

## ORIGINAL ARTICLE

**44<sup>th</sup> Conference drug synthesis and analysis – Part 1****Brno, 2<sup>th</sup> to 4<sup>th</sup> September 2015**

Dear colleagues, dear friends

It is pleasing to introduce the agenda of the “44<sup>th</sup> Conference on the Synthesis and Analysis of Drugs” with a wide range of highly interesting issues relating to the development of drugs.

Our Conference is a very important annual gathering of employees of pharmaceutical universities and research institutions it is a great opportunity to renew contacts and discuss topics of mutual interest.

I would like to thank all presenters and other participants who contributed to increasing the awareness of treatment possibilities. I firmly believe that we can meet the upcoming challenges – especially if we continue the tradition of science in the service of public health.

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organizational committee*

**Determination of biologically active compounds in the fungi of the genus *Cordyceps sinensis* by HPLC and NMR**

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**Introduction**

*Cordyceps sinensis* is the fungi parasiting larvae, pupae and imagoes of insect as well as fruiting bodies of truffles of the genus *Elaphomyces*<sup>1)</sup>. The fungi is known in both traditional Chinese medicine and in modern medicinal methods. It is used as a dietary supplement (*CORDYCEPS MRL*<sup>®</sup>, *ACAI DETOX*<sup>®</sup>). The fact of *Cordyceps sinensis* consequence is supported by many scientific studies, which have shown its positive effects, for example in anti-tumor therapy<sup>2)</sup>, in the treatment of HIV/AIDS, asthma, liver diseases and it also has a positive effect on female fertility etc.<sup>3)</sup>. Chemical compounds are responsible for these properties, which is currently characterized by the parasite. It was found that the fungi are rich in natural substances such as cordycepin, cordycepic acid, respectively, D-mannitol<sup>4–6)</sup>, polysaccharides<sup>7, 8)</sup>, nucleotides<sup>9)</sup>, proteins and amino acids<sup>10, 11)</sup>.

This paper is focused on the basic research of studied biologically active compounds (nucleosides, amino acids) identification. To date nucleosides are believed to be the active compounds in *Cordyceps*<sup>12)</sup>. Several methods

including thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC) and nuclear magnetic resonance spectroscopy (NMR) have been used for the identification of the compounds under study.

**Experimental methods**

The mushrooms for extraction were obtained dried and grinded from the Technical University of Zvolen, Faculty of Forestry, in co-operation with team of Ing. Martin Paulík, PhD.

For the identification of biologically active compounds occurring in *Cordyceps sinensis* we used methanolic extracts of eight fungi samples (**1–8**). For the separation of the content compounds thin layer chromatography was used (silica gel plates by Kiesselgel 60 F<sub>254</sub> from Merck) in chloroform. Detection of chromatographs was provided by UV light at 254 nm and by freshly prepared 2% ninhydrine in methanol. For HPLC detection on a UHPLC Ultimate 3000 (ThermoScientific) a DAD detector was used. The measurements were performed at the temperature of 25 °C on the column Polaris 5 C18-A 250 × 4,6 mm (Varian), the injection volumes were 20 µl, flow rate was 1 ml/min. The mobile phase consisted of 2% acetonitrile hypergrade for chromatography (Merck) in water for chromatography (Merck). Standards were purchased: cytidine and uridine (Sigma-Aldrich), guanosine and adenosine (Acros Organics), thymidine (ABCR) and inosine (Calbiochem). NMR spectra were measured on a Varian VNMRS 600 MHz in D<sub>2</sub>O (Merck).