

# Preventing anaphylaxis to venom of the jack jumper ant (*Myrmecia pilosula*)

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**Dedication**

To my wife Danielle who has somehow managed to cope with two daughters under the age of three as well as my seemingly endless commitment to work and study, and who has kept me feeling loved, fed, and watered. To my daughters Stella and Felicity who's earliest memories will lead them to think that a laptop computer was somehow grafted to my body; their interruptions were always welcome and kept me more-or-less sane. Moreover, to our unborn child Michelle who has provided the inspiration to ensure this thesis is submitted before her birth so that I can return my whole attention to the family.

# Contents

**Summary** i

**Declaration** iii

**Acknowledgements** v

**Abbreviations** vii

**Foreword** ix

## **PART I: INTRODUCTION - 1**

### **Chapter 1: Literature review 3**

<i>1.1 Stinging insects of medical importance in Australia</i>	3
1.1.1 Phylogeny of the Aculeata	3
1.1.2 Family Apidae	4
1.1.3 Family Vespidae	6
1.1.4 Other Vespoidea families	7
1.1.5 Family Formicidae (ants)	8
<i>1.2 Insect sting anaphylaxis</i>	12
1.2.1 Clinical presentation	12
1.2.2 Pathophysiology	18
1.2.3 Management	29
1.2.4 Mortality	32
1.2.5 Pathogenesis	36
1.2.6 Prevalence & causative insects	38
1.2.7 Natural history & predictors of reaction severity	39
<i>1.3 Anaphylaxis classification and grading systems</i>	40
<i>1.4 Diagnosis of venom allergy</i>	45
1.4.1 Serum sIgE analysis	46
1.4.2 Venom skin testing	47
1.4.3 In-vitro tests of leukocyte reactivity to venom	49
1.4.4 Deliberate sting challenge	50
<i>1.5 Specific immunotherapy</i>	51
1.5.1 Evidence for clinical efficacy	52
1.5.2 Initiation phase	60
1.5.3 Safety, tolerability and compliance with immunotherapy	60
1.5.4 Antihistamine pre-medication	61
1.5.5 Maintenance dosing interval	62
1.5.6 Duration of therapy	62
1.5.7 Long term safety	64
1.5.8 Mechanisms	64
1.5.9 New technologies and adjuvants	73
<i>1.6 Myrmecia ant venoms</i>	74
1.6.1 Pharmacological properties	74
1.6.2 Allergenic components	74
<i>1.7 Literature review summary</i>	77

### **Chapter 2: Research objectives and outline 79**

<i>2.1 Objectives</i>	79
<i>2.2 Research outline</i>	79
<i>2.3 Thesis structure</i>	80

## **PART II: EPIDEMIOLOGY OF JACK JUMPER ANT STING ALLERGY IN TASMANIA- 81**

### **Chapter 3: Deaths 83**

- 3.1 *Introduction* 83
- 3.2 *Methods* 83
- 3.3 *Results* 84
- 3.4 *Discussion* 88
  - 3.4.1 Principal findings 88
  - 3.4.2 Study strengths & weaknesses 88
  - 3.4.3 Comparison with related studies 88
  - 3.4.4 Interpretation 90
  - 3.4.5 Unanswered questions 91

### **Chapter 4: Population prevalence and emergency department presentations 93**

- 4.1 *Introduction* 93
- 4.2 *Methods* 93
  - 4.2.1 Emergency Department presentations 93
  - 4.2.2 Population sting exposure and allergy prevalence 94
  - 4.2.3 Statistical analysis 94
- 4.3 *Results* 95
  - 4.3.1 Population sting exposure and allergy prevalence 95
  - 4.3.2 Emergency Department presentations 96
- 4.4 *Discussion* 97
  - 4.4.1 Principal findings 97
  - 4.4.2 Study strengths & weaknesses 97
  - 4.4.3 Comparison with related studies 97
  - 4.4.4 Interpretation 98
  - 4.4.5 Unanswered questions 99

