

**Parasites of sleepy lizards
(*Tiliqua rugosa*) sampled
across an ecological gradient
at Bunday Bore Station,
South Australia**

by

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M.Sc.

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Abstract

Parasites make up a large part of the world's biodiversity and are important components of ecosystems because they influence the fitness of their hosts, and therefore intraspecific and interspecific interactions. Historically, parasites have often been overlooked in ecological studies, so there are gaps in our understanding of their natural history, and even fundamental information, such as their geographic distribution, is often poorly understood. The distribution of parasites is not consistent throughout their distribution range. Variations in limiting factors, such as precipitation, in an area can influence plant communities in such a way that it creates ecological gradients. Species that can inhabit a variety of habitat types often occur throughout such an ecological gradient. However, the parasites that infect them may differ over the ecological gradient due to being more habitat specific or requiring additional hosts that are habitat specific.

In this thesis, I explored parasite community variations in a sleepy lizard (*Tiliqua rugosa*) population inhabiting an area with a precipitation-induced ecological gradient, and whether different plant communities from natural vegetation to those more affected by farming can influence these parasite communities. This work is a continuation of a long-established area of exploration of sleepy lizards and their parasites set up by the late Prof. C. Michael Bull who built upon the earlier work by Michael Smyth.

Through the dissection of road kills I confirmed the gastrointestinal helminths that infect lizards in the study site. I caught lizards along established transects in the study site and examined them for the presence of ectoparasites. I also collected blood and faecal samples that were subsequently examined microscopically to determine the prevalence of the various endoparasites in the sleepy lizard population.

Vegetation data were collected from sample plots along two transects that span both disturbed and rehabilitated areas and are therefore representative of the vegetation of the study site. The floristic data was analysed to determine the various plant communities. Based on the identified plant communities, the habitats along the two transects were assigned to different habitat categories.

The geographic distribution of the various identified ecto- and endoparasites were then mapped

out within the study site. Sub-samples of the data collected along the two transects used for the vegetation studies were used to identify associations between different parasite species and various habitat categories.

This work has furthered our knowledge on associations between the parasite community of sleepy lizards and different habitats. The variations in the parasite community were mainly driven by differences in the prevalence of ixodid tick species as a result of their association with specific habitat categories. Precipitation as well as disturbances to the habitat influenced the distribution of these ectoparasites, which influences the degree of overlap in their parapatric distribution. The information about the prevalence of the various parasites across the ecological gradient form a basis for future studies, which should focus on filling the gaps in our understanding of the life cycles the parasites, and identifying the ecological roles they play.

Declaration

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

A handwritten signature in black ink, appearing to read 'Gerrut Norval', written in a cursive style.

Gerrut Norval
24th of September 2021

Acknowledgements

Although this thesis is the result of years of fieldwork and study, it would not have been possible without the encouragement, support and assistance of my parents and wife, whom I have to express my gratitude to. My interest in animals and natural history started at a very young age, and for as long as I can remember my parents, Andries and Rita Norval, have been supporting this interest, whether it was through the books I received as birthday or Christmas gifts, or by allowing me to catch and keep snakes and other animals for observation. And when I grew older my parents continued to provide support and generally being interested in the research I do. These childhood and later life events helped shape me into the ecologist I am today, and for that I am truly grateful. Ecological research requires fieldwork, which means I am away from home for days, and I thank my wife, Liudmila for being understanding and supportive in these times.

To my study supervisors, Assoc. Prof. Michael (Mike) Gardner, Assoc. Prof. Kirstin Ross and Dr. Robert (Bob) Sharrad, I would like to say thank you very much for providing me with more than just guidance in my doctoral studies. When my intended supervisor unexpectedly passed away before my arrival in Adelaide, Mike took over his position and was willing to accept me into the lab. Under his guidance I managed to formulate a new research question that encompassed my skills and background and the research directions he has for the lab. Knowing what my research will entail, he proposed very suitable co-supervisors. Throughout my doctoral studies he supervised the research I set out to do, but also allowed me freedom to investigate new ideas and incorporating additional aspects into my studies. Mike was very supportive and understanding, and encouraged me to undertake activities and training that would develop my career, and also cared about my personal life and was very supportive during the time when my mom passed away and my daughter, Eliana was born. Kirstin not only provided me with opportunities to develop new laboratory skills, but also ensured that I develop as an academic. She reminded me that I should work towards becoming a balanced academic, and that I should develop my skills as a member of the teaching staff. Bob was an invaluable source of information and encouragement. He provided literature, and verified tick specimens I needed a second opinion on, as well as assist in the identification of some plants. His other “duty” was to “keep me from doing work” for an hour or so almost every week; these regular conversations over a cup of coffee were immensely useful since they provided an opportunity for me to discuss my findings. Our discussions about the gaps in the understanding of the natural history of ticks and potential future research, was always encouraging as it reminded me that my PhD study was part of the route I have to take to work towards a career as a tick ecologist.

There was a time constraint for me to complete my doctoral research and there were limitations to my skills. As a result, I had to ask for assistance from others with some parts of my research. I would like

to thank Dr. Lesley Warner (South Australia Museum) for her advice on examining and identifying nematodes. Dr. Bruce Halliday (Australian National Insect Collection, CSIRO) identified the snake mite (*Ophionyssus natricis*) I collected and also provided guidance for me to learn to identify them myself, and for that I am grateful. I thank Prof. Ian Beveridge (University of Melbourne) for verifying the cestode species I collected from sleepy lizards (*Tiliqua rugosa*), and Prof. Charles Bursey (Pennsylvania State University) and Prof. Stephen Goldberg (Whittier College) who identified and verified various helminth species that I collected from some reptiles in the Mid North region of South Australia. Stephan Hennekens provided me with a TURBOVEG database for the flora of South Australia, and Prof. Leslie Brown (University of South Africa) assisted with the phytosociology aspect of my research, for which I am very grateful. I also thank Dr. Ryan Baring (Flinders University) for his assistance with some of the statistics that were used in my thesis.

I collected most of the specimens and samples for my doctoral research, but some specimens and information were provided by others. So I want to express my gratitude to my supervisor, Mike Gardner and Dr. Jessica Clayton (College of Science and Engineering, Flinders University, Adelaide, Australia) and Chantelle Derez (School of Biological Sciences, University of Queensland, St Lucia, Queensland, Australia) for collecting some of the reptiles that were found dead on roads, and Dr. Jessica Nowicki (Biology Department, Stanford University, Stanford, California, USA) for providing parts of the digestive tracts of the lizards used in her study. I also would like to thank Simon Adamczyk of Animal Relocation & Education (Adelaide, Australia) for collecting snake mite from reptiles being relocated in the Adelaide region, Sarah Churcher and Sophie Hammond for collecting snake mite from the sleepy lizards in captivity at Flinders University, and Dr. Jessica Clayton, Seamus Doherty, Dr. Aaron Fenner, Mike and Joyanne Gardner and Sophie Hammond for their assistance with the capturing of some of the sleepy lizards in 2020 and 2021 that were used in certain of my snake mite and kangaroo soft tick (*Ornithodoros gurneyi*) investigations. I also express my gratitude to Dr. Bruce Halliday (Australian National Insect Collection, CSIRO, Canberra, Australia), Dr. Matthew Shaw (South Australian Museum, Adelaide, Australia) and Dr. Owen Seeman (Queensland Museum, Brisbane, Australia) for information and access to the specimens in the collections under their management. Prof. Stephen Barker (University of Queensland, Brisbane, Australia) provided information on the distribution of the kangaroo soft tick in Australia, and Grant Gully (College of Science and Engineering, Flinders University, Adelaide, Australia) and Pat Vilimas (Flinders Microscopy) assisted with the preparation of the

micrographs of some of the parasites I studied, and for that I am grateful. Many parts of my research would not have been possible without access to areas beyond the roads that were used as transects, so I would like to extend a special thank you to the landholders of the region, and particularly Ron and Leona Clarke, John Bouilly and Chris Mosey for their generous and continued hospitality in allowing access to their land, and Courtney Heinjus (DEW Ranger) for providing access to the Hallett Cove Conservation Park.

