Submission Date: 05/07/2020

2019 Academic Year Bio-SPMs Collaborative Research
Research Report Summary

Title of the research project		Investigating the mechanism of membrane-thinning by antimicrobial peptides	
PI (Person in charge of the research project)	Name	Prof. Bart Hoogenboom	
	Affiliated Institution and Department/Division/etc.	University College London (London Centre for Nanotechnology and Department of Physics and Astronomy)	
	Position	Professor of Biophysics	
Bio-SPMs that you used (Check the boxes)		✖	Super-resolution AFM (FM-AFM/3D-AFM) – Kelvin probe
		☐	High-speed AFM
		☐	SICM (Kelvin probe)
Collaborative NanoLSI Faculty Members		Prof Takeshi Fukuma, Kaito Hirata	

Antimicrobial peptides (AMPs) are produced by nearly all organisms as defense against microbial pathogens. Many AMPs act by targeting the bacterial cytoplasmic membrane. In-liquid AFM of supported lipid bilayers (SLBs) is commonly used to resolve the mechanisms of these peptides. However, AFM is a surface technique that provides no chemical identification. Open-loop electric potential microscopy (OL-EPM) can provide simultaneous topography and electric potential maps of a surface. The aim of the research project was to determine whether we can use OL-EPM to better understand how AMPs attack the bacterial membrane. We developed a protocol for preparing and imaging fluid SLBs by OL-EPM. The system was stable and continuous topographic and surface potential maps could be obtained with nanoscale resolution. We then showed that changes to the topography and surface potential of the SLB can be measured by OL-EPM following addition of a cationic AMP. In our preliminary data, topographical images show that the peptide is solubilizing the lipid bilayer, and the remaining lipid has an extended boundary length characteristic of AMP-induced bilayer re-modelling. Our potential measurements show that the surface potential of the bilayer becomes more positive after peptide addition. We cannot resolve individual AMPs, but because the peptides are themselves cationic, this increase in surface charge indicates we are detecting the AMP diffusing across the lipid surface. Now that we have an optimized sample setup and have demonstrated that stable OL-EPM measurements can detect the changes in the surface potential of SLBs during AMP disruption, future experiments can be planned to validate our preliminary findings and to expand on them.

*This form (Form 3) will be open on the NanoLSI website in the following academic year.

*Note that this form should be prepared in one A4-size paper.

*Submission Deadline: May 8, 2020 (Friday). Submit it as a PDF file.

*Submission Destination: the person in charge of Bio-SPMs collaborative research at WPI-NanoLSI, Kanazawa University (Bio-spmscr_nano@ml.kanazawa-u.ac.jp)