

QK10

### Freie Aminosäuren in Glycerolmazeraten von *Ribes nigrum* L.

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**Einführung:** Die Untersuchungen befassen sich mit freien Aminosäuren (fAS) der Schwarzen Johannisbeere (*Ribes nigrum* L.), welche in der Gemmotherapie (Knospentherapie) erfolgreich eingesetzt wird.

**Ziel:** Ausgewählte fAS in verschiedenen Knospen- und Blattmazeraten von *Ribes nigrum* L. sollen identifiziert und quantifiziert werden.

**Methode:** Zur Bestimmung wurden eine abgewandelte HPTLC-Methode (s. Poster) gemäss Lapke [1] und eine HPLC-Methode der ZHAW (s. Poster) eingesetzt.

**Resultat:** Das 2012 hergestellte Knospenmazerat wies für Ala, Arg, Asp, GABA und Gln die höchsten Konz. mit 15–37 mg/g Knospe auf. Ältere Mazerate wiesen vergleichbare Fingerprints in geringerer Konz. auf. Eine Charge aus dem ausländischen Grosshandel wies deutlich geringere fAS auf. Die Knospenmazerate zeigten weit höhere Konz. als die zum Vergleich gemessenen Blattmazerate (Asparagin bis zu 200fach).

**Diskussion:** Das Vorkommen von in der Literatur erwähnten fAS in Blättern und Knospen [2] konnte für Knospenmazerate von Gemmo-Arzneimitteln der Firma Spagyros AG bestätigt werden.

Um den Einfluss von Bedingungen wie Herkunft oder Erntezeitpunkt auf den Gehalt fAS zu klären, bedarf es weiterer Untersuchungen.

Der im Vergleich zu Blattmazeraten höhere Gehalt von fAS in Knospenmazeraten kann als Fingerzeig gedeutet werden, dass Knospen einen von den Blättern abweichenden Primärstoffwechsel besitzen.

Das Ergebnis ermutigt, Knospen als eigenständigen Pflanzenteil einer Heilpflanze bzw. die Gemmotherapie als entsprechende Therapieform weiter zu untersuchen.

#### Referenzen:

- 1 Lapke C, et al: Detection of amino acids via TLC as a rapid method for the screening of hop and valerian. *Revista de Fitoterapia* 2002;2:S1
- 2 Totan M: *Ribes nigrum* Knospen. Ein natürliches pflanzliches Mittel gegen Entzündungen. Zentralstelle für Dokumentation LPh DOLISOS 1997.

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### Development of Biotechnological Protocols in Medicinal Plant Research – Contribution for Safeguarding and Standardization of Raw Material for Phytotherapy Using *Hypericum* Species Indigenous to the Balkan Region

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**Introduction:** Threats for medicinal plant species are unmonitored trade, over-exploitation, destructive harvesting techniques, as well as habitat loss and habitat changes. As a result, diminution of population sizes, genetic diversity and eventually the extinction of the species could occur.

**Aim:** The PhytoBalk project, a Bulgarian-Swiss Joint Research Project, strives for the development of standardized biotechnological protocols for the conservation of valuable medicinal plants indigenous to the Balkan region outside their natural habitats (*ex situ*), and on the other hand to provide for a platform for the production of medicinal plant raw material containing a standardized quality of secondary metabolites *in vitro*.

**Method:** Standard pharmacognostic methods combined with *in vitro* culturing techniques as well as secondary metabolite analysis based on Ph Eur HPLC and HPTLC methods.

**Results:** Intermediate results of biotechnological cultivation techniques on several not or less used *Hypericum* species and *Hypericum perforatum*

– St. John's Wort (treatment of mild depression) are presented in comparison to *ex situ* plant material. Comparison of the main secondary metabolites known from *Hypericum perforatum* show similar concentration levels of Hypericins in *in vitro* and *ex situ* biomass, whereas production of Flavonoids is rather suppressed in *in vitro* cultures.

**Conclusion:** Genotypes of the *Hypericum* genus, indigenous to the Balkan region, seem to have the potential for the development of a reliable *in vitro* cultivation system for biomass production rich in hypericine and pseudohypericine in laboratory scale experiments. However, special methodical and technological approaches are needed to optimize content of the polyphenolic and other constituents characteristic for this genus.

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QK12

### Bioautographic Xanthine Oxidase Assay: Combining Phytochemical Separation and Activity Assessment as a Tool for Medicinal Plant Research

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**Introduction:** Xanthine oxidase (XO) catalyses the oxidation of hypoxanthine and xanthine to uric acid under the formation of superoxide radicals and hydrogen peroxide. An overproduction of these reaction products in the human body is associated with diseases such as hyperuricemia, gout, hypertension, diabetes and different inflammatory diseases. XO inhibition assays are commonly used for drug discovery screening.

**Aim and Method:** Bioautography offers a rapid and simple tool for assessment of secondary metabolite profiles of medicinal plants by HPTLC combined with screening of potential health beneficial activities. A bioautographic XO Inhibition assay described by [1], has been optimized and validated in this study. A standard spectrophotometric assay format served for comparison of results to a standard method.

**Results:** For the establishment of a reliable, reproducible and validatable bioautographic XO assay, the concentrations of redox dye and substrate, enzyme activity, incubation time and assay temperature, as well as the buffer conditions were optimized using allopurinol as known inhibitory substance with a visual detection limit of 45.4 ng. Extracts of *Camellia sinensis* and *Artemisia alba* showed also to contain constituents with XO inhibitory activity, that could be visually detected as white spots on a purple coloured plate up to an applied amount of 10 µg dw for *C. sinensis* extract and 100 µg dw for *A. alba*.

**Conclusion:** The optimized bioautographic Xanthine Oxidase inhibition assay is a rapid and valid research tool for assessment of active secondary metabolites from medicinal plants.

#### Reference:

- 1 Ramallo I.A., Zaccaro S.A., Furlan R.L.E.: A Rapid TLC Autographic Method for the Detection of Xanthine Oxidase Inhibitors and Superoxide Scavengers. *Phytochemical Analysis* 2006;17:15-19.

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