

Antifungal Activity of *Bacillus amyloliquefaciens* Ethyl Acetate Extract and Fractions Against the Fungus *Khuskia oryzae*

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Fungal infections are common among plants and animals which result in economic losses. Finding new antifungal agents from alternative sources may help to solve the above issue. It was observed that, in culture *Bacillus amyloliquefaciens* isolated from a contamination shows antifungal activity against the fungus *Khuskia oryzae*. Therefore, the objective of the current study is to determine the antifungal activity of the crude ethyl acetate extract and the fractions of crude extract of *B. amyloliquefaciens* against the fungus *K. oryzae*. *B. amyloliquefaciens* was grown on Luria-Bertani Agar (LBA), extracted into ethyl acetate after an incubation period of three days and the antifungal activity of the crude extract was tested against *K. oryzae* at 400 lig disc⁻¹ using agar disc diffusion method. Crude extract of 1.5 g was first fractionated by Kupchan solvent-solvent partitioning scheme, sequentially using hexane, methanol/water (9:1); chloroform, methanol/water (6:4) and ethyl acetate, water. Antifungal activity of the three fractions hexane, chloroform and ethyl acetate was determined and the chloroform fraction was active against *K. oryzae*. The active fraction was further purified using Sephadex LH20 size exclusion chromatography using methanol as the eluent. Fractions were combined according to the thin layer chromatography (TLC) profiles and the antifungal activity was tested for the combined fractions (A-F). Flucanazole and methanol was used as the positive and negative controls respectively. Fraction C (32.8 mg) resulted from size exclusion chromatography of the chloroform fraction exhibited 18 mm radius inhibition zone against *K. oryzae* while none of the other fractions showed any activity. Activity of fraction C was similar ($p > 0.05$) to the activity of the positive control. However, TLC profile of the fraction C showed the presence of more than one compound. Thus, further purification of fraction C is necessary in order to isolate the active compound/s which may lead to a potential antifungal agent.

Keywords: *Bacillus amyloliquefaciens*, Antifungal, *Khuskia oryzae*, Fungal infections, Fractionation

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