B035 TLC detection of the iridoid glucoside aucubin and the phenylethanoid acteoside in Plantaginis lanceolatae folium

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The crude drug Plantaginis lanceolatae folium is described in the DAB (1) and a proposal for a monograph for the European Pharmacopoeia (Ph.Eur.) has been published (2). Characteristic constituents in the crude drug are the iridoids aucubin and catalpol and the phenylethanoid acteoside (3). Aucubin has a liver protective activity (4). Significant antimicrobial activity of the aglycone part of aucubin and catalpol has been found and could nicely be demonstrated using a thin-layer chromatographic bioassay (5). The phenylethanoid acteoside is most likely accountable for an anti-oedemic activity (3). Whereas for the TLC-identification in the DAB monograph (1) aucubin is used as a marker, for the Ph.Eur. (2) it is proposed to use acteoside.

Here we present an easy TLC detection method for both aucubin and acteoside. Using the solvent system, proposed for the Ph.Eur. (2), the iridoid glucoside **aucubin** can be detected on the TLC plate by only heating at about 120°C for 5-10 min. In day light aucubin is visible as a blue zone and in ultraviolet light at 365 nm as a red-brown fluorescent zone. For the detection, **formic acid** (present in the mobile phase) is essential. The detection is very specific; the iridoid glucosides catalpol, harpagoside, gentiopicroside give no such a coloration. The phenylethanoid **acteoside** can also be detected on the TLC plate by only heating. In day light acteoside is visable as a yellow zone, and in ultraviolet light at 365 nm as a blue fluorescent zone. The blue fluorescent zone in ultraviolet light at 365 nm can also be seen without heating. In the new Ph.Eur. monography of Plantaginis lanceolatae folium (6), this new TLC detection method has been adopted.

References: 1. Deutsches Arzneibuch (DAB) (1999). **2.** Pharmeuropa (1999) 11, 513-514 **3.** Wichtl, M (Ed.) (1997) Teedrogen, ein Handbuch für Apotheker und Ärzte, 3e Ed. 443-446, WVG, Stuttgart. **4.** Chang I.M., Yun H.S., in Chang H.M et al. (Ed.) (1985) Advances in Chinese medicinal materials research: 269-285. World Scientific Publ. Co. Singapore. **5.** Van der Sluis W.G. et al. (1983) J. Chromatogr. 59: 522-526. **6.** Ph. Eur. supplement 4.3 (2002).

B036 Occurrence of phenolic substances in artichoke residues

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The common disposal of the artichoke wastes proceeding from the tinning industry process is as organic mass, animal feed or fuel. In order to assess a new usage of this residue, its phenolic composition was tested. The found compounds exhibit activity as free radical scavengers and quenchers of reactive oxygen species (ROS) (1-3).

The phenolic fraction from an artichoke waste extract supplied by Euromed S.A. (Mollet del Vallés, Spain) was purified by a liquid-liquid partioniting. A clean-up of the sample using Sephadex LH-20 was carried out to eliminate interferences. The different fractions were monitored by thin layer chromatography performed in a mixture of ethyl acetate / water / acetic acid and visualyzed under UV (254 and 360 nm). The similar fractions were joined to gave six final fractions. Liquid chromatography coupled to ionspray mass spectrometry in tandem mode (LC-DAD-MS/MS) with negative ion detection was used to identify a variety of phenolic compounds. Mass spectrometric studies were carried out using an API 3000 triple quadrupole mass spectrometer (Applied Biosystems) equipped with a Turbolonspray source. A C_{18} Luna (Phenomenex) column 50 x 2.1 mm i.d. 3.5 µm particle size was employed in a linear gradient profile with water and acetonitrile both containing 0.1% formic acid. Different approaches were used for the positive identification of a given compound: comparison of retention time and product ion scan spectrum with those of a standard and also the use of other MS modes such as the neutral loss scan to rapid test the presence of glycosides in artichoke samples. As a result of this study, gallic, chlorogenic, protocatechuic, quinic and caffeic acids and their quinic derivatives (caffeoylquinic compounds), esculin, cynarine, scolimoside, catechin, epicatechin and their gallate derivates, luteolin and luteolin-glycosides, naringenin, apigenin, apigenin-glycosides and diglycosides, quercetin and quercetin-glycosides, rutin and kaempferol-3-rutinoside, were identified.

Acknowledgements: CRAFT-Project FAIR-CT-98-9517 financed by the EC. Euromed S.A. is acknowledged by supplying the artichoke extract.

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