10th Brazilian Conference on Natural Products
XXXVI RESEM
4-7 November 2025, Belo Horizonte, MG, Brazil
Section: 05

"OSMAC Approach Applied to Modulatethe Biosynthesis of Antimicrobial Natural Products in *Pseudonocardia* ICBG1860"

Guilherme José Brustello Vizin[1*], Rafael Gonçalves Padilha[1], Carlismari Oliveira Grundmann[2], Claudia Castania de Macedo[1], Mônica Tallarico Pupo[1]

gjbvizin@usp.br

[1] *School of Pharmaceutical Sciences of Ribeirão Preto, University of São Paulo, Ribeirão Preto, Brazil.*
[2] *Chemistry and Biochemistry Department, University of California, Santa Cruz, United States of America*

The rise of microbial resistance poses a critical challenge to public health, underscoring the urgent need for new strategies in antibiotic discovery and development. Actinobacteria of the genus *Pseudonocardia*, symbionts of attine ants, are well known for producing bioactive metabolites with ecological and pharmaceutical relevance. In this study, we applied the One Strain Many Compounds (OSMAC) approach to *Pseudonocardia* sp. ICBG1860, which was isolated from *Paratrachymyrmex* ants. The study was conducted under the hypothesis that modifying culture conditions could activate silent biosynthetic pathways and promote the production of minor or novel antimicrobial metabolites. Among these metabolites is a putative tetracycline analogue, detected in previous work (GRUNDMANN, 2023), which is the main target of this study. The strain was cultivated in 11media providing diverse nutrients, sugars, cofactors, and pH conditions. Solid cultures were extracted with ethyl acetate, while liquid cultures were processed using HP20 resin and acetone. The resulting extracts underwent solid-phase extraction (SPE, C18 cartridge) and were analyzed by HPLC-DAD-ELSD, enabling comparison of chromatographic profiles and identification of promising culture conditions. Among the tested media, D-Mannitol–Peptone Agar stood out by inducing a unique set of secondary metabolites that were absent under the other evaluated conditions. UV-Vis spectra suggested the presence of tetracycline analogues, while ELSD data revealed a marked increase in the production of a putative novel tetracycline analogue, previously detected only at low concentrations in ISP2 medium (GRUNDMANN, 2023). Accordingly, this medium was selected for large-scale cultivation (450 Petri dishes, 90 mm), followed by SPE fractionation into six fractions. The MeOH:H_2O (50% and 75%) and MeOH 100% fractions concentrated the elicited compounds and exhibited antimicrobial activity in overlay assays against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escovopsis* sp., a fungal pathogen of attine gardens. These results highlight the effectiveness of the OSMAC approach in modulating the metabolism of *Pseudonocardia* ICBG1860 and reinforce its potential as a source of novel bioactive compounds. Next steps include HPLC-MS/MS analysis, dereplication through molecular networking using the Atlas of Natural Products and GNPS platforms, followed by isolation and structural elucidation of the metabolites by NMR. The authors thank their institutions and acknowledge financial support from CAPES (Finance code 001) and FAPESP (grants 2013/07600-3; 2025/10223-4; 2024/21508-7).

Keywords: *Pseudonocardia, antimicrobial, tetracycline, natural products, symbiosis, OSMAC*