

V. Table of Contents

I. Acknowledgements	2
II. List of publications and presentations resulting from this thesis	4
III. List of Abbreviations	6
IV. Summary.....	8
V. Table of Contents.....	11
1 Introduction	14
1.1 GAS Epidemiology.....	14
1.1.1 Different clinical manifestations.....	14
1.1.1.1 Superficial infections.....	15
1.1.1.2 Invasive infections	15
1.1.1.3 Immune-mediated conditions.....	16
1.1.2 Burden of disease	17
1.1.2.1 Superficial infections.....	17
1.1.2.2 Invasive infections	18
1.1.2.3 Immune-mediated conditions.....	19
1.2 GAS Immunology.....	20
1.2.1 Innate.....	20
1.2.1.1 Humoral innate immunity	20
1.2.1.2 Cellular innate immunity	24
1.2.2 Adaptive.....	25
1.3 GAS Bacteria and virulence factors	26
1.3.1 Group A Carbohydrate and Capsule.....	27

1.3.2	Secreted proteins.....	28
1.3.3	Surface expressed proteins	31
1.3.3.1	Mga regulon.....	31
1.3.3.2	M protein.....	32
1.3.3.2.1	M protein structure	33
1.3.3.2.2	A multifunctional molecule	34
1.3.3.2.3	An epidemiological marker.....	37
1.3.3.2.4	M type specific immunity.....	43
1.3.3.3	M-like proteins	44
1.3.3.4	Other Surface proteins.....	57
1.4	Vaccines	60
1.4.1	100 years of vaccine development.....	60
1.4.2	M protein based	61
1.4.3	Non-M protein based	64
2	Chapter 1: Investigation of immunity within <i>emm</i> -clusters	65
3	Chapter 2: Antibody response to the M protein J8 peptide during skin infection	76
4	Chapter 3: Development of a genomic repertoire for investigation of GAS vaccine candidates.....	85
5	Chapter 4: Majority of GAS encode a trio of M and M-like proteins.....	99
6	Chapter 5: Updated <i>emm</i> -typing PCR protocol to reduce misclassification of <i>emm</i> as <i>emm</i> -like genes	116
7	Chapter 6: Clusters of closely related Mrp and Enn proteins and their relationships to M-clusters.....	130

8 Chapter 7: Enn proteins bind to C4BP, an inhibitor of the complement system

149

8.1	Methods	149
8.1.1	Bacterial Strains and Culture conditions	149
8.1.2	Cloning of <i>enn</i> genes	150
8.1.3	Co-purification assays.....	151
8.1.4	C4BP deposition by Flow Cytometry.....	152
8.1.5	<i>In silico</i> modelling.....	153
8.2	Results	153
8.2.1	Development of C4BP-interaction motif.....	153
8.2.2	Selection of representative Enn proteins	155
8.2.3	C4BP bound to the surface of GAS in absence of M protein binding.....	157
8.2.4	Enn proteins bind C4BP	158
8.2.5	IgG effects C4BP binding by Enn.....	159
8.2.6	Enn proteins bind C4BP by the same pattern as M proteins.....	160
8.3	Discussion	163
9	Overall Discussion	165
9.1	Antibody responses to M proteins can be cross-reactive.....	165
9.2	GAS infection does not always lead to a strong anti-M protein response 167	
9.3	M-like proteins are carried by the majority of isolates	169
9.4	M-like proteins are closely related (but slightly different) to M proteins..	170
9.5	M-like proteins are functionally similar to M proteins.....	173
10	Conclusions and Future Perspectives.....	175
	References.....	178

