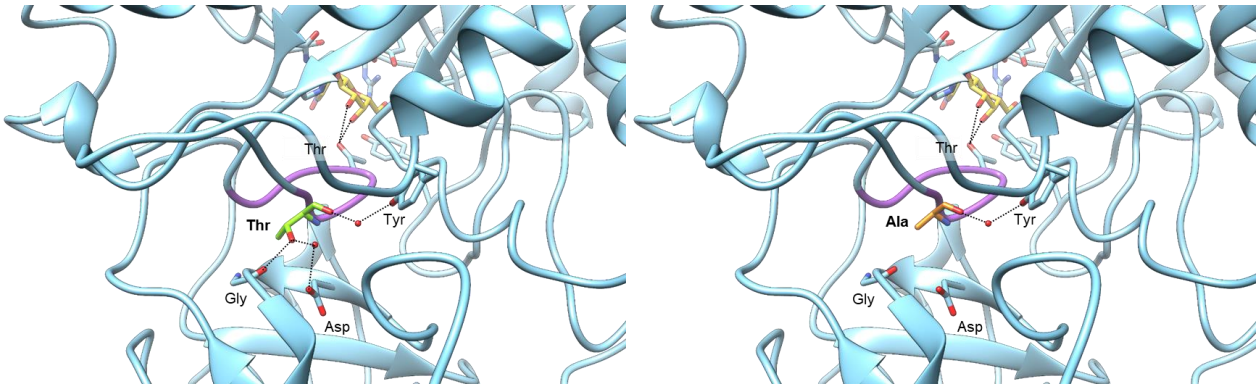
Molecular Case Study of a *GALC* Mutation Causing Infantile Krabbe Disease

Eileen Socher[1], Philipp Arnold[1], Friederike Zunke[2]

[1]Functional and Clinical Anatomy, Institute of Anatomy, Friedrich-Alexander-Universität Erlangen-Nürnberg (FAU), Erlangen, Germany;
[2]Department of Molecular Neurology, University Hospital Erlangen, Friedrich-Alexander-Universität Erlangen-Nürnberg (FAU), Erlangen, Germany

The Krabbe disease is a rare lysosomal disorder affecting the white matter of the central and peripheral nervous system. It is characterized by neurodegeneration and the severity depends on the age of onset. The most common form is the infantile Krabbe disease, which is usually diagnosed within the first year of life and has a high morbidity and mortality. The causes of this autosomal recessive disease are mutations in the *GALC* gene, which encodes the lysosomal enzyme galactocerebrosidase catalyzing the cleavage of galactose from galactocerebroside and galactosylsphingosine. This study presents a galactocerebrosidase variant found as homozygous mutation in the *GALC* gene of a little child with infantile Krabbe disease.



In order to investigate the effect of this mutation on the protein structure at the atomic level, at first, the homology model of the human galactocerebrosidase was build based on the crystal structure of its murine ortholog. In the second step, the structural stability of the mutated enzyme was analysed in several all-atom molecular dynamics (MD) simulations with protonation states corresponding to cytosolic pH (pH 7) and compared to the stability of the wild type enzyme under the same conditions. Furthermore, to account for the fact that the subcellular location of the galactocerebrosidase is the lysosome (pH 4.5-5.5), additional MD simulations were performed with protonation states corresponding to the acidic environment of the lysosome (pH 4.5). Differences in protein flexibility between the wild type and the mutated enzyme were only be observed at acidic pH and not at neutral pH. Similarly, effects of the mutation on the size of the binding pocket were observed at pH 4.5, although the mutation site itself is not part of the active site/binding site of the enzyme. Thus, these MD simulations provide insights into how this mutation affects the structure of the human galactocerebrosidase in the acidic environment of the lysosome.