

Conference abstract PO-55

Analysis of *Citrullus colocynthis* Cucurbitacine Derivatives with HPLC-SPE-NMR

S. STURM, P. SCHNEIDER, C. SEGER, H. STUPPNER

Institute of Pharmacy / Pharmacognosy, University of Innsbruck, Innrain 52, 6020 Innsbruck, Austria

E-mail: sonja.sturm@uibk.ac.at (S. Sturm)

Sci Pharm. 2009; 77: 254

doi:10.3797/scipharm.oephg.21.PO-55

Cucurbitacines are well known for their bioactivities and toxicity [1]. From the analytical point of view, this class of triterpene derivatives holds some challenges, due to their structural similarity and the presence of glycosides.

In *Citrullus colocynthis*, the colocynth, a series of structurally related glucoside derivatives, the cucurbitacines E, I, J, K, and L are present. These are solely differentiated by subtle changes in the C-8 sidechain. With classical phytochemical approaches, this analyte class is well tackleable. However, the isolation of mg amounts needed for conventional NMR based structure characterization requires a remarkable investment of manpower, lab-time and consumables [2]. Hence, minimizing the analyte amount needed for identification (without losing the required chemical information) is a major goal of modern phytochemistry. This goal has been recently realized by the HPLC-SPE-NMR platform allowing to obtain NMR information from μg amounts of analytes [3]. Combining the analytical HPLC based separation of the cucurbitacines with the trapping capabilities and the structural characterization power of the SPE-NMR device enabled the recording of all conventional 1D and 2D homo- and heteronuclear correlation spectra of *Citrullus* cucurbitacines from a crude methanolic extract of the plant material. The timeframe for recording a complete data set ranged between 18 and 24 hours and is comparable to off-line data acquisition times of 4–5 mg sample material. Comparison of different SPE stationary phases showed the superior performance of the C-2 material and the GP (general purpose) resin, whereas C-8 and C-18 phases were of limited usability. Our data demonstrate, that HPLC-SPE-NMR is a valuable analytical platform with a broad applicability. The trap and release process of the online SPE device linking the HPLC and NMR instruments needs careful optimization of at least each analyte class investigated.

- [1] Bauer R, Wagner H. Cucurbitacin-containing drugs. Analysis and standardization of medicinal drugs and plant preparations by high-performance liquid chromatography (HPLC) and other chromatographic methods. *Dtsch Apoth Ztg.* 1983; 123: 1313–1321.
- [2] Seger C, Sturm S, Mair ME, Ellmerer E, Stuppner H. ^1H and ^{13}C NMR signal assignment of cucurbitacine derivatives from *Citrullus colocynthis* (L.) Schrader and *Ecballium elaterium* L. (Cucurbitaceae). *Magn Reson Chem.* 2005; 43: 489–491. doi:10.1002/mrc.1570
- [3] Seger C, Sturm S. HPLC-SPE-NMR - a novel hyphenation technique. *LC-GC Europe.* 2007; 20: 587–597.