

ISOLATION AND CHARACTERISATION OF FLAVONOIDS FROM Ptychopetalum olacoides Benth.

Karen Danielle Borba Dutra, <u>Arthur Ladeira Macedo</u>, Alessandra Leda Valverde, Thatyana Rocha Alves Vasconcelos, Rosane Nora Castro and Rosângela de Almeida Epifanio (*in memoriam*)

Department of Organic Chemistry, Chemistry Institute, Federal University Fluminense, Outeiro de São João Batista s/n, 24020-141 - Centro, Niterói RJ, Brazil

Abstract: Purpose of study: Ptychopetalum olacoides Benth. belongs to Olacaceae family, popularly known as muirapuama [1]. Preparations with the stem of the species P. olacoides are used to "weakness of the nerves," where the main symptoms are fatigue, depression, tremors, and sexual impotence [2], however, little is known about the phytochemical profile of the species. Therefore, the aim of the present work is contribute for the chemical knowledge of this species. Methods: The powder of bark and wood of P. olacoides (500g) was extracted with a solvent gradient of polarity under sonication using hexane, ethyl acetate and methanol, respectively. An amount of 2.6 g of the methanolic extract was restructured in methanol and was chromatographed using Sephadex LH-20 using MeOH as eluent. A total of 438 fractions were analyzed by thin layer chromatography (TLC) on silica gel using Hex: EtOAc (1:1) (v/v), revealed with UV lamp and 1% vanillin solution in ethanol and gathered to 12 fractions (MEF1-12). MEF8 (20 mg) was subjected to separation by preparative HPLC using Phenomenex C18 column (30 cm x 10 mm x 5 microns) with mobile phase consisting of a mixture of H₂O/AcOH (99:1) (solvent A) and MeOH (solvent B) at a constant flow rate of 5 mL/min using 65% of B and 35% of A in a isocratic elution mode. Three compounds were isolated and analyzed by ¹H and ¹³C NMR, UV and LC/MS. Results: Compounds were identified as 3-O-methylquercetin (1), 3,4'-O-dimethylquercetin (2) and 3,7-O-dimethylquercetin (3) (Figure 1). NMR data of compounds 1-3 were in agreement with those reported in the literature [3]. UV analysis with AlCl₃/HCl confirmed the presence of a free –OH in C-5 and the absence of O atom in C-6 for all compounds and the presence of free –OH in C-3' and C-4' for the compounds 1 and 3 [4]. Conclusions: This class of compounds has already been identified in this species however they were isolated for the first time in P. olacoides.

References:

- [1] Silva, R.A.D. 1925. Plantas medicinaes Brasileiras Estudo Botânico e Pharmacognostico. Rev. Bras. Med. Pharm. 1: 37-41.
- [2] Siqueira, I.R., Fochesatto, C., da Silva, A.L., Nunes, D.S., Battastini, A.M., Netto, C.A., Elisabetsky, E. Ptychopetalum olacoides, a traditional Amazonian "nerve tonic", possesses anticholinesterase activity. 2003. Pharmacol. Biochem. Behav., 2003. 75: 645-650.
- [3] Shi, Z.H., Li, N.G., Tang, Y.P., Li, W., Yin, L., Yang, J.P., Tang, H., Duan, J.A. 2012. Metabolism-based synthesis, biologic evaluation and SARs analysis of O-methylated analogs of quercetin as thrombin inhibitors. Eur. J. Med. Chem. 54: 210-222.
- [4] Markham, U.R. 1982. Techniques of flavonoid identification. Academic Press. London.

Figure 1: Flavonoids present in the methanolic extract of the species *P. olacoides*.