The preparation of this thesis required literature from various sources, in addition to the staff at the Central Library on the Bedford Park University campus, and I would also like to express my gratitude to Marc Baldwin, David Bird, Jan Grathwohl, Dr. Jean-Jay Mao (National Ilan University, Taiwan), Darrell Raw, Dr. Richard Robbins (Walter Reed Biosystematics Unit, Smithsonian Institution, Washington DC, USA), and my father for their kind assistance in obtaining some of the references that I used.

I would like to thank the Australian Government for the Australian Government Research Training Program Scholarship (International) and the financial support from the College of Science and Engineering of Flinders University to undertake my doctoral studies. My doctoral research was partly funded by grants from the Royal Society of South Australia in 2017 and 2019, Friends of Private Bushland in 2018, and the Joyce W. Vickery Research Fund from the Linnean Society of New South Wales in 2019, and I thank these organizations for their support. I also would like to express my gratitude to the Animal Welfare Committee of Flinders University who provided clearance for my research and the Department of Environment, Water and Natural Resources of the Government of South Australia for the issue of the necessary research permits.

In addition to my supervisors, I would also like to thank all my co-authors for their contributions to the manuscripts that form part of the various chapters of my thesis. I also want to express my gratitude to the editors and reviewers whose suggestions improved the eventual publications. To Dr. Jessica Clayton and Dr. Bonnie Derne I express my gratitude for their advice on the writing and formatting of my thesis, and to all the members of the LEGS lab past and present for their encouragement and support. If I failed to acknowledge someone it is because my memory failed me and not because I am not grateful. In that case, I am sorry and thank you. Last, but not least, I want to thank the lizards and their parasites for being a continuous source of inspiration for further study and for filling my life with amazement, discoveries, and opportunities.

Details of co-authorship

Chapter 1

Published: Norval, G. (1), and M. G. Gardner (2). 2020. The natural history of the sleepy lizard, *Tiliqua rugosa* (Gray, 1825) – Insight from chance observations and long-term research on a common Australian skink species. *Austral Ecology* 45:410-417.

The candidate was the primary author of the manuscript, conducted the literature review, and drafted the manuscript. The candidate was also responsible for the submission of the manuscript, and the subsequent changes for its publication. Author 2 critically reviewed the manuscript.

1: 95%; 2: 5%

Published: Norval, G. (1), K. E. Ross (2), R. D. Sharrad (3), and M. G. Gardner (4). 2019. Taking stock: a review of the known parasites of the sleepy lizard, *Tiliqua rugosa* (Gray, 1825), a common lizard endemic to Australia. *Transactions of the Royal Society of South Australia* 143:216-234.

The candidate was the primary author of the manuscript, conducted the literature review, and drafted the manuscript. The candidate was also responsible for the submission of the manuscript, and the subsequent changes for its publication. Authors 2, 3 and 4 critically reviewed the manuscript.

1: 85%; 2: 5%; 3: 5%; 4: 5%

Chapter 2

Published: Norval, G. (1), C. R. Bursey (2), S. R. Goldberg (3), R. D. Sharrad (4), K. E. Ross (5), and M. G. Gardner (6). 2021. New host and locality records for gastrointestinal helminths of five reptile species from the Mid North region of South Australia. *Transactions of the Royal Society of South Australia*:1-15.

The candidate collected 21 (70%) of the reptiles that were found dead on roads, and dissected all the collected roadkills and collected helminths from them. He also dissected all the digestive tracts from eastern blue-tongue skink (*Tiliqua scincoides*) and sleepy lizards from an unrelated study that were donated to the candidate for study and collected helminths from them. The candidate determined the species of nematode that was collected from all the sleepy lizards, which was then verified by author 2. Author 2 determined the species of all the other collected

helminths. The candidate drafted the 1st draft of the manuscript and submitted it after incorporating all the input and corrections of co-authors 2-6 who critically reviewed the manuscript. The candidate made the changes as suggested by the reviewers and was the corresponding author for the final editing of the manuscript for publishing.

1: 60%; 2: 20%; 3: 10%; 4: 5%; 5: 5%

Chapter 3

Published: Norval, G. (1), B. Halliday (2), A. Sih (3), R. D. Sharrad (4), and M. G. Gardner (5). 2020. Occurrence of the introduced snake mite, *Ophionyssus natricis* (Gervais, 1844), in the wild in Australia. *Acarologia* 60:559-565.

The candidate was the primary author of the manuscript, and with the assistance of author 2 drafted the manuscript. He collected all the lizards that were brushed for snake mite in 2019, brushed them, and examined the material that was obtained for the presence of snake mites. The candidate identified the snake mite that were collected from the brushed lizards, and author 2 verified the identifications. Authors 2, 3, 4 and 5 critically reviewed the manuscript. The candidate submitted the manuscript, and made the changes as suggested by the reviewers and was the corresponding author for the final editing of the manuscript for publishing.

1: 70%; 2: 20%; 3: 2%; 4: 5%; 5: 3%

Published: Norval, G. (1), B. Halliday (2), R. D. Sharrad (3), and M. G. Gardner (4). 2021. Additional instances of snake mite (*Ophionyssus natricis*) parasitism on sleepy lizards (*Tiliqua rugosa*) in South Australia. *Transactions of the Royal Society of South Australia*:1-11.

The candidate collected some of the lizards that were used in this study, but brushed all the lizards that were used for this study for snake mite, and examined the material that was obtained for the presence of snake mites. He identified the snake mite that were collected, and author 2 verified the identifications. The candidate drafted most of the 1st draft of the manuscript, with the assistance of author 2 and submitted it after he incorporated all the input and corrections of authors 2, 3 and 4. He made the changes as suggested by the reviewers and was the corresponding author for the final editing of the manuscript for publishing.

1: 80%; 2: 15%; 3: 3%; 4: 2%

Chapter 4

Published: Norval, G. (1), R. D. Sharrad (2), and M. G. Gardner (3). 2022. A mammal tick

with a taste for lizard blood: parasitism by the kangaroo soft tick, (*Ornithodoros gurneyi*) on sleepy lizards (*Tiliqua rugosa*). Ticks and Tick-borne Diseases 13:101859.

The candidate collected the majority of the lizards (93%), and examined all the lizards used in this research for soft ticks, and identified the species of all the ticks that were found. He collected all the samples from kangaroo wallows and identified all the ticks that were collected. He also conducted the feeding and moulting experiment and collected all the related data. The candidate analysed all the data that was collected for this project, and drafted the first draft of the manuscript. He made the changes as suggested by authors 2 and 3, and submitted the manuscript. He made the changes as suggested by the reviewers and resubmitted the manuscript.

1: 90%; 2: 5%; 3: 5%

Chapter 5

Manuscript: Norval, G. (1), L. R. Brown (2), R. D. Sharrad (3), & M. G. Gardner (4). The roadside plant communities along two transects spanning an ecological gradient, surveyed during a period of drought at a locality near Mt. Mary, South Australia.

