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Title of the Master's Thesis: Bioanalytical assessment of new prospective drugs derived from thiosemicarbazone I.

Abstract

Development of novel anti-cancer drugs which would boost current therapeutic regimes and/or overcome the resistance against common chemotherapy ranks among the most progressive fields of modern drug discovery. Selective targeting of intracellular iron (Fe) has been recognized as a new strategy for anti-cancer drug development. Biocompatible Fe chelators structurally derived from thiosemicarbazone belong to the most intensively studied anti-cancer agents. Apart from Triapine, currently in phase II of a clinical trial, the systematic investigation led to development of a new generation of iron chelators – di-2-pyridylketone thiosemicarbazones (DpTs). Based on the outcomes of numerous *in vitro* and *in vivo* experiments and di-2-pyridylketone-4,4-dimethyl-3-thiosemicarbazone (Dp44mT) was selected as the lead compound for further development.

The aim of this study was to develop an LC-MS method for the analysis of Dp44mT and its main phase I metabolites in biological materials (plasma, urine and faeces). The pilot *in vivo* experiment revealed that di-2-pyridylketone (DpK) and Dp4mT are the main metabolites of this drug. All compounds were separated on Discovery HS C18 column (75 × 4.6 mm, 3 µm, Sigma Aldrich) using the mixture of 2 mM ammonium formate and acetonitrile as the mobile phase. The structural analog of the parent drug di-2-pyridylketone-3-thio-semicarbazone (DpT) was synthesized and used as an internal standard. The plasma and urine samples were treated by solid phase extraction while a liquid-liquid extraction using solvents of various polarities was tested for isolation of the compounds from faeces. Linearity of the method for the determination of Dp44mT and its main metabolites in plasma was tested. LC-MS method developed in this study will be fully validated and employed for the analysis of samples from the pharmacokinetic experiment.