

Contents

1	Introduction	1
1.1	Cell Membrane: Structure and Function	1
1.1.1	Function of the Cell Membrane	2
1.1.2	Composition of the Cell Membrane	2
1.1.3	Structure of the Cell Membrane	3
1.1.4	Phospholipid in Cell Membrane	4
1.2	Membrane Proteins and in Vitro Reconstitution	7
1.3	Membrane Binding Protein	10
1.4	NMR Methods to Study Membrane Protein	11
1.4.1	Background of NMR	11
1.4.2	Use NMR to Study Protein Structure	13
1.4.3	Using NMR to Study Protein Function	19
	References	25
2	Lipid Molecular-Ion Interaction Study Based on Nanodisc	29
2.1	What Is a Nanodisc?	29
2.2	Optimizing Nanodisc Assembly for NMR Studies	31
2.2.1	Experiment Methodology and Materials	31
2.2.2	Results	32
2.3	Lipid Molecular-Ion Interaction	35
2.3.1	Experiment Methods	36
2.3.2	Results	36
	References	41
3	Calcium Regulates T Cell Receptor Activation by Modulating Phospholipid Electrostatic Property	45
3.1	Introduction	46
3.2	Experiment Methods	47

3.2.1	Methods Summary	47
3.2.2	Cells and Reagents	47
3.2.3	CD3 ϵ _{CD} Peptide Expression and Labelling	48
3.2.4	Tyrosine Fluorescence Polarisation (FP) Assay	48
3.2.5	FRET Measurement.	48
3.2.6	T Cell Activation and Immunoblotting	49
3.2.7	Preparation of Nanodisc, Bicelles and Large Unilamellar Vesicles (LUVs).	49
3.2.8	NMR Experiments.	50
3.2.9	Aromatic NOESY	50
3.2.10	³¹ P Spectrum.	50
3.3	Experiment Result	50
	References.	69
4	Study on the Structure of Dengue Virus NS2B–NS3p in Complex with Aprotinin.	71
4.1	Experiment Methods.	71
4.1.1	Clone, Expression and Purification of NS2B–NS3p	71
4.1.2	NMR Sample Preparation	72
4.1.3	NMR Spectra Collection and Data Processing.	72
4.2	Experiment Results.	73
	References.	75
	Conclusion and Future Work.	77