The candidate determined the plot sizes and conducted all the surveys. He entered the data into the programme JUICE and performed the phytosociology analyses, which was then verified by the authors 2 and 3. With the assistance of author 2, the candidate determined the species richness and diversity indices of the different plant communities. He drafted the first draft of the manuscript, which was critically reviewed by author 2. The candidate revised the manuscript, which was then critically reviewed by authors 2, 3 and 4.

1: 70%; 2: 20%; 3: 7%; 4: 3%

Chapter 6

Manuscript: Norval, G. (1), R. Baring (2), R. D. Sharrad (3), M. G. Gardner (4), and L. R. Brown (5). The distribution of ecto- and endoparasites of sleepy lizards (*Tiliqua rugosa*) sampled across an ecological gradient in the Mid North region of South Australia and their habitat association.

The candidate caught and examined all the lizards that were used for this research for the presence of ectoparasites, and identified all the observed ectoparasites. He collected faecal samples and performed the faecal flotation and direct smear examinations, and identified the observed helminths. He also collected all the blood samples and prepared blood smear slides,

which he stained, cover-slipped and examined for the presence of haemoparasites. Where possible, he identified the observed haemoparasites, and in other instances assigned the observed intracellular inclusions to preliminary morpho-groups. With the assistance of author 2, the candidate examined the association between the different parasite species and the habitat categories. He drafted the manuscript, which was then critically reviewed by authors 2, 3, 4 and 5.

1: 70%; 2: 15%; 3: 5%; 4: 5%; 5: 5%

Preface

This thesis consists of five data chapters (Chapters 2 to 6) that sets out the research contributing to the fulfillment of a doctoral program. These chapters are preceded by a general introduction (Chapter 1) containing a broad introduction to parasites and their ecological roles, the natural history of the host species, a summary of its parasites, and background and main aims of my doctoral study. The data chapters are followed by a general discussion of the key findings, their implications and future research directions (Chapter 7). A ‘note to examiners’ precedes the introduction and each of the data chapters and details the main aspects of the chapter and what it entails. Chapters 1 and 3 consist of more than one published journal article, which are presented as separate sections within the chapter, each as a stand-alone article, with its own abstract (if included in the published version) and own bibliography. If stylistic changes were made to these articles, these changes are stipulated in the ‘note to examiner’ section. Chapters 2, 4, 5 and 6 are set out as individual research articles, each with an abstract and bibliography, as well as supplementary materials in the case of Chapter 6. Some background overlap of chapters was therefore inevitable. Chapter 4 was reviewed by an anonymous reviewer and the section editor of the journal *Ticks and Tick-borne Diseases*, and the resubmitted revised version of the manuscript was included in this thesis. Chapters 5 and 6 are manuscripts that are yet to be submitted to scientific journals for consideration.

I conducted the majority of the work presented in this thesis, but in most of the chapters of my thesis the manuscripts were not altered so the pronoun “we” is used as apposed to the singular “I”, acknowledging the contributions of the co-authors, whose contributions are outlined in the details of co-authorship (the preceding section of this thesis).

Chapter 1

Thesis Introduction

Note to examiners

The introductory chapter of my thesis consists of four parts, each with a different focus, but a small degree of unavoidable overlap exists. The first part provides introduces the importance of studying parasite communities and then sets out the research aims. The second part is an introduction to the sleepy lizard (*Tiliqua rugosa*), the host species of my study, presented as a review of what is known about its natural history, published in the journal *Austral Ecology*. In that publication, the associated tables were made available as online appendixes due to manuscript length limits. Herein the tables were incorporated into the text for ease of reading. The third part of the chapter is a review of the known parasites of sleepy lizards, which was published in *Transactions of the Royal Society of South Australia*. The final part of this chapter is the outline of my thesis. The second and third part of this chapter formatted according to the specifications of the respective journal (e.g. *Transactions of the Royal Society of South Australia* requires referencing and indicating the authority and publication date when a species is first mentioned in the text). The referencing style of the published reviews and the abbreviations in the associated tables were changed to make them consistent with the style of the other chapters of the thesis. The numbering of the tables was changed so that they appear chronologically in the chapter and additional figures were also incorporated. Unless stated otherwise, I took the photographs and micrographs and therefore own the copyright.

Parasites and their ecological importance

What are parasites?

A parasite is an organism that is reliant on a host (i.e. another organism) for its existence by living in or on the host and feeding on its tissue or nutrition (Fripp 1979, Roberts and Janovy 2005). Parasites can be unicellular or multicellular organisms, and most phyla have parasitic forms (Poulin and Morand 2000). Some parasites, such as a mosquitoes (Fig. 1.1), are only parasitic during part of their life cycle, while others, like pentastomes (Fig. 1.1), are parasitic at all post-embryonic stages of their life cycle (Byford and Maurice 2007).

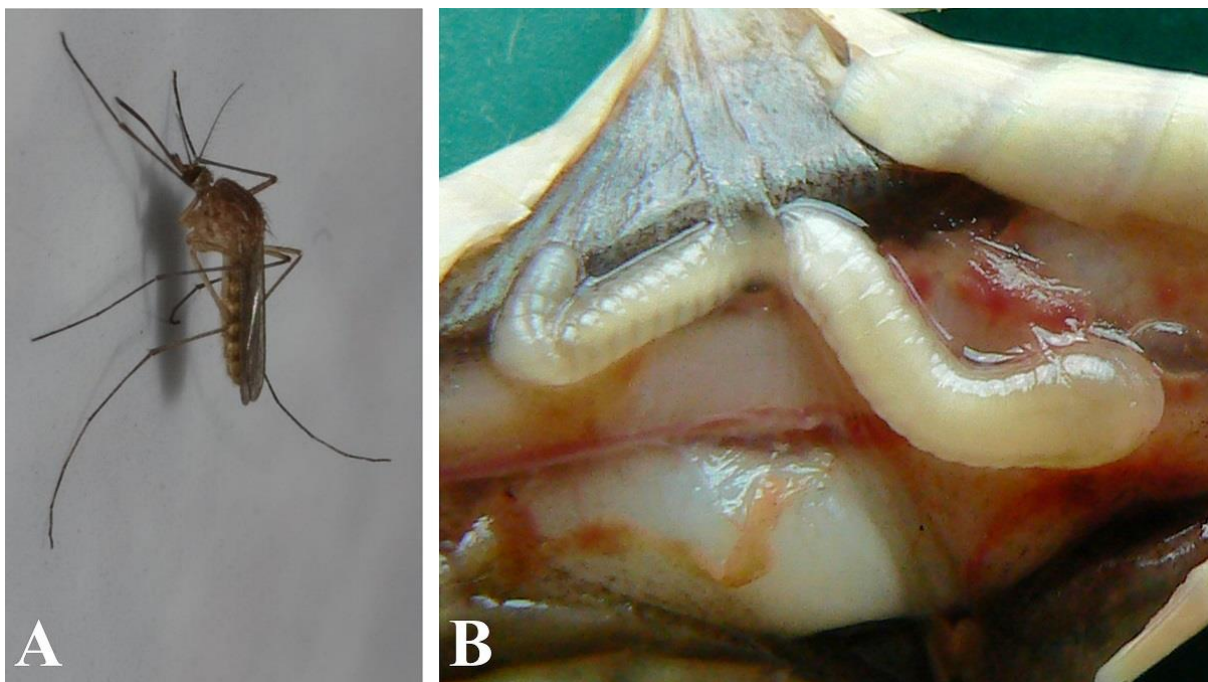


Figure 1.1. Only the adult female of *Culex* sp. (A) are parasitic and needs a blood meal to reproduce, whereas the larvae, nymphs (B) and adults of the pentastome *Kiricephalus pattoni* are endoparasites of some Asian amphibians and reptiles.