### **Chapter 5: Initial assessment and prospective follow-up of JJA sting allergic volunteers 101**

- 5.1 *Introduction* 101
- 5.2 *Methods* 101
  - 5.2.1 Allergic volunteers; clinical features and serum IgE analysis 101
  - 5.2.2 Accidental field stings 102
  - 5.2.3 Statistical analyses 102
- 5.3 *Results* 102
  - 5.3.1 Baseline clinical characteristics 102
  - 5.3.2 Prior reaction severity; correlation with other baseline clinical characteristics 103
  - 5.3.3 Serum JJA venom-specific IgE 103
  - 5.3.4 Serum venom-specific IgE to other Myrmecia, bee and wasp venoms 105
  - 5.3.5 Accidental field stings 106
- 5.4 *Discussion* 109
  - 5.4.1 Principal findings 109
  - 5.4.2 Study strengths & weaknesses 109
  - 5.4.3 Comparison with related studies 110
  - 5.4.4 Interpretation 111
  - 5.4.5 Unanswered questions 112

## **PART III: VENOM ANAPHYLAXIS AND IMMUNOTHERAPY - 113**

### **Chapter 6: Introduction 115**

- 6.1 *Clinical trial of venom immunotherapy* 115
- 6.2 *Ancillary studies* 115
  - 6.2.1 Sting anaphylaxis; clinical observations, management and diagnosis 115
  - 6.2.2 Predicting sting challenge reaction risk 116
  - 6.2.3 Changes in laboratory parameters with VIT 117
- 6.3 *Ethical issues* 117
  - 6.3.1 Use of a placebo group 117
  - 6.3.2 Determining reaction severity 118
  - 6.3.3 Double blinding 118
- 6.4 *Ethics approval* 118

### **Chapter 7: Methods 119**

- 7.1 *Trial participants* 119
- 7.2 *Venom extracts* 119
- 7.3 *Venom skin testing* 120
- 7.4 *Immunotherapy* 120
  - 7.4.1 Placebo-controlled randomised phase 120
  - 7.4.2 Crossover phase 122
- 7.5 *Sting challenge procedure* 122
  - 7.5.1 Clinical 122
  - 7.5.2 Sting challenge blood sampling and laboratory analyses 124
- 7.6 *In vitro laboratory studies* 124
  - 7.6.1 Blood sampling 125
  - 7.6.2 RAST 125
  - 7.6.3 Basophil activation tests (BAT) 125
  - 7.6.4 Histamine release tests (HRT) 125
  - 7.6.5 Leukotriene release tests (LRT) 125
  - 7.6.6 Venom concentrations used for BAT, HRT and LRT 126
  - 7.6.7 Venom induced lymphocyte proliferation and cytokine excretion 126
- 7.7 *Analysis and power studies* 127
  - 7.7.1 Clinical immunotherapy trial 127
  - 7.7.2 Sting anaphylaxis; clinical observations, management and diagnosis 127
  - 7.7.3 Predicting sting challenge reactivity in the placebo group 128
  - 7.7.4 In vitro parameter changes during VIT 129
- 7.8 *Role of the funding sources* 129

### **Chapter 8: Clinical trial of venom immunotherapy 131**

- 8.1 *Results* 131
- 8.2 *Discussion* 136
  - 8.2.1 Principal findings 136
  - 8.2.2 Study strengths & weaknesses 137
  - 8.2.3 Comparison with related studies 137
  - 8.2.4 Interpretation 139
  - 8.2.5 Unanswered questions 139

### **Chapter 9: Sting anaphylaxis; manifestations, management and diagnosis 141**

- 9.1 *Results* 141
  - 9.1.1 Clinical description of reactions in the placebo group 141
  - 9.1.2 Serum tryptase and plasma histamine 144
- 9.2 *Discussion* 148
  - 9.2.1 Principal findings 148
  - 9.2.2 Study strengths & weaknesses 149

9.2.3	Comparison with related studies	150	
9.2.4	Interpretation	152	
9.2.5	Unanswered questions	153	
<b>Chapter 10:</b>	<b>Predicting sting challenge reaction risk</b>	<b>155</b>	
10.1	<i>Results</i>	155	
10.1.1	Correlations between laboratory parameters and skin testing		155
10.1.2	Predicting sting challenge reactions in the placebo group		155
10.2	<i>Discussion</i>	158	
10.2.1	Principal findings	158	
10.2.2	Study strengths & weaknesses	158	
10.2.3	Comparison with related studies	159	
10.2.4	Interpretation	160	
10.2.5	Unanswered questions	160	
<b>Chapter 11:</b>	<b>Changes in laboratory parameters with VIT</b>	<b>161</b>	
11.1	<i>Results</i>	161	
11.2	<i>Discussion</i>	166	
11.2.1	Principal findings	166	
11.2.2	Study strengths & weaknesses	166	
11.2.3	Comparison with related studies	167	
11.2.4	Interpretation	168	
11.2.5	Unanswered questions	168	
<b>Chapter 12:</b>	<b>Conclusion</b>	<b>169</b>	
12.1	<i>Major findings</i>	169	
12.2	<i>Interpretation</i>	171	

