ABSTRACT

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Title of diploma thesis: Study of protein-protein interaction of DHRS7 enzyme by pull-down assay

Dehydrogenase/reductase (SDR family) member 7 (DHRS7) is one of the less studied enzymes of SDR superfamily. It has been proven that this enzyme is *in vitro* involved in reductive metabolism of various compounds, such as steroids, retinoids and xenobiotics. Recently results pointing out to possible role of this enzyme in the pathogenesis of prostate cancer or other diseases has been published. It would be suitable to better characterize this enzyme to clarifying its patho/physiological role in the organism.

Because protein-protein interactions seem to be important for the function of proteins, the aim of this study was to identify interaction partners of the DHRS7, and thus contribute to the improvement of understanding of this enzyme. For our experiments, pull-down assay, *in vitro* method was utilized. The first step was immobilization of DHRS7 enzyme (bait protein) to suitable carrier (His Mag Sepharose Ni particles and nonmagnetic Protino Ni-IDA particles). Subsequently, the carrier with immobilized DHRS7 was incubated with the lysate of Hep G2 cells – the source of possible interacting proteins. The SDS-PAGE electrophoresis of sample after elution consist of several bands containing potential interacting partners of DHRS7. The MS analysis was used to identify these proteins.

The pull-down assay, *in vitro* method for study of protein-protein interaction, was implemented and optimized that could be used for further experiments at the Faculty of Pharmacy in Hradec Kralove.