Parasitism is a symbiotic relationship that is in favour of the parasite as it exists at the expense of the host. The costs associated with parasite infection or infestation can vary. On the one end of the spectrum it is a mere irritation and the associated energetic cost of carrying the parasites and seeking relief from the discomfort they cause. Pinworm infections in mammalian hosts are a good example of this because pinworms do not feed on the ingested food or the tissue of the host, but instead feed on endosymbiotic bacteria and protists living in the digestive tract of their hosts (Hugot et al. 2014). Under most conditions the only costs linked to them, in otherwise healthy hosts, is the discomfort these worms cause when they deposit their eggs around the anal opening of the host (Lapage 1963, Fripp 1979). On the other end of the spectrum, parasite

infections or infestations can be severely debilitating. For instance, infections by certain digenian flukes cause polydactyly in some North American anurans (Johnson et al. 2003) and the filarial nematodes *Brugia malayi*, *Brugia timori* and *Wuchereria bancrofti* cause disfiguring lesions in humans in parts of Africa, Asia and Oceania (Hoerauf et al. 2011, Gordon et al. 2018). Many parasites, like some hemipterans, mosquitoes, and ticks are also vectors of pathogens that impact humans directly by causing morbidity and even mortality (Cholewiński et al. 2015), while a variety of parasites impact humans indirectly through economic losses associated with parasite infections (Evans and Jamison 1994, Nalradkar 2018). As a result, parasites are generally viewed with disdain and are often regarded as organisms that should be eradicated or at least controlled and, even among ecologists and conservationists, parasites have been overlooked as organisms that warrant conservation (Lapage 1963, Wood and Johnson 2015).

Why do parasites matter?

The loss of some parasites may reduce human morbidity, mortality and /or financial losses, but the loss of parasites from the ecosystems they are a part of can have cascading negative impacts due to their ecological roles (Wood and Johnson 2015). Although not readily observable, some parasites play a role in species distributions and the community structure of their hosts (Hatcher et al. 2006). For instance, the tick *Amblyomma hebraeum* is the main vector for the rickettsial causative agent of heartwater disease, *Ehrlichia ruminantium* (Jongejan et al. 2020). The tick is limited to warm humid wooded regions of southern Africa and is absent in grasslands (Norval 1977, Spickett 2013). Some antelope such as the greater kudu (*Tragelaphus strepsiceros*) has a habitat preference for wooded areas and are the hosts of *A. hebraeum* and evolved an innate immunity to *E. ruminantium* (Grunow 1980, Burkepile et al. 2013, Oberem 2017). The impala (*Aepyceros melampus*) and springbuck (*Antidorcas marsupialis*) are mixed feeders (i.e. grazers and browsers), but while the impala primarily occurs in wooded savanna, the springbuck is limited to savanna desert and other semi-desert habitats (Grunow 1980, Burkepile et al. 2013). The impala is susceptible to *E. ruminantium* but can acquire immunity (Oberem 2017), whereas the springbuck is highly susceptible to *E. ruminantium* and unable to acquire immunity, and as a result, naturally occurs outside areas where this pathogen is present (Neitz 1944, Oberem 2017). Under natural conditions *A. hebraeum* and *E. ruminantium* therefore restricts habitat overlap between species like the impala and springbuck and the associated competition, illustrated by the fact that when springbuck is vaccinated against *E. ruminantium* this species

can be sympatric with other antelope in areas outside its native range (Anderson et al. 2016, Oberem 2017). It should also be noted that parasites do not only prevent species from being sympatric but can also promote it (Hatcher et al. 2006). An example of such a condition can be seen in St. Maarten, where the less competitive lizard, *Anolis watsi*, is only sympatric with the more competitive *Anolis gingivinus* in habitats where the malarial parasite *Plasmodium azurophilum* infects *A. gingivinus* (Schall 1992). *Plasmodium azurophilum* rarely infects *A. watsi* but *A. gingivinus* is more susceptible and when these lizards are infected they tend to be less competitive (Schall 1992).

Parasites do not just manipulate the community structure of potential hosts but often also influence energy flow. Some parasitic plants, like the harlequin mistletoe (*Lysiana exocarpi*), produce fruit (Fig. 1.2) that is a food source for some animals such as the mistletoe bird (*Dicaeum hirundinaceum*), while some parasitic arthropods, such as mosquitoes, are preyed upon by a variety of other animals, such as spiders and some insectivorous bats (Yan 1993, Ndava et al. 2018, Puig-Montserrat et al. 2020).



Figure 1.2. The fruit of the harlequin mistletoe (*Lysiana exocarpi*) (A) is eaten by a bird, such as the mistletoe bird (*Dicaeum hirundinaceum*), but the parasite does not infect the bird. Instead, it simply uses the bird to transport the seed to a potential host, where it will germinate if it is deposited along with the faeces of the bird on a stem of the potential host (B).

Other parasites, like some nematomorphs induce their arthropod hosts to seek out water, and by doing so contribute significantly to the energy flow in riparian ecosystems (Sato et al. 2011). Various parasites manipulate their intermediate hosts in bizarre manners in order to be ingested by their definitive hosts. For instance, the cestode, *Anomotaenia brevis*, infects the workers of

the ant, *Temnothorax nylanderi*, and increases the longevity of infected ants and manipulates them into remaining within the nest to increase the likelihood of them being eaten by an ant-eating woodpecker (Beros et al. 2021). Others make the infected intermediate host more obvious to potential definitive hosts. For example, the nematode, *Myrmeconema neotropicum*, changes the colour of the abdomen of infected workers of the ant, *Cephalotes atratus* so that they resemble berries and are therefore more likely to be eaten by a bird (Poinar and Yanoviak 2008), and the trematode *Euhaplorchis californiensis* induces California killifish (*Fundulus parvipinnis*) to flash their sides and therefore become more visible to piscivorous birds (Lafferty and Morris 1996).

By making intermediate hosts, and even definitive hosts, more susceptible to predation, parasites also play a role in regulating host populations (Scott and Dobson 1989, Tompkins and Begon 1999). Many parasitic infections and infestations are asymptomatic in healthy hosts because the parasite does not benefit from killing the host (Fripp 1979). In fact, in most instances the death of the host will eventually lead to the death of the parasite. Killing the host would therefore not be an evolutionary stable strategy if it means limited dispersal for the parasite (Ewald 1995). However, hosts with reduced immunity or resistance are at risk of greater co-infections and intensities of parasites, which often causes mortality, and by doing so parasites keep the host population healthy by reducing the population size and the associated competition, and at the same time prevent less fit individuals from breeding (Marcogliese 2004, Wood and Johnson 2015). The death of hosts is not always necessary in order for parasites to reduce host population sizes. For instance, the saurian malarial parasite, *Plasmodium mexicanum*, reduces the clutch sizes of infected western fence lizards (*Sceloporus occidentalis*), but does not cause mortality in these lizards even though infected lizards also tend to have lower fat reserves (Schall 1990, Eisen 2001). In addition, Vézilier et al. (2012) found that in the mosquito, *Culex pipiens*, *Plasmodium* sp. infected females had lower fecundity but increased survival.