**Appendix: Published papers- 173**

**Bibliography- 175**

## Summary

**Background** *Myrmecia pilosula* (the “jack jumper” ant, JJA) is the principal cause of ant venom anaphylaxis in Australia. Whereas honeybee and wasp venom allergy can be treated by venom immunotherapy (VIT), no such treatment is available for ant sting allergy. In addition, information on the natural history of JJA sting allergy is required to identify those most likely to benefit from immunotherapy. The main objectives of this research were to establish: (i) the prevalence, natural history and determinants of reaction severity for JJA allergy, and; (ii) the efficacy and tolerability of JJA VIT.

**Methods** A search of the Royal Hobart Hospital (RHH) forensic register, a random telephone survey, and a review of emergency department (ED) presentations were performed. Three hundred eighty-eight JJA allergic volunteers were assessed, including serum venom-specific IgE RAST, and then followed up for accidental stings over a 4-year period. Finally, a randomised double-blind, placebo-controlled, crossover trial of JJA VIT was performed. Laboratory parameters measured during the trial were; leukocyte stimulation index (SI), IL-4 production, IgE RAST, histamine release test (HRT), leukotriene release test (LRT) and basophil activation test (BAT). Intradermal venom skin testing (VST) was also performed at trial entry.

**Findings** The prevalence of JJA sting allergy was 2.7% in the Tasmanian population, compared to 1.4% for honeybee. People aged  $\geq 35$  had a greater risk of both sting allergy and hypotensive reactions. Four deaths were identified, all in adults with significant comorbidities. During follow-up, 79 (70%) of 113 accidental jack jumper stings caused systemic reactions. Only prior worst reaction severity predicted the severity of follow-up reactions, with the majority of people experiencing similar or less severe reactions when stung again.

Sixty-eight otherwise healthy JJA allergic adult volunteers were enrolled in the clinical trial. Systemic reactions to therapy were recorded in 34% during VIT. Objectively defined systemic reactions to sting challenges arose in 1/35 after VIT (mild self-limiting urticaria only) versus 21/29 in the placebo group. Treatment with oxygen, intravenous adrenaline infusion and volume resuscitation was effective and well tolerated. Hypotension was always accompanied by a relative bradycardia, which was severe and treated with atropine in two patients.

In the placebo group, only VST and HRT were predictive of sting challenge results. Although IgE RAST, leukocyte SI and IL-4 production, LRT and BAT all correlated well with VST, they did not predict sting challenge outcome. After successful VIT,

venom-induced leukocyte IL-4 production tended to fall, whereas IgE RAST increased and a natural decline in HRT reactivity was reversed.

**Interpretation** VIT is highly effective in prevention of JJA sting anaphylaxis and is likely to be of most benefit to people with a history of severe systemic reactions, which usually occur in people aged over 35. Neurocardiogenic mechanisms &/or direct cardiac effects may be important factors in some anaphylaxis deaths. Systemic reactions to immunotherapy are common and require immediate access to resuscitation facilities. The HRT warrants further investigation as a test for selecting those most likely to benefit from VIT. None of the tests evaluated appear to be reliable markers of successful VIT.

## Declaration

I certify that this thesis does not incorporate without acknowledgement any material previously submitted for a degree of diploma with any university; and that to the best of my knowledge and belief does not contain any material written by another person except where due reference is made in the text.

Simon G. A. Brown, 15 October 2003



## Acknowledgements

The development of an effective venom immunotherapy for people allergic to the sting of the “jack jumper” began in Tasmania in the 1980’s thanks largely to the late Dr Paul Clarke who brought the extent of the problem to the attention of doctors, media and local politicians. Dr Clarke initiated the process leading to this trial by involving the Royal Hobart Hospital Department of Emergency Medicine through my predecessor, Dr Bryan Walpole.

A milestone for our team was establishing a link with the Flinders Medical Centre. My decision to pursue PhD studies with Flinders University was determined by early interactions with Dr Bob Heddle. It was immediately apparent that his enthusiastic supervision would make a world of difference. Our research has benefited enormously from Bob’s keen clinical and analytical skills, unparalleled experience of venom immunotherapy for bee sting allergy, generous provision of time, and not least of all his Australian Rules football-oriented humour. I am also grateful for the academic guidance provided by Professor Peter Roberts-Thomson.

