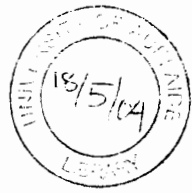


09 PH
0 151



**Studies on the systematics of the cestodes infecting
the emu, *Dromaius novaehollandiae* (Latham, 1790)**

Michael O'Callaghan

Department of Environmental Biology

School of Earth and Environmental Sciences

The University of Adelaide

TABLE OF CONTENTS

	Page
ABSTRACT	i-ii
Declaration	iii
Acknowledgements	iv-v
Publication arising from this thesis (see Appendices H, I, J).	
Chapter 1. INTRODUCTION	
1.1 General introduction	1
1.2 The host, <i>Dromaius novaehollandiae</i> (Latham, 1790)	2
1.3 Cestode nomenclature	3
1.3.1 Characteristics of the family Davaineidae	4
1.3.2 <i>Raillietina</i> Fuhrmann, 1909	5
1.3.3 <i>Cotugnia</i> Diamare, 1893	7
1.4 Cestodes of emus	8
1.5 Cestodes from other ratites	8
1.6 Records of cestodes from emus in Australia	10
Chapter 2. GENERAL MATERIALS AND METHODS	
2.1 Cestodes	11
2.2 Location of emu farms	11
2.3 Collection of wild emus	11
2.4 Location of abattoirs	12
2.5 Details of abattoir collections	12
2.6 Drawings and measurements	13

2.7	Effects of mounting medium	13
2.8	Terminology	13
2.9	Statistical analysis	14

Chapter 3. TAXONOMY OF THE CESTODES INFECTING STRUTHIONIFORMES IN AUSTRALIA

3.1	Introduction	15
3.2	Material examined	
3.2.1	Australian Helminth Collection	16
3.2.2	Parasitology Laboratory Collection, South Australian Research and Development Institute	17
3.2.3	Material collected at abattoirs from farmed emus	17
3.3	Preparation of cestodes	
3.3.1	Staining of cestodes	18
3.3.2	Measurements	19
3.3.3	Terminology	20
3.4	Results	
3.4.1	Species descriptions	
3.4.1.1	<i>Raillietina australis</i> (Krabbe, 1869)	20
3.4.1.2	<i>Raillietina beveridgei</i> O'Callaghan, Davies and Andrews, 2000	25
3.4.1.3	<i>Raillietina chiltoni</i> O'Callaghan, Davies and Andrews, 2000	30
3.4.1.4	<i>Raillietina dromaius</i> O'Callaghan, Davies and Andrews, 2000	34
3.4.1.5	<i>Raillietina mitchelli</i> O'Callaghan, Davies and Andrews, 2000	38
3.5	Comparison with other species	40

3.6	Closely related species in the Casuariidae	
3.6.1	Species descriptions	
3.6.1.1	<i>Raillietina geraldshmidti</i> O'Callaghan, Andrews, Davies and Spratt, 2001.	45
3.6.1.2	<i>Raillietina casuarii</i> (Kotlan, 1923)	48
3.6.1.3	<i>Raillietina infrequens</i> (Kotlan, 1923)	52
3.7	Cestodes in Struthionidae	
3.7.1	Species descriptions	
3.7.1.1	<i>Raillietina australis</i> (Krabbe, 1869)	57
3.8	Calcareous corpuscles	61
3.9	Identification of archived material	61
3.10	Summary of cestode identifications and prevalence of species	61
3.11	Discussion	65
3.12	Summary	72

Chapter 4. HOST-PARASITE RELATIONSHIP BETWEEN *RAILLIETINA* SPECIES AND THE EMU

4.1	Introduction	73
4.2	Materials and methods	
4.2.1	Enumeration and distribution of cestode species	76
4.2.2	Distribution of cestode species in relation to <i>diverticulum caecum vitelli</i> , 'Meckel's diverticulum'	77
4.2.3	Histological examination	77
4.2.4	The crowding effect	77
4.2.5	Biochemical analysis	78
4.2.6	Statistical analysis	79

4.3	Results	
4.3.1	Length of intestine	79
4.3.2	Intensity of cestodes	79
4.3.3	Distribution of cestode species in the small intestine of emus	80
4.3.4	Statistical analysis	90
4.3.5	Distribution of cestode species in relation to Meckel's diverticulum	94
4.3.6	De-strobilised cestodes	94
4.3.7	Histopathology of the small intestine	95
4.3.8	Effect of crowding	
4.3.8.1	Size of cestodes measured by weight	96
4.3.8.2	Size of cestodes measured by scolex diameter	97
4.3.8.3	Number of egg capsules per proglottis	97
4.3.8.4	Number of eggs per capsule	98
4.3.9	Biochemical analysis	98
4.4	Discussion	98
4.5	Summary	107

Chapter 5. THE LIFE CYCLE OF *RAILLIETINA* SPECIES INFECTING THE EMU

5.1	Introduction	109
5.2	Materials and methods	
5.2.1	Collection of organisms	110
5.2.2	Organisms examined	110
5.2.3	Identification of ants and beetles	110
5.2.4	Collection of cysticercoids	112
5.2.5	Statistical analysis	112

5.3	Results	
5.3.1	Intermediate host	113
5.3.2	Description of cysticercoids	
5.3.2.1	<i>Raillietina australis</i> (Krabbe, 1869)	114
5.3.2.2	<i>Raillietina beveridgei</i> O'Callaghan, Davies and Andrews, 2000	115
5.3.2.3	<i>Raillietina chiltoni</i> O'Callaghan, Davies and Andrews, 2000	115
5.3.2.4	<i>Raillietina dromaius</i> O'Callaghan, Davies and Andrews, 2000	116
5.3.2.5	<i>Raillietina mitchelli</i> O'Callaghan, Davies and Andrews, 2000	117
5.3.3	Number of cysticercoids recovered	122
5.3.4	Appearance of the cysticercoid wall	122
5.3.5	Size of cysticercoids	124
5.4	Discussion	129
5.5	Summary	134

