Interaction of insecticidal toxin proteins with target membranes

Supervisory team:
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Project description:

Insecticidal proteins are of increasing importance in agriculture to control pest species that damage crops and reduce yields. Many structurally-distinct families of proteins are able to lyse target insect cells in a selective manner, making them both safe in terms of human use/consumption and safe for non-target species, including beneficial insects. An in depth understanding of the mechanisms of action of these proteins is important for their development and to tackle problems that may arise due to insect resistance.

This project will study the biophysical and biochemical interactions of insecticidal pore-forming proteins with target membranes, using a combination of state-of-the art methods and novel microscopy techniques developed in-house. Recombinant proteins will be expressed and purified for analysis in test systems of increasing complexity from artificial lipid bilayers with controlled chemical composition to cell membranes. The formation of pores will be studied using state-of-the-art equipment (electrophysiology, fluorescence microscopy) to characterise pore properties and their dependence on membrane composition and physical conditions (eg temperature, pH).

At the interface between biochemistry and physics, cutting edge label-free optical microscopy techniques developed in-house will be used to analyse the detailed interaction of individual proteins with lipid membranes at the nanoscale with sub-millisecond time resolution, without introducing structural-functional artefacts.

This will enable us to answer key questions including i) how proteins remodel and diffuse within membranes in space and time, ii) how and where proteins partition, depending on the heterogeneous lipid membrane chemical composition and curvature, iii) how is the protein function modulated by the lipid environment and how is the lipid membrane local composition and curvature affected by the protein (an interplay often overlooked). Notably, bacteria, insects and mammals have very different cell membrane chemical compositions. This is likely to be important in regulating toxin specificity, an aspect not yet well studied that will be specifically explored in this project.

Protein-membrane interaction studies will be complemented by investigations on the structure and function of the pore forming toxins, that may include structural analysis (crystallography, electron microscopy, modelling) and mutagenic studies.

In this project, the successful candidate will join a vibrant cross-disciplinary team at the life science/physics interface and will learn a wide range of techniques likely to include: - Recombinant protein production and purification - Protein structural analysis - Production and characterisation of mutant proteins - Cell culture - Electrophysiological studies - Fluorescence microscopy - Differential Interference Contrast microscopy - Label-free optical microscopy based on interferometric gated off-axis reflectometry. Opportunities for wider training experiences and participation in conferences and seminars will also be promoted.