An often-overlooked role of parasites is their impact on each other. Co-infections may result in a greater burden on the health of the host (Bordes and Morand 2011), but this is not always the case. As in free-living organisms, there can be direct and indirect competition among parasites for resources (Dobson 1985). In some species, such as the oxyurid *Protrellus dixonii*, overcrowding within the arthropod host is prevented through interference competition which prevents infection by conspecifics (Zervos 1988b, a). such competition can have health benefits

to the host. For instance, the endangered woylie (*Bettongia penicillata*) are parasitized by at least four trypanosome species, of which only *Trypanosoma copemani* has been determined to be pathogenic, but only when there is a lack of competitive interaction from the other trypanosome species (Thompson et al. 2014, Thompson et al. 2018). Parasite interactions can be complex and may involve different species that infect or infest different parts of the host's body (Shen et al. 2019). For instance, in free-ranging rabbits (*Oryctolagus cuniculus*) in the United Kingdom the blood-feeding stomach worm *Graphidium strigosum* initially suppresses the immune response of the host, which results in an increase in the numbers of the intestinal mucosal browsing worm *Trichostrongylus retortaeformis*, but as the host age and the numbers of *T. retortaeformis* increase they stimulate an acquired immunity in the host which causes a decrease in the numbers of *G. strigosum* (Lello et al. 2004). Similar indirect interactions have also been noted in sheep (*Ovis aries*), involving the stomach worm *Haemonchus contortus* and the intestinal worm *Trichostrongylus colubriformis* (Lello et al. 2018). It is therefore evident that an understanding of the parasite communities of a particular host species is required in order to elucidate the roles that the parasites play in the ecology of the host species.

Parasites and ecological gradients

As a result of various ecological factors parasites tend to be heterogeneously distributed within a host population (Pullan et al. 2012). The diet and social network of individuals of the host species determines to some extent their parasite communities (Godfrey 2013, Grear et al. 2013, Leung and Koprivnikar 2019). However, many parasites are for part of their life cycle not in or on the host, and during this stage their survival can be negatively influenced by environmental conditions such as humidity and temperature (Cumming 2002, Blum and Hotez 2018, Preisser 2019). Others have complex life cycles, involving intermediate hosts or vectors, and are therefore indirectly influenced due to the constraints imposed by the specific environmental condition requirements of the intermediate hosts or vectors (Zamora-Vilchis et al. 2012). A striking example of this is the expanding range of human malaria (*Plasmodium* spp.) and tick-borne diseases such as Lyme disease, caused by *Borrelia burgdorferi*, as a result of climate change, which makes former unsuitable areas more suitable for the vectors of these pathogens (Caminade et al. 2014, Gilbert 2021). If the parasite or vector has a wide environmental conditions tolerance and is not very host specific its distribution range can exceed that of some of its host species. For instance, the tick, *Ixodes ricinus* parasitizes a variety of birds, mammals and reptiles in Europe, and has a wide environmental conditions tolerance,

so it is not surprising that its distribution range exceeds that of some of its hosts (Estrada-Peña and de la Fuente 2017, Černý et al. 2020). However, most parasites and vectors have more limited environmental conditions tolerances and as a result they often do not occur throughout the distribution range of the host. For example, *Ixodes holocyclus* (Fig. 1.3), like *I. ricinus*, is not very host specific and has been recorded from 34 species of mammals and seven species of birds, and yet its distribution is limited to coastal eastern Australia due to the susceptibility of its eggs and larvae to desiccation and its dependency on stable populations of bandicoots, *Isodon macrourus* and/or *Perameles nasuta* (Barker and Walker 2014).



Figure 1.3. The paralysis tick, *Ixodes holocyclus* occurs along the eastern coastal region of Australia in suitable habitats, with stable populations of bandicoots, its primary hosts. The chemical compounds secreted by feeding females of this tick can cause paralysis if the tick remains attached for extended periods (days) of time.

Parasite communities therefore spatially vary, and these variations can be explored by comparing parasite species assemblage across ecological gradients. This can be done from a broad scale, involving multiple host species, such as the comparisons across latitudinal gradients (Guernier et al. 2004, Nunn et al. 2005, Preisser 2019), or the comparisons can also be done on a more regional scale, often focussing on specific parasites and hosts, as done by Zamora-Vilchis et al. (2012), who examined avian haemoparasite prevalence in 40 bird species along an elevational gradient. Such comparisons can even be used to examine the impact of anthropogenic disturbance on parasites, as were done by Coates et al. (2017) and Calegario-Marques and Amato (2014) who investigated the prevalence of specific parasites across a

degree of urbanization gradient.

Study species and study site location

The sleepy lizard (*Tiliqua rugosa*) (Fig. 1.4) is a common large skink species that inhabits a variety of habitats of southern Australia (Cogger 2014). Various aspects of the ecology of this species have been studied and many chance observations on its biology and behaviour have been reported. It is therefore one of Australia's better-studied species and in the second section of this chapter the natural history and ecology of sleepy lizards will be elaborated on in more detail.



Figure 1.4. The sleepy lizard (*Tiliqua rugosa*), also known as a bogey, pinecone lizard, shingleback, stumpy-tailed skink or two-headed skink, is a common lizard species in many parts of southern Australia.

Throughout its range *T. rugosa* has been reported as a host of a variety of parasites, but as indicated in the third section of this chapter, most remain relatively poorly studied (Norval et al. 2019). At the Bunday Bore study site ca. 160 km north of Adelaide near Mt. Mary in the Mid North region of South Australia (Fig. 1.5), long-term studies (since 1982) initiated by the late Prof. Mike Bull found that the ticks *Amblyomma limbatum* (Fig. 1.6) and *Bothriocroton hydrosauri* (Fig. 1.7) have an abrupt parapatric boundary that is maintained along a gradual

environmental gradient (Bull and Burzacott 2001). Apart from the research pertaining to the two ticks, and the infection dynamics of the haemogregarine blood parasite, *Hemolivia mariae* in the ticks and *T. rugosa* (Smallridge and Bull 2000, Smallridge and Bull 2001a, b), surprisingly little is known about the parasite community of this sleepy lizard population.



Figure 1.5. The Bundey Bore study site in relation to Adelaide, South Australia, and the layout of the dirt roads (white lines in inset) that are used as the transects for the long-term monitoring of sleepy lizards (*Tiliqua rugosa*).

Only one study surveyed helminth parasites, and it focussed only on the prevalence of the cestode *Oochoristica trachysauri* and the nematode, *Thelandros trachysauri* in this *T. rugosa* population in part of the study site (Gyawali et al. 2013). Sharrad (1979) recorded the kangaroo soft tick (*Ornithodoros gurneyi*) (Fig. 1.8) as an occasional parasite of *T. rugosa* in this locality, but this tick species has not been incorporated into any of the research done at the Bundey Bore

study site.



Figure 1.6. *Amblyomma limbatum* is a reptile tick adapted to xeric habitats so occurs throughout the arid and semi-arid parts of Australia.



Figure 1.7. *Bothriocroton hydrosauri* is a reptile tick that is sensitive to desiccation and therefore is limited to more humid habitats of southern parts of Australia.