The Royal Hobart Hospital Research Foundation, Department of Health and Human Services (via the Dick Butfield Memorial Scholarship) and Cosy Cabins Tasmania (in memory of Mr Arthur F. Park who died following a jack jumper sting in 1999) all provided generous financial support. NSL Health Ltd (Melbourne), at the instigation of Dr Brian Baldo, then provided a substantial funding boost enabling us to expand the scope of our project. I am also indebted to many staff in the Emergency Departments and Pathology Departments of the Royal Hobart Hospital and North West Regional Hospital Burnie, for their assistance during all stages of our work.

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Numerous co-investigators, students and research assistants helped during various stages of the project. These are listed as authors &/or acknowledgments in our published papers included in the Appendices. In particular I thank pharmacist Michael Wiese without whom the clinical trial could not have occurred, Dr Konrad Blackman, Vikki Stenlake (Research Nurse), Sandra Ahokas (Administrative Assistant), Matilda

Haas (Research Assistant and Science Honours Student), Andrew Black and Anand Parameswaran (Medical Honours Students), and David Spiers (Research Assistant).

Medical research requires strong collaborations. Therefore, this thesis necessarily includes some data that has been described in theses of my collaborators. Immunological studies reported in brief for one case from our series of deaths (Chapter 3) also forms part of the PhD thesis of Qi Xuan Wu (University of Sydney). Three honours students performed a wide range of laboratory studies under my supervision for the project (Matilda Haas- cellular proliferation and cytokine production studies; Andrew Black- histamine/leukotriene release and basophil activation studies; Anand Parameswaran- serum IgE analysis). Their theses report detailed laboratory methods and preliminary data. However, the analysis of all results (including clinical correlation) is reported here for the first time (Chapters 10 and 11).

The 68 Tasmanians who enrolled in the clinical trial deserve the highest praise. Without them, all our endeavours would have been in vain. Each had the choice of waiting until a proven treatment was available thus leaving it for others to take the risk. To each of them I extend my thanks on behalf of at least 10,000 Australians who stand to benefit immediately and many more who will benefit in the future.

Finally, Dr Bryan Walpole will not be forgotten. His vision and moral support have been inspirational throughout my career at the Royal Hobart Hospital, and his masterstroke can be summed up in his own words: “Mate, would you mind taking on this research for me after I leave? It’ll just be a matter of giving some injections to a few people, a couple of hours each week for a few months...”

## Abbreviations

BAT	Basophil activation test
ED	Emergency department
HRT	Histamine release test
IFA	Imported fire ant, <i>solenopsis</i> spp.
IFN- $\gamma$	Interferon gamma
IL-4	Interleukin 4
JJA	Jack jumper ant, <i>Myrmecia pilosula</i> species complex
LRT	Leukotriene release test
<i>Myr p 1</i>	<i>Myrmecia pilosula</i> venom peptide allergen 1
<i>Myr p 2</i>	<i>Myrmecia pilosula</i> venom peptide allergen 2
RAST	Radioallergosorbent test
slgE	Allergen-specific Immunoglobulin E
slgG	Allergen-specific Immunoglobulin G
SI	Stimulation index
SIT	Specific immunotherapy (aeroallergen and venom)
sp., spp.	Species (sp.- singular, spp.- plural)
VIT	Venom immunotherapy
VST	Intradermal venom skin testing
WBE	Whole body extract



## Foreword

In 1964, an analysis of 5-years of enquiries to Commonwealth Serum Laboratories for medical advice found that allergy to the sting of *Myrmecia pilosula* (the jack jumper ant, JJA) was a problem mainly in Tasmania and Victoria.<sup>1</sup> The potential extent and severity of this problem was outlined in 1986 by Dr Paul Clarke, who also questioned the contemporary practice of desensitisation using ant whole body extracts (WBE) and suggested further research into the use of pure venom (venom immunotherapy, VIT) to treat ant venom allergy.<sup>2</sup> *Myrmecia* WBE preparations were withdrawn in the early 1990s and at a scientific meeting in 1995 were reported to be ineffective.<sup>3</sup>

This thesis describes clinical research conducted in Tasmania to develop a venom immunotherapy for JJA sting allergy. Behind the scenes, development of a method for extracting large amounts of JJA venom in the field, further investigation of the native venom allergens, development of improved analytical techniques, and venom stability studies have been underway. That work has been expertly managed by my pharmacist colleague Michael Wiese, and will be reported elsewhere.