Chapter 6. SOME ASPECTS OF THE FINE STRUCTURE OF RAILLIETINA SPECIES

6.1	Introduction	136
6.2	Material and methods	
6.2.1	Histological examination	139
6.2.2	Scanning Electron Microscopy	139
6.2.3	Transmission Electron Microscopy	140
6.2.4	Measurements	141
6.2.5	Specimens examined	141
6.2.6	Release of eggs from capsules	141
6.3	Results	

6.3.1	Histological features of the scolex	
6.3.1.1	<i>Raillietina australis</i>	142
6.3.1.2	<i>Raillietina beveridgei</i>	143
6.3.1.3	<i>Raillietina chiltoni</i>	145
6.3.1.4	<i>Raillietina dromaius</i>	146
6.3.1.5	<i>Raillietina mitchelli</i>	149
6.3.2	Scanning Electron Microscopy of the scoleces of <i>Raillietina. dromaius</i> and <i>Raillietina. beveridgei</i>	
6.3.2.1	<i>Raillietina dromaius</i>	150
6.3.2.2	<i>Raillietina beveridgei</i>	151
6.3.3	Transmission Electron Microscopy	
6.3.3.1	The scolex of <i>Raillietina dromaius</i>	152
6.3.3.2	The sucker of <i>Raillietina australis</i>	155
6.3.3.3	The scolex of <i>Raillietina mitchelli</i>	156
6.3.4	Fine structure of the egg capsule of <i>Raillietina beveridgei</i>	159
6.3.5	Observation of the embryophore in released eggs	164
6.4	Discussion	164
6.5	Summary	171

Chapter 7. GENETIC ANALYSIS OF RAILLIETINA SPECIES 1 FROM EMUS

7.1	Introduction	172
7.1.1	Molecular analysis using nucleotide sequence data	173
7.2	Materials and methods	
7.2.1	DNA based characterisation	174
7.3	Results	
7.3.1	18S rDNA	177

7.3.2	ITS2 rDNA	187
7.3.3	CO1 mtDNA	192
7.4	Discussion	196
7.5	Summary	198
Chapter 8. GENERAL DISCUSSION		199
Appendix A	Updated list of the species of <i>Fuhrmannetta</i> <i>Raillietina</i> , <i>Paroniella</i> and <i>Skrjabinia</i> .	205
Appendix B	Comparison of mean length of rostellar hooks in Lactophenol and De Faurés medium	208
Appendix C	Size of rostellar hooks	209
Appendix D	Statistical representation of rostellar hook length	210
Appendix E	Intensity of helminths other than cestodes	213
Appendix F	Biochemical analysis of emu liver	214
Appendix G	Biochemical analysis of emu plasma	215
 Publications arising from this thesis		
Appendix H	O'Callaghan, M.G., Davies, M. and Andrews, R.H. (2000). <i>Transactions of the Royal Society of South Australia</i> . 124 : 105-116.	216
Appendix I	O'Callaghan, M.G., Andrews, R.H., Davies, M. and Spratt, D.M. (2001). <i>Transactions of the Royal Society</i> <i>of South Australia</i> . 125 : 133-139.	217
Appendix J	O'Callaghan, M.G., Davies, M. and Andrews, R.H. (2003). <i>Systematic Parasitology</i> . 55 : 19-24.	218
BIBLIOGRAPHY		219

ABSTRACT

Four new species of cestode are described from the emu, *Dromaius novaehollandiae* Latham, 1790 and one from the southern cassowary, *Casuarius casuarius* Linnaeus, 1758. All are assigned to the genus *Raillietina* Fuhrmann, 1920 (*sensu lato* Jones and Bray, 1994) on the basis of the possession of two rows of numerous hammer-shaped rostellar hooks, unilateral genital pores, a small cirrus sac which does not reach or just crosses the osmoregulatory canals and egg capsules containing several eggs. In addition, *R. australis* (Krabbe, 1869) from the emu and *R. casuarii* (Kotlan, 1923) from the cassowary are redescribed from Australian specimens. These specimens are compared to and distinguished from all known congeners in the Struthioniformes. The principle diagnostic characters are identified and tabled. This study incorporates a range of analyses to establish the identity of the cestodes infecting *Dromaius novaehollandiae* in Australia.

Large numbers of cestodes were recovered from farmed and wild emus with a maximum intensity of 4795 in a wild bird. *Raillietina beveridgei* was the most predominant species encountered.

The distribution of the cestode species infecting emus indicated that each species occupied a preferred and predictable portion of the intestinal tract that is believed to reinforce reproductive advantage.

Cysticercoids of the five species of *Raillietina*, *R. australis*, *R. beveridgei*, *R. chiltoni*, *R. dromaius* and *R. michelli*, were recovered from ants belonging to the genus *Pheidole* and are described. Each species was identified on the basis of the number and size of rostellar hooks which corresponds to that of adult worms. There was a trend towards an inverse relationship between size of the cysticercoid and the parasite burden in the intermediate host.

Ultrastructural studies showed that the microtriches present on scoleces do not differ from those reported from other davaineids. Examination of the fine structure of the egg capsule revealed a thin and greatly-folded embryophore. The mechanism of egg hatching appears to begin with mechanical release of the embryo from the capsule followed by expansion of the folded embryophore to encircle the oncosphere.

DNA sequencing techniques were applied to confirm the morphological distinction of adult worms. 18S, ITS2 and CO1 gene sequence analysis provided additional characters to enable species separation.