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The natural history of the sleepy lizard (*Tiliqua rugosa*) – insight from chance observations and long-term research on a common Australian skink species

Natural history is the basis for the understanding of the biology of a species and its ecological roles (Bury 2006, Lindenmayer et al. 2012). The development of an understanding of the natural history of a species often require long-term research, since aspects such as longevity, are usually difficult to determine otherwise (Fitch 2006). In addition, many ecological processes, like succession and the associated community changes, usually take place at a pace that cannot be observed over a relatively short period of time, making long-term data collection invaluable (Fitch 2006). In spite of this, the current trend among most academic institutions is to favour biochemical and/or molecular studies, which produce results relatively quickly, over long-term natural history or ecological research (Greene and Losos 1988, Pianka 2001, Tewksbury et al. 2014). This is a result of pressures that these institutions face for obtaining funding and other support, coupled with a lack of appreciation of natural history and ecological studies and decrease in interest in the natural environment among members of the general public (Greene 1994, Mccallum and Bury 2013). To enable long-term research, ecologists and other natural historians, therefore often have to run short-term projects, with more immediate outputs, in order to find a balance between meeting the demands of producing timely results and obtaining the necessary information that only long-term research can provide. This requires a genuine interest in the study subjects and a degree of resourcefulness. A successful strategy is to use a long term system as a scaffold to ‘hang’ individual projects from. The research on *Tiliqua rugosa* (Gray, 1825), mostly done in South Australia by the late Professor Michael Bull and his collaborators, is a clear illustration of how chance observations and short-term studies can be incorporated with long-term data collection for the unravelling of the ecology of a species. Herein we summarise the natural history findings from this impressive long-term data collection which started in the late 1970’s and from 1982 was continuous around road transects surrounding Bunday Bore Station (33°54’S; 139°20’E) in the Mid North region of South Australia. We incorporate these data with chance observations and other research to provide a comprehensive overview of what is known about the natural history of *T. rugosa*.

Distribution and Habitat

The sleepy lizard (*Tiliqua rugosa*, formerly classified as *Trachydosaurus rugosus*) is a relatively large (total length ca. 40 cm) skink species (Gray 1825, Shea 1990, Bull and Pamula 1996, Cogger 2014), endemic to the semi-arid parts of southern Australia, from Western Australia eastwards to southern Queensland (Cogger 2014, Parsons et al. 2015). It is a habitat generalist and can inhabit arid *Acacia* scrublands, chenopod shrublands, coastal heaths, dry sclerophyll forests, eucalypt scrublands, gibber plains, mallee scrublands, spinifex-dominated sandy deserts and woodlands (Cogger 2014). This species however, favours fairly open habitats dominated by shrubs, as opposed to habitats with dense tree canopy cover (Henle 1990, Bull 1995). Within these habitats these lizards establish home ranges that, depending on the season and/or climate, vary in size from about three to nine hectares (Satrawaha and Bull 1981, Dubas and Bull 1992, Kerr and Bull 2006a, Bull et al. 2017). This home range size is relatively small compared to those of similar-sized carnivorous lizards, and is possibly due to its omnivorous diet (Satrawaha and Bull 1981).

The initial studies into the home ranges of sleepy lizards found that the home ranges of individuals overlap and are spatially and temporally stable over consecutive years (Bull 1978, 1987, Bull and Freake 1999). It was also found that the size of the home range of each lizard is not influenced by food availability (Dubas and Bull 1991, 1992) but rather by access to water (Kearney et al. 2018), since during periods of drought lizards have smaller home ranges (Kerr and Bull 2006a) and lizards with access to water tend to maintain larger home ranges (Leu and Bull 2016). The analyses of data gathered over consecutive years indicates though that there is a subtle, but crucial association between spatial food resources and refuges, which influences the patterns in which sleepy lizards utilize their home ranges (Leu et al. 2010a, Malishev et al. 2017).

Sleepy lizards are ectotherms and prefer a body temperature of about 33° to 35°C (Warburg 1965, Licht et al. 1966, Bennett and John-Alder 1986, MacMillen et al. 1989, Firth and Belan 1998), which they maintain through basking or seeking shelter when thermal conditions become unfavourable, so it is not surprising that their activity patterns are influenced by the time of year and the time of day. Studies into their activity patterns have determined that the lizards are primarily active during the warmer mid part of the day (i.e. unimodal) on days of mild temperature, usually during the late winter to spring period, to being active mainly during

the cooler morning and afternoon periods (i.e. bimodal) on hot days, primarily during the late spring to summer period (Firth and Belan 1998, Kerr and Bull 2006b). However, it should also be noted that there are inter-sexual differences in activity patterns. Data collected over multiple years indicates that male sleepy lizards are more active than the females prior to mating as a result of remaining active later in the day than the females and traveling greater distances (Bull et al. 1991, Kerr and Bull 2006b), likely to forage or even for opportunities of extra-pair mating (Leu et al. 2011b). After mating, the activity of the males decrease, most likely as a result of more effective foraging in conjunction with reduced energetic demands associated with reproduction (Leu et al. 2011b). The females, on the other hand, need to thermoregulate and meet the energetic demands of the developing embryos, so do not reduce their activity to the same extent as the males and likely only become inactive when weather conditions become unsuitable for activity (Fergusson and Algar 1986, Bull et al. 1991, Leu et al. 2011b).

When prevailing temperatures are unfavourable sleepy lizards usually seek shelter in the burrows of other animals (Kerr et al. 2003). At other times of inactivity, the lizards usually make use of shrubs, hollow logs, or man-made objects such as sheets of corrugated iron (Fig. 1.10) as refuges (Henle 1990, Kerr et al. 2003, Lancaster et al. 2012). Studies into the utilization of refuges found that sleepy lizards make use of numerous refuges within their home ranges (Leu et al. 2010b, 2011a) and that they do not select the refuges at random, but rather select refuges and positions within them depending on their thermal requirements (Kerr et al. 2003, Kerr and Bull 2004b, Auburn et al. 2009).

The multiple-year study into the home ranges of sleepy lizards have also found that males usually occupy larger home ranges than those of females (Godfrey et al. 2013, Spiegel et al. 2018), and that both sexes do not make constant use of their entire home-ranges. Instead they utilise exclusive core areas, which usually overlap with that of their paired partner, which was proposed as suggestive of intra-sexual territoriality (Kerr and Bull 2006a). Unlike other species in the sub-family Egerniinae, sleepy lizards do not make use of scat piling to indicate occupancy of their home ranges, but rather likely employ other forms of scent-marking (Fenner et al. 2015, Leu et al. 2016). The periphery of their home ranges (i.e. the part of their ranges outside of the core areas) overlap with those of neighbours and earlier studies suggested that although neighbours may periodically utilise the same refuges within these periphery areas they usually do so asynchronously, which was interpreted as a form of avoidance behaviour (Spiegel et al. 2015). However, more recent analysis of the available data, incorporating lizard

movements, social networks (i.e. which individuals interact with each other and how often), and personality (i.e. the aggressiveness and boldness of individuals) suggest intra-sexual interactions in the peripheral areas are more likely a form of excluding behaviour since they take place relatively far from the centre of exclusive core areas and were at least in some cases motivated by agonistic behaviour (Spiegel et al. 2018), thereby supports earlier assumptions of intra-sexual territoriality.



Figure 1.10. A discarded sheet of corrugated iron in a paddock in the Bunday Bore study site (A) and a sleepy lizard (*Tiliqua rugosa*) that was using it as a shelter (B).

