Title: Improving antibody specificity towards heparan sulfate molecules for new therapeutic targets in Parkinson's disease.
Master research internship ~6 months, starting earliest February 2023

Hello everyone, my name is Thierry van Wessel, and I am currently looking for a student for a master research internship. I am a third year PhD candidate in the Matrix Biochemistry workgroup located at the 5th floor in the RIMLs building, https://www.radboudumc.nl/en/research/research-groups/matrix-biochemistry.

Parkinson's disease (PD) is the second most common brain disease. Without good biomarkers or drugs that target the cause of the disease, patients are left in despair for their future. Nowadays diagnosis is based on the presence of resting tremors, stiffness, and slow movement. Drugs are only available for reducing symptoms. This severe neurological disorder forms a drastic burden on our ageing society.

In PD patients, cells of the substantia nigra die. In this midbrain area, dopamine is produced. Loss of dopamine affects basal ganglia downstream and eventually causes pathology. To find new therapeutic strategies, we focus on the role of glycosaminoglycans, especially heparan sulfate (HS).

HS is a linear polysaccharide with high structural and functional diversity. This negatively charged molecule is present intracellularly, on cell membranes and in the extracellular matrix. During PD development, it is thought that modified HS molecules have a detrimental effect on the brain. Moreover, HS can interact with misfolded α-synuclein, preventing it being degraded by proteasomes. This results in the formation of α-synuclein fibrils and is toxic for neurons. Therefore, understanding HS motif related functioning might help to find a way in reversing factors that cause PD.

Differences in HS chains arise from several sugar modifications, like epimerization and sulfation. These modifications are not equally distributed across the chain, allowing sulfation motifs to form. It is challenging to study functions of specific HS motifs due to a lack of sequencing techniques, the enormous number of potentially different HS chains and large differences between each chain.

Our approach to study the function of HS in PD, is to use specific anti-HS single chain variable fragment (scFv) antibodies. In our lab, we have scFv antibodies that can recognize HS, but these do not identify a single motif. Detection and localization of specific HS motifs is relevant to reveal HS motifs related to diseases and provide a target for therapy.

The student project will focus on the production of more specific scFv antibodies against HS. 3D modelling of the interaction of the scFv antibody and HS molecules (figure 1) will be performed to visualize the binding in detail. Based on good docking results, antibody DNA will be mutated accordingly with a polymerase chain reaction (PCR). Isolated DNA will be sequenced to confirm mutagenesis. Mutated scFv antibodies will be produced by bacterial expression and tested in a western blot assay. The reactivity will be analyzed in enzyme-linked immune sorbent assays (ELISA) and immunofluorescence assays (IFA) and specificity in immunoprecipitation experiments (IP).

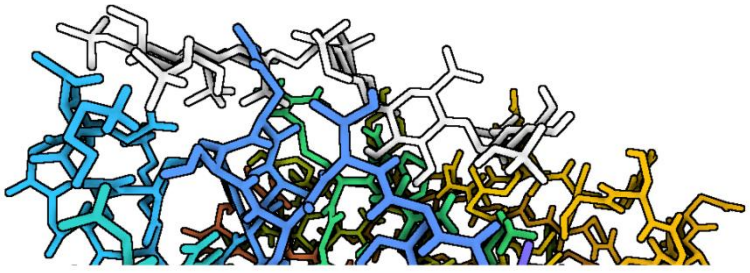

Figure 1. 3D model to visualize the interaction of heparan sulfate with a scFv antibody. White: heparan sulfate molecule, other colours: interacting surface of scFv antibody.

I am looking forward meeting a motivated student for this research internship. The duration will be ~6 months. If you are interested, please send a motivation letter with CV and intended starting date to Thierry.vanwessel@radboudumc.nl.