

## COMPARATIVE STUDY OF ANTILEISHMANIAL COMPOUNDS EXTRACTION METHODS IN *Moringa oleifera* LAMARCK

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### Introduction

*Moringa oleifera* Lamarck is a medicinal plant native to northeastern India with numerous pharmacological properties.<sup>[1]</sup> The antileishmanial activity of its bioactive compounds may represent a source for the discovery and production of new drugs, especially for visceral leishmaniasis, the most severe form of the disease.<sup>[2,3]</sup> As the protozoan *Leishmania* depends on the purine salvage pathway for nucleotide synthesis, this pathway becomes an important source of biological targets.<sup>[4]</sup> Thus, the study compares the chemical profiles of extracts from the leaves of *M. oleifera* obtained by different methods, aiming the future search for antileishmanial active compounds.

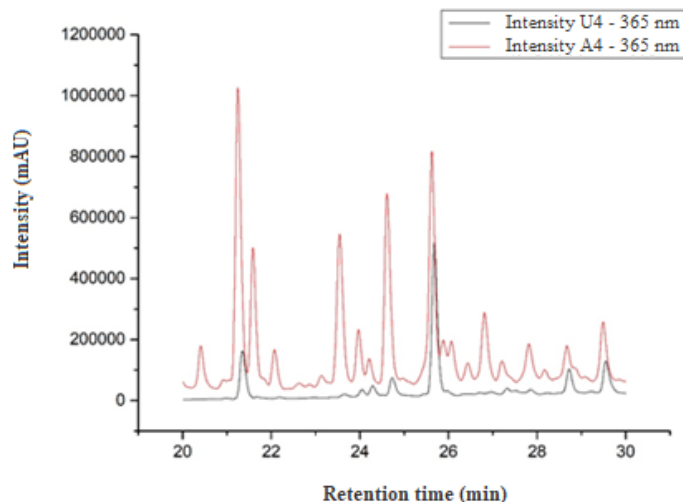
### Method

Air dried leaves of *M. oleifera* were supplied by the Herbarium of the Federal University of Goiás (UFG). Leaf extracts were prepared using two techniques: infusion and ultrasound-assisted solvent extraction. For each technique, experiments in triplicate were performed, using different solvents: ultra-pure water or ethanol P.A. grade. Extracts were dried, the residue was diluted in MeOH (ethanolic extracts) or water (aqueous extracts), homogenized in a vortex and analyzed by HPLC. The chromatographic conditions were: a C18 column (25 x 0.46 cm, 5 µm), gradient elution with 0.1% formic acid buffer/methanol, 0.6 mL/min flow rate and 20 µL injection volume. Detection was monitored at 280 and 365 nm.<sup>[5,6]</sup>

### Results / Discussion

The exploratory data analysis allowed the selection of the best extraction technique and solvent. The chromatograms obtained by the HPLC/DAD system were superimposed to compare the degree of extraction of bioactive compounds. Water was the best extraction solvent in both extraction techniques used. The samples obtained by the infusion technique presented a higher number of peaks, with slightly higher intensity, when compared to the samples extracted by the ultrasound-assisted technique, at 365 nm (Figure 1) as well as at 280 nm. These wavelengths are commonly used in the detection of extracts from natural products and reveal the presence of flavones, flavonols and flavonones at 285 nm, and the presence of flavonoids at 365 nm.<sup>[7]</sup> Phenolic compounds and derivatives are usually detected at 280 nm and alkaloids have an absorption band between 268-282 nm.<sup>[8]</sup>

**Figure 1:** Superimposed chromatograms (20-30 min) of 30 mg/mL aqueous extracts obtained by the ultrasound-assisted (U4) or infusion (A4) techniques. Detection at 365 nm.



In the available literature, some compounds obtained from the leaves and roots of *M. oleifera* with inhibitory activity against the protozoan *Leishmania* were identified. This activity has been attributed to the glycosides thiocarbamate, carbamate and nitrile.<sup>[9]</sup> These compounds are detected at 220 nm, a wavelength considered nonspecific, since many organic compounds absorb at this energy.<sup>[10]</sup>

## Conclusion

From the results obtained, the best solvent extractor and extraction method were water and infusion, respectively. The chemical profile of the extracts obtained from the leaves of *M. oleifera* was not fully elucidated using the HPLC/DAD system. Further studies are needed to confirm the compounds in order to explore the inhibitory profile of the NH enzyme. For a better analysis of chemical profile of the extracts, the LC/MS coupled to a mass spectrometer with ESI ionization source can be used.

## Acknowledgments

The authors would like to acknowledge the Herbarium of the Federal University of Goiás (UFG) for the plant samples provided and the financial support from the Brazilian agencies CAPES (Finance Code 001) and FAPERJ (Emergency Support for Stricto Sensu Graduate Programs and Courses in the State of Rio de Janeiro Project E -26/200.930/2017).

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