Diet and feeding behaviour

The diet of a species is a crucial component of its natural history because in addition to growth and maintenance, the food sexually mature lizards consume provides energy for reproduction (Derickson 1976, Storer et al. 1979). *Tiliqua rugosa* has conical teeth that tend to be relatively larger along the middle of the jaw, compared to the other teeth (Greer 1989, Shea 1990) This is believed to be an evolutionary adaptation for an omnivorous diet (Estes and Williams 1984). Although sleepy lizards are omnivores that will opportunistically feed on invertebrates and even carrion (Table 1.1), they primarily feed on plant material (Table 1.2), and even have specific adaptations, such as cecal ridges in the colon, which are adaptations associated with herbivorous lizards (Herrel 2007). From long-term data collection it has been determined that *T. rugosa* mainly feed on the dominant available food (i.e. being opportunistic), so their diets tend to vary seasonally, and they favour a varied diet and thus consume different food items

each day (Dubas and Bull 1991). They also do not usually consume all the available food (Fig. 1.11), but feed for a short while, and then move several metres to feed in a different locality, even when food is still available in the initial locality (Henle 1990, Dubas and Bull 1991, 1992). Even though this may suggest that a large part of each day should be allocated to feeding, through the long-term studies into the foraging behaviour and activity patterns of *T. rugosa* it has been determined that irrespective of the gender or the time of year, these lizards spend about the same amount of time foraging, which constitutes only a small part (on average < 12 minutes) of each day (Dubas and Bull 1991).



Figure 1.11. A sleepy lizard (*Tiliqua rugosa*) feeding on the shoots of wards weed (*Carrichtera annua*) growing along the sides of a dirt road in the Mid North region of South Australia.

Considering the extensive distribution of *T. rugosa*, and the fact that it is an opportunistic omnivorous species, it is unlikely that its diet would be the same throughout its range. Additional dietary studies and observations are therefore required from throughout the distribution range of this species to elucidate a comprehensive understanding of its diet, and how environmental change (due to climate change and/or anthropogenic habitat disturbances) may influence it or may already be having an impact on it. In addition to that, studies are also needed to determine if there are changes in the diet of different age classes, and if so, to what extent.

Table 1.1. Reported non-plant dietary items of *Tiliqua rugosa* (* – non-native species; WA – Western Australia; NSW – New South Wales; VIC – Victoria; SA – South Australia).

Family	Species	Locality	Reference
Cyanobacteria			
Nostocaceae	<i>Nostoc commune</i>	SA	Kerr and Bull (2004a)
Fungi			
	Unspecified	WA, SA	Longley (1944), Peters (1973), Yeatman (1988), Shea (1989)
Molluscs			
Hygromiidae	<i>Cerneuella virgate</i> *	SA	Satrawaha and Bull (1981), Yeatman (1988)
Helicidae	<i>Theba pisana</i> *	SA	Dubas and Bull (1991)
	Unspecified snails	WA	Yeatman (1988), Shea (1989)
Arthropods			
Acrididae	<i>Chortoicetes terminifera</i>	SA	Dubas and Bull (1991)
Acrididae	Unspecified grasshopper	WA	Dell and Chapman (1979), Chapman and Dell (1980)
Carabidae	<i>Calosoma schayeri</i>	WA	Chapman and Dell (1980)
Carabidae	<i>Promecoderus</i> sp.	WA	Shea (1989)
Carabidae	Unspecified ground beetle	SA	Yeatman (1988)
Curculionidae	Unspecified weevil	WA, SA	Dell and Chapman (1979), Yeatman (1988)
Ixodidae	Unspecified tick	SA	Yeatman (1988)
Scarabaeidae	<i>Onthophagus duboulayi</i>	WA	Shea (1989)
Scarabaeidae	Unspecified (Melolonthinae)	WA	Shea (1989)
Scarabaeidae	Unspecified scarab beetle	SA	Yeatman (1988)
Tenebrionidae	Unspecified piedish beetles	WA	Henle (1990), Shea (1989)
	Unspecified arachnid	WA	Shea (1989)
	Unspecified beetles	SA	Satrawaha and Bull (1981), Yeatman (1988)
	Unspecified bugs	WA	Chapman and Dell (1980)

Table 1.1. continued

	Unspecified crustacean	WA	Shea (1989)
	Unspecified dipteran (pupae)	WA	Shea (1989)
	Unspecified dragonfly	WA	Chapman and Dell (1980)
	Unspecified insect larva	WA	Shea (1989)
	Unspecified lepidopteran larva	WA	Yeatman (1988), Shea (1989)
	Unspecified millipede	WA	Shea (1989)
	Unspecified roach	WA	Yeatman (1988), Shea (1989)
	Unspecified spider	SA	Yeatman (1988)
Vertebrates			
Muridae	<i>Mus</i> sp.	SA	Yeatman (1988)
Myobatrachidae	<i>Heleioporus psammophilus</i>	WA	Brooker (2016)
Scincidae	Unspecified	SA	Dubas and Bull (1991)
Unspecified	Nestling birds	SA	Dubas and Bull (1991)
Other			
	Lizard scales (dermatophagy)	SA	Yeatman (1988)
	Unspecified invertebrates	SA	Dubas and Bull (1991)
	Rabbit bones	WA	Dell and Chapman (1979)
	Rabbit scats	SA	Dubas and Bull (1991)
	Carrion	SA	Satrawaha and Bull (1981)
	Carrion (mammalian caudal vertebrae)	WA	Shea (1989)
	Carrion (<i>Macropus</i> sp.)	SA	Dubas and Bull (1991)
	Carrion (<i>Macropus fuliginosus</i>)	SA	Norval et al. (2018)
	Carrion (<i>Oryctolagus cuniculus</i>)	VIC	Simpson (1973)
	Carrion (<i>Pogona barbata</i>)	VIC	Simpson (1973)
	Carrion (<i>Varanus</i> sp.)	NSW	Henle (1990)

Table 1.2. Reported plant-based dietary items of *Tiliqua rugosa* (* – non-native species; WA – Western Australia; NSW – New South Wales; VIC – Victoria; SA – South Australia).

Family	Species	Notes	Locality	Reference
Aizoaceae	<i>Mesembryanthemum crystallinum</i> *	fruit	WA	Tubb (1938)
Aizoaceae	<i>Tetragonia implexicoma</i>	fruit	SA	Yeatman (1988), Dubas and Bull (1991)
Amaranthaceae	<i>Chenopodium baccatum</i>	fruit	SA	Dubas and Bull (1991)
Anacardiaceae	<i>Schinus molle</i> *	fruit	SA	Norval et al. (2018)
Asparagaceae	<i>Acanthocarpus preissii</i>	fruit	WA	Peters (1973), Shea (1989)
Asparagaceae	<i>Asparagus asparagoides</i> *	fruit	SA	Yeatman (1988)
Asphodelaceae	<i>Dianella</i> sp.	fruit	SA	Yeatman (1988)
Asteraceae	<i>Angianthus tomentosus</i>	flowers	SA	Dubas and Bull (1991)
Asteraceae	<i>Arctotheca calendula</i> *	flowers	SA, VIC	Mattingley (1909), Dubas and Bull (1991)
Asteraceae	<i>Craspedia</i> sp.		NSW	Henle (1990)
Asteraceae	<i>Pallenis spinosa</i> *	flowers	SA	Dubas and Bull (1991)
Asteraceae	<i>Polycalymma stuartii</i>		NSW	Henle (1990)
Asteraceae	<i>Reichardia tingitana</i> *	flowers	SA	Dubas and Bull (1991)
Asteraceae	<i>Senecio lautus</i>	flowers	SA	Yeatman (1988)
Asteraceae	<i>Vittadinia gracilis</i>	leaves	SA	Norval et al. (2018)
Boraginaceae	<i>Echium plantagineum</i> *		NSW	Henle (1990)
Brassicaceae	<i>Brassica tournefortii</i> *	leaves	NSW	Henle (1990)
Brassicaceae	<i>Cakile</i> sp. *	leaves	SA	Yeatman (1988)
Brassicaceae	<i>Cakile maritima</i> *	flowers and leaves	SA	Dubas and Bull (1991)

