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Grant title: Deciphering the molecular role of sialylated glycans for the tropism of LASV

In the framework of the research funded by the Mizutani foundation we have investigated the role of sialylated glycan on the surface of LAMP1 during the cell entry process of the pathogenic Lassa virus. We aimed to reveal the biochemical role of sialylation for the interaction of LAMP1 with Lassa virus, understand how they interact and find out the functional role of this interaction.



The first part of our research was devoted for understanding the functional role of engaging with LAMP1 by the GP1 receptor-binding domain of Lassa virus during cell entry. GP1 binds to LAMP1 as part of a novel receptor switching activity that was recently revealed. Using pseudo-viruses in cell-based assays and based on structural information we discovered that the glycoprotein spike complex of Lassa virus senses acidic pH as it enters endocytic compartments by protonation of His230. This protonation arrests the spike and prevents undesired premature triggering of fusion. Furthermore, this protonation event is likely to prime the spike complex for interaction with LAMP1. For engaging with LAMP1, the GP1 of Lassa virus needs to have a positive charge at His230 implying that LAMP1 donates a negative charge for this interaction, possibly a negatively charged sialic acid. We further demonstrated that the binding to LAMP1 facilitates the triggering of the spike, likely by placing a negative charge near His230. Thus, Lassa virus enter cells, is primed and arrested by the acidic pH until it reaches a compartment with LAMP1 molecules that serve as the actual triggering cue.

The second aim of our Mizutani funded research was to reveal how GP1 of Lassa virus binds the glycosylated LAMP1, and what is the role of the sialylation for this interaction. The first effort was to map the LAMP1 binding site of the surface of GP1. For that, we crystalized and solved the structure of a GP1 domain from Morogoro virus. This virus is genetically close to Lassa but cannot bind LAMP1. This structural information allowed us to map the structural differences between the two proteins and using biochemical approaches we were able to tease out the differences that prevent LAMP1 binding. This comprehensive mapping provided us with a sequence signature that is needed for binding LAMP1. Knowing this sequence signature allowed us to conclude that switching to LAMP1 is unique to Lassa virus. Also, we were able to demonstrate that a fully mature and sialylated glycan on LAMP1 is critical for the interaction as LAMP1 that was produced in kifinensine-treated cells cannot bind GP1.