9 ABSTRACT

Charles University in Prague

Faculty of Pharmacy in Hradec Králové

Department of Pharmaceutical Chemistry and Drug Control

Candidate: Zuzana Kollárová

Supervisor: PharmDr. Petra Kovaříková

Title of diploma thesis: **Bioanalytical evaluation of new potential agents derived from thiosemicarbazone II.**

Nowadays, malignant neoplasms (cancer) are listed as the 4th most frequent cause of death. Therefore, its successful therapy and proper drug treatment attract scientific attention all over the world. New potent anticancer agent, 2-benzoylpiridine-4-ethyl-3-thiosemicarbazone (Bp4eT), developed by Prof. D. Richardson (University of Sydney, Australia) was identified as a leading compound of the 2-benzoylpiridine thiosemicarbazone series. These compounds, known as iron chelators, were originally developed to treat metabolic iron overload but recent investigations revealed their antiproliferative properties and highly selective mechanism of action in the therapy of malignant neoplasms.

In this study optimal conditions for HPLC-MS analysis of Bp4eT and its degradation products were developed. Also, di(2-pyridilketone)-4-phenyl-3-thiosemikarbazone (Dp4pT) was chosen as an internal standard.

Previous studies, during which Bp4eT was incubated with rat and human liver microsomal fractions, confirmed oxidative way of metabolism and revealed two phase I metabolites. HPLC-MS analysis detected that both of them resulted from the oxidation of thiocarbonyl group. First metabolite, identified as 2-benzoylpyridin-4-ethyl-3-semikarbazone, has the mass of 269 *m/z* and is present in

the form of two *E/Z* isomers, while the second, N^3-ethyl-N^1-[fenyl(pyridin-2-yl)methylen]-formamidrazne, has the mass of 253 *m/z*. Our further goal was to prepare chemical standards of these metabolites. Isolation of the chemical standards was performed by TLC. Their identification was confirmed using the NMR and IR spectra.

Finally, the reproductibility of the SPE extraction of Bp4eT and internal standard from plasma was confirmed.