

## Characterization of a Broad Range Antimicrobial Substance from *Pseudozyma aphidis*

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**Abstract :** Natural product-based pesticides may serve as an alternative to the traditional synthetic pesticides, which have a potentially damaging effect, both to human health and for the environment. Along with plants, microorganisms are a prospective source of such biological pesticides. A unique and active strain of *P. aphidis* (designated isolate L12, Israel 2004), an epiphytic and non-pathogenic basidiomycete yeast, was isolated in our lab from strawberry leaves. *P. aphidis* L12 secretions were found to inhibit broad range of plant pathogens. This work demonstrates that metabolites isolated from the biocontrol agent *P. aphidis* (isolate L12) can inhibit varied fungal and bacterial phytopathogens. Biologically active metabolites were extracted from *P. aphidis* biomass, using the organic solvent ethyl acetate. The antimicrobial activity of the extract was demonstrated, both in vitro and in planta. Using disk diffusion assays, the following inhibition zones were obtained: 43cm<sup>2</sup> for *Pseudomonas syringae* pv. tomato, 28.5cm<sup>2</sup> for *Xanthomonas campestris* pv. vesicatoria, 59cm<sup>2</sup> for *Clavibacter michiganensis* subsp. *michiganensis*, 34cm<sup>2</sup> for *Erwinia amylovora* and 34cm<sup>2</sup> for *Agrobacterium tumefaciens*. Additionally, strong inhibitory activity of the extract against fungi mycelial growth was established, with IC<sub>50</sub> values of 606µg ml<sup>-1</sup> for *Botrytis cinerea*, 221µg ml<sup>-1</sup> for *Pythium* spp., 519µg ml<sup>-1</sup> for *Rhizoctonia solani*, 455µg ml<sup>-1</sup> for *Sclerotinia sclerotiorum*, 2270µg ml<sup>-1</sup> for *Fusarium oxysporum* f. sp. *lycopersici*, and 2038µg ml<sup>-1</sup> for *Alternaria alternata*. The results of the in planta experiments demonstrated a dose-dependent reduction in disease infection. Significant inhibition of *B. cinerea* lesions on tomato plants was obtained when a spore suspension of this pathogen was treated with extract concentrations higher than 4.2mg ml<sup>-1</sup>. Concentration of 7mg ml<sup>-1</sup> caused a reduction of over 95% in the lesion size of *B. cinerea* on tomato plants. The strong antimicrobial activity demonstrated both in vitro and in planta against varied phytopathogens, may indicate that the extracted antimicrobial metabolites have potential to serve as natural pesticides in the field.

**Keywords :** antimicrobial, *B. cinerea*, metabolites, natural pesticides, *P. aphidis*

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