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

Section: 02

ISOLATION OF GLYCOSYLATED FLAVONIDS FROM THE ETHYL ACETATE LEAF EXTRACT OF Siparuna cymosa Tolm. *BY COUNTERCURRENT CHROMATHOGRAPHY*

Adérito José Cavele[1], *Diégina Fernandes Araújo*[1], *Gilda Guimarães Leitão*[1*]

ajcavele2@gmail.com; *ggleitao@ippn.ufrj.br

[1] Laboratório de Fitoquímica e Cromatografia contracorrente – Instituto de Pesquisa de Produtos Naturais – Universidade Federal do Rio de Janeiro - 21491 599, Brazil.

Countercurrent chromatography (CCC) is a liquid–liquid separation technique based on the partitioning of solutes between two immiscible liquid phases, one of which is retained as the stationary phase within the column, while the other is used as the mobile phase during the chromatographic process. This technique has proven to be highly effective for the isolation of secondary metabolites from complex matrices. In this context, *Siparuna cymosa* Tolm., an endemic Brazilian species from the Siparunaceae family, also popularly known as *erva-do-rato*, has been extensively studied in the field of natural products due to its rich diversity of secondary metabolites, particularly alkaloids and flavonoids. Previous studies have demonstrated that extracts of this species, especially the ethyl acetate extract (SCA), exhibit a broad range of pharmacological activities, including antiviral effects against influenza virus (H1N1) and SARS-CoV-2, potential antivenom activity against *Bothrops jararaca*, and antitumor activity against myeloid cell lines. Given this therapeutic potential, the present study aimed to isolate secondary metabolites from the SCA by CCC fractionation. The fractionation was carried out using the biphasic solvent systems HEMWat (hexane-ethyl acetate-methanol-water, 3:6:3:6, v/v) and EBuWat (ethyl acetate-butanol-water, 8:2:10, v/v), employing high-speed countercurrent chromatography (HSCCC) on an HTPrep apparatus equipped with a 120 mL column (2.0 mm i.d.), operated in normal mode (head-to-tail). The mobile phase was delivered at a flow rate of 2.0 mL/min. A total of 60 fractions were collected - 30 during the elution phase and 30 during the extrusion phase - with each fraction having a volume of 4 mL. The collected fractions were monitored by thin-layer chromatography (TLC), grouped according to chromatographic similarities, and analyzed by nuclear magnetic resonance (NMR) spectroscopy. The findings, supported by comparison with reported chemical shifts in the literature, suggest the presence of glycosylated flavonoids in the analyzed fractions. The purified compounds are currently being analyzed by liquid chromatography coupled with mass spectrometry (LC-MS), as well as by ^{13}C-NMR and 2D NMR techniques (COSY, HSQC, HMBC) for full structural elucidation. Ongoing studies aim to further characterize the isolated substances and evaluate their biological activities.

Keywords: *Siparunaceae, Countercurrent chromatography, Bioactivity compounds, Flavonoids.*