

# ABSTRACT

Diploma Thesis

## **Proteomic characterization of host cell after interaction with intracellular pathogen *Francisella tularensis***

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*Francisella tularensis* (*F. tularensis*) is an intracellular gramnegative bacterium, causative agent of the serious human disease tularemia. It is a typical bacterium with unknown mechanisms of pathogenesis. A proteome characterization of host-pathogen interaction could significantly contribute to their elucidation. The aim of this diploma thesis was to reveal the protein expression changes in macrophage mouse cell line J774.2 after infection with *F. tularensis* using the quantitative „shotgun” proteome technology. The proteomes of host cells infected with *F. tularensis* for three various time intervals (6, 12 and 24 hours) were compared with the proteome of non-infected cells. Four subcellular fractions were prepared using ProteoExtract Subcellular Extraction Kit (S-PEK, Calbiochem). Fraction enriched for cytosolic proteins was further analyzed and quantified by iTRAQ-HPLC-MS/MS technology. This approach led to the successful identification of 248 proteins, the production of 37 proteins was then observed to be different compared to the non-infected cells. Twelve distinct proteins were found in the six hours infection period, eighteen distinct proteins in the twelve hours infection period and nineteen distinct proteins in the twenty four hours infection period. The applied technology has been demonstrated to be suitable for the proteome analysis of host-pathogen interaction and will be used in further studies for the confirmation and specification of the observed changes in proteome of host cell infected with *F. tularensis*.