Fuente: www.fitoterapia.net

SL21 Characterisation of St. John's wort extracts by multivariate analysis of spectroscopic data

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Herbal medicines, produced from plant materials, often present a unique problem for manufacturers desiring the characterisation, reproducibility and standardisation that are required of pharmaceuticals. This problem is primarily due to the plurality of components contained in a herbal medicine and the large variation in composition. For the majority of plant extracts there is no evident correlation to be found between pharmacological activity and certain characteristic compounds. Although it is evident that the whole extract must be considered as the therapeutic agent, the characterisation is carried out referring to one single -often inactive- compound. Accordingly, there is a need to provide alternative methods for standardising complex botanical materials.
In our present work we choose a new approach for the classification of thirty different extracts of *Hypericum perforatum* adopting a method that has been developed in studies of "metabolic profiling" (1). We show the application of proton NMR spectroscopy as a very general analytical chemical tool for the characterisation of crude plant extracts. This technique can quantitatively and simultaneously detect all proton-bearing compounds and consequently all relevant substance classes in the samples. However, the spectra obtained are too complicated to be analysed visually. Therefore, the classification of spectra in this study was carried out using several multivariate statistical methods: Principal component and discriminant analysis as well as nonlinear regression techniques were used for the visualisation of the complex data set. In order to correlate the spectral data with pharmacological information, we describe the calibration of a quantitative model using a PLS algorithm. We also show that principal component loading plots and factor spectra are an effective tool in the interpretation of the differences between the substance composition of each extract.

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SL22 Ability of hederacolchiside A_1 to bind melanin may partly explain its strong antiproliferative activity on human melanoma cells

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It was demonstrated that hederacolchiside A_1 (Hcol-A_1), a monodesmoside from *Hedera colchica* K. Koch, with the sugar sequence O-L-rhamnopyranosyl (1→2)-α-L-arabinopyranoside at C3 of oleanolic acid and a complementary glucopyranosyl moiety branched at C1 of arabinose, exhibits *in vitro* stronger anti-proliferative effects in human malignant melanoma cell-line M4 Beu (IC_{50}: ca 4.5 µM) than in a panel of carcinoma cells, with differential cytotoxicity versus normal fibroblasts (IC_{50}: ca 7.5 µM) (1). The present study focused on mechanisms involved in the stronger activity on melanoma cells. Complementary investigations on four melanoma cell-lines, showed the weakest activity on human melanoma M3Dau, a cell-line which do not express melanin, suggesting that the anti-melanoma activity of Hcol-A_1, might be partly related to a specific ability to bind melanin. This hypothesis was verified by *in vitro* experiments with a new NMR technique, high resolution magic angle spinning and, insoluble synthetic melanin. ^1H spectra of Hcol-A_1 in D_20 phosphate buffer pH 7.4 were recorded at 500 MHz with a Bruker AVANCE DRX spectrometer fitted with a HRMAS probe, in presence and in absence of melanin. Interaction with melanin was demonstrated by a concentration-dependent linear broadening of line-widths which allows to determine the equilibrium dissociation constant (Kd) saponin-melanin.

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