practice. The explanation for this scenario could be found in high production costs and overall cost-effectiveness of the industrial level production. Cultivation medium preparation is one of the most critical elements in the cost structure of microbial biopesticides production (30–40% of total production costs). Carbon sources play a crucial role in the production of biocontrol agents and influence the growth of bacterial cells as well as the synthesis of secondary metabolites. Reducing the cultivation media preparation costs using alternative nutrient sources was recognized as a solution that could significantly decrease overall production costs. This approach would contribute to the industrialization of biocontrol agents' biotechnological production and result in increased chances of microbial pesticides' adoption and positioning in the world market.

## Objectives

The present study includes the selection of optimal carbon source for cultivation of novel isolate of *Bacillus* genus, originating from the rhizosphere sample of *Phaseolus vulgaris*. The antagonistic effect of the producing strain was evaluated against aflatoxigenic *Aspergillus* flavus isolated from the infected maize grown in the Republic of Serbia.

### Method

Seven different carbon sources, including glucose, fructose, glycerol, maltose, lactose, cellulose, starch, were used for cultivation media preparation. The selected carbon sources were chosen to represent the most common components of the industrial waste streams, which could serve as alternative sources of nutrients. The cultivation was carried out in an Erlenmeyer flask for 96 h, at the temperature of 28 °C, with an agitation rate of 180 rpm. The percentage of mycelial growth inhibition was estimated based on the antimicrobial activity testing of cultivation broth by the diffusion method. One-way ANOVA was performed to determine the carbon sources' effect statistical significance on inhibition zone diameter. Duncan's multiple range test was conducted to define homogenous groups of medium with different carbon sources with the same level of statistical significance regarding their effect on antimicrobial activity of the tested cultivation broth against tested aflatoxigenic strain.



# Conclusions

Cellulose being chosen as the most promising carbon source opens a new chapter of possibilities regarding agricultural waste utilization as an alternative source of a key nutrient. The conversion of lignocellulosic waste as a raw material in biotechnological production represents a promising approach for creating bio-based products in a cost-efficient way. Additionally, it is also important from the aspect of production sustainability and contributing to agricultural waste recycling. As the present study results revealed high antagonistic activity of the producing strain using a cellulose-based medium, further research will include the utilization of various agricultural waste in the media preparation. The following investigation would be a key step in developing viable and ecofriendly bioprocess solutions for the production of biocontrol agents, with the possibility of scaling up to the industrial level and potential commercialization.

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