Protein folding is a very complicated and error-prone process, which can be disrupted by multiple factors including genetic mutations, stress conditions, cell aging, or pathogen infections. As a result, cells are continuously producing misfolded proteins, with a vast majority appearing in the endoplasmic reticulum. Therefore, the cellular protein quality control eliminates misfolded proteins with the use of the ERAD (Endoplasmic Reticulum-Associated Degradation) pathway, where E3 ubiquitin ligases constitute key components. The ligases are responsible for the attachment of ubiquitin to the improperly folded proteins, which mediates their subsequent degradation. The diversity among E3 ubiquitin ligases in mammalian cells is enormous. More than 600 different human E3 enzymes have been identified so far, but only a small number of them have been fully characterized, while the function of most E3s still needs to be elucidated.

Viruses are masters of ERAD manipulation. One of the examples is bovine herpesvirus 1 (BHV-1, BoHV-1), which possesses a unique ability to induce antigenic transporter TAP degradation, which, in consequence, leads to the evasion of the $CD8^+$ cytotoxic T lymphocyte-mediated immune response. The viral protein responsible for this activity – UL49.5 – has been already identified; however, the exact mechanism of TAP degradation has remained unknown. Since UL49.5 has no E3 ubiquitin ligase-like domain, it has been speculated that this viral protein hijacks cellular degradation machinery, which leads to TAP degradation.

The aim of this thesis was the attempt to identify cellular factors, mainly ERAD components, utilized by UL49.5 to trigger TAP degradation.

To achieve this goal, I have constructed and validated a cellular fluorescent platform to study TAP degradation in the presence of UL49.5. This model was applied for high throughput siRNA screening with the use of the ubiquitin-proteasome system siRNA library. Analysis of the effect of knock-down of approximately 2000 cellular factors led to selection and validation of those, which (when silenced) exhibited increased stability of TAP complex in the presence of UL49.5. Obtained results demonstrate the role of the ubiquitin-proteasome system and retrotranslocation machinery in UL49.5-mediated TAP degradation, identify the key multicomponent E3 ligase being responsible, and explain the mechanism of UL49.5 action in targeting of TAP transporter to the proteasome pathway.