

Evaluation of the Chemical Profile and Antibacterial Activity of Extracts Obtained from the Endophytic Fungus *Colletotrichum dianesei*

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Endophytic fungi are microorganisms that live in the internal tissues of plants without causing apparent damage. Fungi of the genus Colletotrichum are little studied regarding its secondary metabolism, making it an excellent group of micro-organisms for the bioprospection of biomolecules. The conditions in which the organisms are exposed dictates the production of metabolites, many of these molecules are of great interest to the industry. Thus, the objective of this work was to perform the cultivation in different media of endophytic fungus Colletotrichum dianesei with was isolated from Palicourea corymbifera (Rubiaceae) and evaluates the antibacterial potential of its extracts. The cultivation media was composed of Czapek (CZ) with and without the addition of 0.2% yeast extract (12 repetitions each). The cultivation was performed in orbital shaker incubator at 120 rpm under the temperature of 30 °C for 20 days, after this period it was performed the filtration of the metabolite broth and mycelium. The mycelia were subjected to extraction with organic solvents: dichloromethane (DCM), ethyl acetate (AcOEt) and methanol (MeOH) each one by using ultrasonic bath during 20 minutes then the obtained extracts were concentrated in a rotary evaporator yielding 0.5884 g: 0.2319 g and 7.2421 g, respectively. The extracts were analyzed by thin-layer chromatography (TLC) using physical and chemical revealers, then 15 milligrams (mg) of each extract was solubilized in dimethyl sulfoxide-d₆ and analyzed by ¹H-Nuclear Magnetic Resonance (NMR) at 300 MHz (Bruker, Fourier model-300). The analyzes of ¹H-NMR of DCM extracts showed the presence of triglycerides, but only in the extract obtained from the add of yeast, showed signs of aromatic substances in the regions between 7 and 7.7 ppm. The AcOEt extracts without yeast extract showed few signs of aliphatic and aromatic substances, however, when yeast was add to media the extract showed greater diversity of signs of aliphatic and aromatic substances, plus signs in the region between 10.3 and 11.5 ppm, being indicative of indole alkaloids (together TLC). For MeOH extracts, all presented sugars, but only the extract obtained from the media add with yeast presented a greater diversity of signs of aliphatic substances in the regions between 5 and 6 ppm and aromatic substances between 6.3 and 8.5 ppm. So, the extracts obtained from the media add with yeast were assayed against the bacteria: Serratia marcescens (ATCC 13880), Edwardsiella tarda (ATCC 15947), Pseudomonas aeruginosa (ATCC 10145), Acinetobacter baumannii (ATCC 1906-143), Citrobacter freundii (ATCC 13316), Aeromonas hydrophila (ATCC 7966), Escherichia colli (ATCC 11775), Staphylococcus aureus (ATCC 12600) and Klebsiella pneumoniae (ATCC 13883). 10 mg of DCM, AcOEt and MeOH were assayed in micro dilution method to determine Minimal Inhibitory Concentration (MIC) at 500 and 1000 µg/mL, and measure at 625 nm. As positive control it was used oxytetracycline and DMSO as negative control. As results, DCM extract was active against Serratia marcescens (24,4% at 1000 µg/mL and 29,3% at 500 µg/mL). AcOEt extract was active against Edwardsiella tarda (20,2% at 1000 µg/mL) and 8,8% at 500 µg/mL against Serratia marcescens. MeOH extract was active against Serratia marcescens (22,3% at 1000 µg/mL and 11,4% at 500 µg/mL). Thus, this study indicates that there is a better performance for the production of secondary metabolites when the fungus is cultivated in CZ with the addition of yeast extract. So, these results encourage us to perform the chemical fractionation.

Keywords: Endophyte; Secondary Metabolites; Antibacterial activity.

Acknowledgments: To CNPq, CAPES and FAPEAM.