Abstract

Curcumin with it's derivates belongs to pharmacologically acting diarylheptanoids. They are obtained from the herbal rhizomes of genus *Curcuma* spp. *(Zingiberaceae)*, which are widespread in tropical regions of south-east Asia. For last decades the drug has been in focus of scientists. On the human organism there have been proven many interesting effects. Therapeutically are used it's choleretic, antioxidant and antiinflammatory activities. Nowadays the interest orientates to the prevention and treatment of cancer and to the favourable influence on Alzheimer's disease. After oral administration, however, curcumin achieves the system circulation only in traces.

In introduced study we developed HPLC method for the determination of curcuminoids. The most advantageous way is extracting the pigments from the herb or extract with methanol as relatively polar solvent. In the aqueous medium is practically unsoluble. The presence of three acid hydrogens exhibits in the alkaline aqueous solutions. The resultating ionisation is documented by color change from yellow to red. These conditions prejudice the stability of the substance – in part we may prevent the degradation by using the antioxidants and keeping in dark place.

The analysis utilizes a reversed-phase C_{18} column and involves the use of an external standard. A mobile phase consisted of 45% acetonitrile and 55% phosphate buffer 0,025M, adjusted to pH 2,6. Sample detection was achieved by two different detectors. UV-VIS detection has been run at 422 nm. The fluorescence detection at λ_{ex} = 422 nm and λ_{em} = 525 nm, respectively. The achieved validations parameters confirm that method is precise sensitive and robust sufficiently. The linearity has been demonstrated on a wide range of concentrations. The benefit of UV-VIS detection is ability to quantify relatively high concentration of curcuminoids and the correlation of AUC on the amount of single curcuminoids. However, it accords lower order detection and quantification limit. Conversely the fluorescence detection is able to determine the curcuma pigments at level 10 ng/ml. The individual substances are different by their capability of emitting the fluorescence. Thus, the amount of single curcuminoids is possible to determine only when the sample and the standard are chemically equal in these conditions.

Potentially, the single-curcuminoid separation from the mixture (e.g. preparative columns or HP-TLC) and subsequent analysis of each substance alone, is the right solution. The obtained

fluorescence intensity data could allow the quantification of all three substances independently on the source.