UNIVERSITÉ DE GENÈVE
FACULTÉ DES SCIENCES

LE DEPARTEMENT DE CHIMIE PHYSIQUE

a le plaisir de vous inviter à la

CONFERENCE

Intitulée

CAUGHT IN THE ACT : MOLECULAR DYNAMICS SIMULATIONS OF MEMBRANE TRANSPORTERS

donnée par

Prof. Dr. Lars SCHÄFER

RUHR UNIVERSITY BOCHUM (GERMANY)

le MARDI 5 MAI à 16h30

SALLE 1S081
Sciences III

30 quai Ernest-Ansermet ou 4 bld d'Yvoy

Responsable : Prof. Enrica BORDIGNON

Département de Chimie Physique, Sciences II, 30 quai E.-Ansermet 30 – 1211 Genève 4 / Tél 022 379 65 47

Caught in the Act: Molecular Dynamics Simulations of Membrane Transporters

Prof. Dr. Lars Schäfer

Center for Theoretical Chemistry, Ruhr University Bochum, D-44780 Bochum, Germany
lars.schaefer@rub.de

The talk is about recent molecular dynamics (MD) simulations of the functional mechanisms of bacterial transporters. Both an ABC transporter (TM287/288)[1-4] and a transporter from the major facilitator superfamily (MHAS2168)[4] will be covered.

ABC transporters couple ATP binding and hydrolysis in the nucleotide-binding domains (NBDs) to large-scale conformational changes of the transmembrane domains (TMDs), which ultimately lead to the translocation of substrate molecules across biological membranes. Despite the progress in the determination of high-resolution structures of substrate-bound ABC exporters, the inherently dynamic mechanisms of substrate transport remain unclear at the atomic level. Our all-atom MD simulations visualize how substrate molecules are translocated by the heterodimeric ABC exporter TM287/288 from *T. maritima*. Unguided multi-microsecond MD simulations show how the drugs daunorubicin and verapamil, which were initially docked into the inward-facing (IF) conformation of the transporter, move through the protein by following its large-scale alternating access transition from the initial IF conformer via an occluded intermediate to an outward-facing (OF) state. Substrate reuptake is prevented by marked differences between the IF and OF conformations concerning the hydration of the TMD inner cavity, which leads to an affinity difference between the two conformations and ultimately triggers the dissociation of the drugs from the OF conformation.

The second part of the talk is about the MFS transporter MHAS2168 from *M. hassiacum*, a homologue of Rv1410 from *M. tuberculosis*. Together with the periplasmic lipoprotein LprG, Rv1410 is involved in the transport of triacylglycerides (TAGs), which seal the mycomembrane. X-ray crystallography and cryo-EM structures of MHAS2168 show an OF conformation, with unusual extensions of TM helices 11 and 12 into the periplasm. Based on these experimental data, augmented by molecular docking and Alphafold modelling, a TAG-bound model of the MHAS2168-LprG complex was constructed, followed by extended coarse-grained MD simulations. The MD simulations show the spontaneous loading of TAG from a cavity in the transporter along TM helices 11 and 12 into the hydrophobic cavity of LprG, which is a key step in the functional transport mechanism.

Taken together, the combination of experimental and computational structural biology enabled us to propose functional mechanisms for both, the ABC exporter and the MFS transporter studied. These mechanistic working cycles will be discussed.

1. H. Göddeke, L. Schäfer. Capturing Substrate Translocation in an ABC Exporter at the Atomic Level. *J. Am. Chem. Soc.* 142, 12791 (2022). DOI 10.1021/jacs.0c05502
2. H. Göddeke, M. H. Timachi, C. A. J. Hutter, L. Galazzo, M. A. Seeger, M. Karttunen, E. Bordignon, L. Schäfer. Atomistic Mechanism of Large-Scale Conformational Transition

in a Heterodimeric ABC Exporter. *J. Am. Chem. Soc.* 140, 4543 (2020). DOI 10.1021/jacs.7b12944
3. C. A. J. Hutter, M. H. Timachi, L. M. Hürlimann, I. Zimmermann, P. Egloff, H. Göddeke, S. Kucher, S. Štefanić, M. Karttunen, L. Schäfer, E. Bordignon, M. A. Seeger. The Extracellular Gate Shapes the Energy Profile of an ABC Exporter. *Nature Commun.* 10, 2260 (2019). DOI 10.1038/s41467-019-09892-6
4. S. Remm, D. De Vecchis, J. Schöppe, C. A. J. Hutter, I. Gonda, M. Hohl, S. Newstead, L. Schäfer, M. A. Seeger. Structural Basis for Triacylglyceride Extraction from Mycobacterial Inner Membrane by MFS Transporter Rv1410. *Nature Commun.* 14, 6449 (2023). DOI 10.1101/2023.01.25.525346