

Oct. 26-29th 2015

BIOSYNTHESIS OF SECONDARY METABOLITES IN Peperomia obtusifolia: A PROTEOMIC APPROACH

<u>Andrea N. L. Batista^{*}, ^a José R. A. Santos-Pinto, ^b Silvia N. López, ^c Massuo J. Kato, ^d</u> <u>Mario S. Palma^b and Maysa Furlan^a</u>

^aInstituto de Química, Universidade Estadual Paulista – UNESP, Araraquara-SP, Brazil, ^bInstituto de Biociências, Universidade Estadual Paulista – UNESP, Rio Claro-SP, Brazil, ^cFacultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario, Rosario, Argentina, ^dInstituto de Química, Universidade de São Paulo – USP, São Paulo-SP, Brazil *andrluca@yahoo.com.br

Purpose of study: *Peperomia obtusifolia*, an ornamental plant from Piperaceae family, accumulates bioactive secondary metabolites such as prenylated chromanes, lignans, amides, flavonoids and others phenolic derivatives [1,2]. Interestingly, this species seems to produce orsellinic acid-derived benzopyrans only, with some of them being isolated as racemic mixtures. The latter fact suggests either a chemical cyclization of the geranylated orsellinic acid precursor or the presence of two separate enzymatic systems devoted to the formation of each enantiomer. Therefore, *P. obtusifolia* represents an interesting plant for biosynthetic studies. Herein we report a comparison between the protein (soluble and microsomal fractions) and metabolic profiles of roots, stems and leaves of *P. obtusifolia* using shotgun proteomics and chromatographic methods.

Methods: Tryptic peptides were analyzed using an UFLC-DAD system coupled with mass spectrometry (ESI-IT-TOF). Proteins were identified from online databases (Swiss-Prot and NCBI) using the MASCOT protein search engine within Viridiplantae taxonomy. The metabolic profiles were determined by using HPLC-PAD-MS and GC-MS analyses.

Results and Conclusions: The shotgun proteomic analysis of the solution and microsomal fractions of different organs from *P. obtusifolia* led to the identification of more than 1500 proteins. Although the majority of these proteins were associated with the primary metabolism, a significant number of proteins involved in the secondary metabolism were also found (5% in soluble fraction and 20% in microsomal fraction), including enzymes related to the biosynthesis of prenylated chromanes, flavonoids and terpenes. These findings were corroborated by the metabolic profiles of the crude extracts of different organs from *P. obtusifolia* obtained via HPLC-PDA-MS and GC-MS analyses.

References:

[1] Mota, J.S., Leite, A.C., Batista, J.M., López, S.N., Ambrósio, D.L., Passerini, G.D., Kato, M.J., Bolzani, V.S., Cicarelli, R.M.B. and Furlan, M. 2009. *In vitro* trypanocidal activity of phenolic derivatives from *Peperomia obtusifolia*. Planta Medica 75:620-623.

[2] Batista, J. M., Batista, A. N. L., Mota, J. S., Cass, Q. B., Kato, M. J., Bolzani, V. S., Freedman, T. B., López, S. N., Furlan, M. and Nafie, L. A. 2011. Structure elucidation and absolute stereochemistry of isomeric monoterpene chromane esters. J. Org. Chem. 76: 2603-2612.