Student: Volodymyr Tsybulskyi
Supervisor: Mgr. Tomáš Fessl, Ph.D.
Co-supervisor: RNDr. Marek Scholz, Ph.D.
Opponent: Mgr. Filip Dyčka, Ph.D.
Title of the thesis: Comparison of fast single-molecule experiments and simulation of translocation through the SecY translocon.

Overall assessment:
Translocation of proteins across SecYEG channel in the inner membrane of Gramm-negative bacteria is a complex phenomenon, which includes several large proteins and it usually lasts for tens of seconds. Molecular dynamics (MD) simulation of such large system is on the edge of capability of current super-computers. To enable testing of current hypotheses, Volodymyr wrote a software package for Monte Carlo simulation of simplified one-dimensional Brownian motion through the channel. In Volodymyr's simplified model, the essential parameters affecting translocation are: diffusional force, friction in the channel, conformational entropy of substrate and trans-membrane potential. To link simulation to available experimental data, Volodymyr also implemented kinetics of opening and closing of the channel. These events are allosterically controlled by ATP hydrolysis in cytosolic SecA protein, which is thought to be the main driving unit of the molecular machine.
Apart from writing the simulation package, Volodymyr also helped to introduced novel correlative techniques for processing of MD simulations. These techniques should facilitate future design of our single-molecule fluorescence experiments, and thus help to test and develop new models of protein translocation.

Evaluation of student's performance:
Volodymyr is active and independent student. During this projects, he independently searched literature and implemented found biophysical phenomena into software. He also greatly improved his knowledge of diffusion, molecular machines, chemical kinetics and protein translocation.
We were finalising this project only few days before deadline for thesis submission, therefor there was not enough time to assemble sufficient amount of time-trajectories to get robust statistical analysis, which could be compared to experimental data. This wasn't Volodymyr's fault. I believe, it was caused mostly by my choice of ambitious project and limited time, cross-border students can dedicate to bachelor projects.

Future recommendations:
Volodymyr might benefit from improving his language skills and time management.

Conclusions:
In my opinion, Volodymyr's thesis fulfils all requirements posed on theses aimed for obtaining bachelor degree.

Proposed mark: very good.

České Budějovice, 9th May 2018

Tomáš Fessl

Mgr. Tomáš Fessl, Ph.D.