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PHYTOCHEMICAL CHARACTERIZATION OF ZIZIPHUS (RHAMNACEAE) SPECIES AND THE ISOLATION OF METABOLITES BY COUNTERCURRENT CHROMATOGRAPHY (CCC)

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The genus *Ziziphus* (Rhamnaceae) is a promising source of bioactive secondary metabolites, particularly cyclopeptide alkaloids, which exhibit several pharmacological activities, including the potential to inhibit enzymes like α-glucosidase, a key therapeutic target in diabetes management (1). This work focused on the phytochemical study of the Brazilian native species *Ziziphus joazeiro* and *Ziziphus undulata*, employing chromatographic techniques to isolate their chemical constituents, and evaluate their biological activities. Ethanolic extracts were prepared from the leaves and twigs of *Z. joazeiro* and *Z. undulata*. The crude extracts were subjected to liquid-liquid partitioning and fractionation by Countercurrent Chromatography (CCC), a support-free liquid-liquid partition technique. Biphasic (BPSS) and triphasic solvent systems (TPSS) were used to optimize the separation. Additionally, an acid-base extraction was performed to obtain alkaloid-enriched fractions. The phytochemical profile of the samples was monitored by Thin-Layer Chromatography (TLC) using specific spray reagents. The TLC analysis confirmed the presence of several classes of metabolites, including flavonoids, terpenoids, and alkaloids, in the extracts and fractions. The fractionation of the crude ethanolic extract from *Z. joazeiro* leaves (ELZJ) by CCC using the BPSS HEMWat (6:1:6:1, *v/v/v/v*) proved to be particularly effective, allowing for the direct isolation of a major compound, which was identified as methyl pheophorbide A (MPhA) by spectroscopic analysis by ^{1}H NMR (2). This study validated CCC as a practical tool for the fractionation of *Ziziphus* extracts, enabling the isolation of MPA in a single chromatographic step. The identification of diverse metabolic classes reinforces the pharmacological potential of the genus and establishes a foundation for further investigation. Future steps will include the application of TPSS and the use of molecular networking for dereplication and characterization of new compounds. Biological assays will be conducted to assess the therapeutic potential of the isolated substances.

(1) EL MAAIDEN, E. *et al*. J. Ethnopharmacol, 259, 112950, 2020.
(2) HA, J. Y. *et al.* Appl. Biol. Chem., 65, 1, 52, 2022.

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