Culture medium modelisation for optimization of anti methicillin resistant *Staphylococcus aureus* metabolites by a coal mining soil derived *Streptomyces rochei* CMB47 strain

Ibtissem Djinni[1,2], Warda Djoudi[1], Chahinaz Boumezoued[1], Halima Barchiche[1], Samiha Souagui[1], Mouloud Kecha[1], Ines Mancini[2]

[1] Laboratoire de Microbiologie Appliquée, Faculté des Sciences de la Nature et de la Vie, Département de Microbiologie, Université de Bejaia, Bejaia 06000, Algeria
[2] Laboratory of Bioorganic Chemistry, Department of Physics, University of Trento, Via Sommarive 14, Povo, 38123 Trento, Italy

ibtissem.djinni@univ-bejaia.dz ; ines.mancini@unitn.it

The urgent need for the development of new antibacterial drugs arises from the global increase of antibiotic resistance. In addressing this challenge, *Streptomyces rochei* CMB47, isolated from a Saharan soil of a coal mine provided an ethyl acetate extract which exhibited a minimum inhibitory concentration of <0.439 µg/mL against MRSA, resulting therefore a promising source of bioactive metabolites. Medium formulation and optimization is essential for the success of an industrial production by fermentation, as it directly affects time and costs of the bioactive products. A statistical medium optimization experimental design employing a response surface methodology (RSM) based on a second-order rotatable central composite design (RCCD) was employed to optimize the fermentation process, in favor of an enhanced production of the metabolites responsible of the bioactivity. Optimal conditions for starch and $NaNO_3$ concentrations, incubation time, and initial pH were determined, resulting in an inhibition zone diameter of 20 mm, closely matching the experimental value after model validation. Bioassay-guided fractionation of the crude extract yielded the most active fractions, afterwards subjected to high-performance liquid chromatography equipped with a photodiode array detector and coupled online with electrospray mass spectrometry (HPLC-DAD/ESI-MS). These results provided preliminary insights into the molecular structures of the metabolites.

Keywords: Antimicrobials, Central composite rotatable design, Methicillin-resistant *S. aureus*, statistical optimization, LC-ESIMS analysis, *Streptomyces rochei*