

Nowadays, high performance liquid chromatography (HPLC) belongs among the most modern and most frequently used analytical separation techniques. Biocompatible iron chelators are mainly used for the treatment of iron overload. Nevertheless, in the light of new information about physiological and pathophysiological role of iron in organism, their potential use was widened into other diseases. Iron chelators were proved to have antioxidative, cardioprotective and cytostatic effects. Salicylaldehyde isonicotinoyl hydrazone is a biocompatible iron chelator, which also shows these activities. However, its short half life in plasma makes its use difficult. Nowadays, novel derivatives of SIH were prepared to improve its stability in plasma. 2'-hydroxypropiophenone isonicotinoyl hydrazone (HPP-INH) is one of newly prepared SIH derivatives.

The aim of this study was to develop optimal chromatographic conditions for HPLC analysis of HPP-INH and to develop sample pretreatment method using precipitation for its analysis in plasma.

The analysis was carried out on a Merck 250x4 mm column I.D. LiChrospher® 100, RP-18e (5 µm) protected with Purospher® 100, RP-18e (5 µm) guard column. The following mobile phase gradient condition was developed for HPP-INH purity evaluation: phosphate buffer (0,01 mol/l  $\text{NaH}_2\text{PO}_4 \cdot 2 \text{H}_2\text{O}$  water solution) with 2 mmol/l EDTA, pH 6 was adjusted by 0,1 mol/l NaOH water solution : methanol (A:B), 0:00 – 10:00 min B 35 – 70 % (v/v), flow 1,0 ml/min. UV detection was set to 252 and 288 nm. Following mobile phase isocratic condition was used for the analysis of HPP-INH in plasma: phosphate buffer (0,01 mol/l  $\text{NaH}_2\text{PO}_4 \cdot 2 \text{H}_2\text{O}$  water solution) with 2 mmol/l EDTA, pH 6 was adjusted by 0,1 mol/l NaOH water solution : acetonitrile, 59:41 (v/v), flow 1,1 ml/min. Precipitation using acetonitrile was developed as the sample pretreatment method. These extraction recoveries were achieved: HPP-INH 67 %, SIH 66 %, HPP 80 %. The stability experiment revealed that HPP-INH is much more stable in plasma than SIH, since 60 % of the initial amount stayed undegradated after 1 hour of exposition. Nowadays, antioxidative and cardioprotective effects of HPP-INH are tested at standardized *in vitro* models.