The Role of Glycosylation in the Bactericidal Pore Forming Protein Perforin-2

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Perforin-2 (P2) is a member of a family of immune proteins that employs its MACPF domain to form a β-barrel pore in bacterial membranes.

- The transition of the MACPF domain occurs around a rotational axis known as the hinge.
- The tip of the MACPF domain is glycosylated at residue N185 and is close proximity to the hinge.

Hypothesis: The N185 glycan prevents rotation of the MACPF domain at neutral pH.

Methods

- Transfect cells with WT or N185A P2 with Dox inducible system
- Validate Dox inducible expression of WT and N185A P2
- Test bactericidal capabilities of WT P2 vs N185A P2
- Compare stability of WT P2 vs N185A P2

Discussion

- Acidification of the phagosome may disrupt electrostatic interactions between the N185 glycan and the hinge allowing rotation to occur.
- Does P2 with the N185A mutation have the same functional capabilities as WT P2?

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