Table 1.2. continued

Brassicaceae	<i>Carrichtera annua</i>	flowers, leaves and fruit	SA	Spiegel et al. (2015)
Chenopodiaceae	<i>Enchylaena tomentosa</i>	fruit	SA, NSW	Henle (1990), Dubas and Bull (1991)
Convolvulaceae	<i>Convolvulus remotus</i>	flowers and leaves	SA	Norval et al. (2018)
Dioscoreaceae	<i>Dioscorea hastifolia</i>	leaves	WA	Brooker (2016)
Ericaceae	<i>Astroloma macrocalyx</i>	fruit	WA	Keighery (1984)
Ericaceae	<i>Leucopogon parviflorus</i>	fruit	SA	Yeatman (1988)
Euphorbiaceae	<i>Euphorbia paralias</i> *	flowers	SA	Dubas and Bull (1991)
Fabaceae	<i>Medicago minima</i> *	flowers and leaves	SA	Norval et al. (2018)
Fabaceae	<i>Medicago</i> sp. *	flowers and leaves	SA	Dubas and Bull (1991)
Goodeniaceae	<i>Goodenia pinnatifida</i>	flowers	SA	Dubas and Bull (1991)
Iridaceae	<i>Moraea setifolia</i> *	flowers	SA	Dubas and Bull (1991)
Iridaceae	<i>Patersonia</i> sp.	flower	WA	Shugg (1983)
Lamiaceae	<i>Salvia verbenaca</i> *	flowers and leaves	SA	Norval et al. (2018)
Lamiaceae	<i>Teucrium sessiliflorum</i>	flowers	SA	Dubas and Bull (1991)
Malvaceae	<i>Sida intricata</i>	leaves	SA	Norval et al. (2018)
Nitrariaceae	<i>Nitraria billardierei</i>	fruit	SA	Dubas and Bull (1991)
Orchidaceae	<i>Dipodium punctatum</i>	flowers	VIC	Turner and Doery (1981)
Oxalidaceae	<i>Oxalis corniculata</i> *	leaves	SA	Dubas and Bull (1991)

Table 1.2. continued

Phormiaceae	<i>Dianella revoluta</i>	fruit	SA	Dubas and Bull (1991)
Plantaginaceae	<i>Plantago drummondii</i>		NSW	Henle (1990)
Plumbaginaceae	<i>Limonium ramosissimum</i> *	flowers	SA	Dubas and Bull (1991)
Polygonaceae	<i>Muehlenbeckia</i> sp.	fruit	SA	Yeatman (1988)
Primulaceae	<i>Anagallis arvensis</i> *	flowers	SA	Dubas and Bull (1991)
Scrophulariaceae	<i>Eremophila glabra</i>	flowers	SA	Dubas and Bull (1991)
Scrophulariaceae	<i>Eremophila longifolia</i>	flowers	SA	Norval et al. (2018)
Scrophulariaceae	<i>Myoporum insulare</i>	fruit	WA	Tubb (1938)
Scrophulariaceae	<i>Myoporum platycarpum</i>	flowers	SA	Norval et al. (2018)
Solanaceae	<i>Lycium ferocissimum</i> *	flowers and fruit	SA	Cole (1930), Satrawaha and Bull (1981), Dubas and Bull (1991)
Solanaceae	<i>Solanum nigrum</i> *		NSW	Henle (1990)

Reproduction

Tiliqua rugosa is viviparous, and produce one to three neonates, which are born in late autumn after a gestation period of ca. five months (Bull et al. 1993b, Munns 2013). There is no direct parental care, but through long-term research it has been determined that the female can recognize her offspring via chemical signals and they are tolerated and remain within the mother's home range in their first year, after which they disperse (Bull et al. 1994, Main and Bull 1996, Bull and Baghurst 1998). *Tiliqua rugosa* young grow fairly rapidly (Bull 1987). They reaching sexual maturity within three to four years (Bull 1987), which is longer than most skink species listed in Greer (1989). However, species such as *Egernia cunninghami* and *Egernia rugosa*, which are related to *T. rugosa*, can take up to 5 years to reach sexual maturity but have life spans that are almost half of that of sleepy lizards (Chapple 2003, Peck et al. 2016). The age at which sleepy lizards reach sexual maturity is therefore quick for such a long-lived species. Mortality rates are high among juveniles, most likely as a result of predation, parasitism and unfavourable environmental conditions, so only a few survive to adulthood (e.g. ca. 4% at the site near Mt. Mary), but adults have lower mortality rates and are estimated to live for ca. 50 years (Bull 1995, Jones et al. 2016).

Prior to the long-term research into the natural history of *T. rugosa* it was noted that in spring these lizards walk in tandem (Fig. 1.12), and it was assumed that the male was attending and guarding the female (Bull 1987), however the long-term data collection revealed that this simple explanation was an understatement of a behaviour that is extremely rare in lizards. Sleepy lizards are usually solitary, but during the mating season (September to December) they form pairs (Bull 1988). The onset of the pairing period is determined by warm temperatures, but mating takes place in late October to early November, so the pairing period may vary (Bull and Burzacott 2002, Brooker 2016). Although some extra-pair fertilization may take place (Bull et al. 1998), the majority of sleepy lizard pairs are both socially and genetically monogamous (Leu et al. 2015). These pair bonds can be extremely stable, lasting multiple (e.g. one pair was recorded for 27 consecutive years in 2012) mating seasons (Leu et al. 2015), and can be so strong that males will remain with their partners even after the female died (Sharrad et al. 1995, How and Bull 1998, Kerr and Bull 2001). The partners are not constantly together during the pairing period, and separations, which may range in time from a few hours to several days, may take place (Bull 1988, How and Bull 2002, Leu et al. 2011b). It has been noted that male and female *T. rugosa* exhibit various personalities (i.e. behavioural types) and that

aggressive males have weaker pairing bonds with females than do less aggressive males (Godfrey et al. 2012). By observing individuals over consecutive weeks, it was found that partners usually search for each other to re-establish the bonds and the reunion intensity increases later in the pairing period prior to the short period when females are receptive (Bull 1988, Bull et al. 1993a, How and Bull 2002, Leu et al. 2011b). Although both genders may re-establish the bond, it is more frequently initiated by the males (Leu et al. 2011b).



Figure 1.12. During the mating season sleepy lizards (*Tiliqua rugosa*) can often be found in pairs, usually with the female walking in the front and the male trailing behind her.

Mate-guarding either entails a male guarding a female against rival males because he is unable to defend the female's home range (Cuadrado 2001), or in the case of non-territorial species, to ensure paternity (Marco and Pérez-Mellado 1999). Earlier studies into the reason for the pairing behaviour of sleepy lizards disproved mate-guarding as an explanation (How and Bull 2002). Priming, a form of female coercion in which the female can only become receptive after repeated courtship, was proposed as a likely alternative explanation (How and Bull 2002). An evaluation of data from multiple years supports the assumption that it is a form of female coercion (Bull 2000). The female benefits energetically since there are indications that the males will defend the female from rival males (Bull and Pamula 1996, Kerr and Bull 2002, Murray and Bull 2004) and act as a sentinel for potential threats (Bull and Pamula 1998), both which permits the attended female to feed more effectively. Males benefit genetically from the pairing because males that remain monogamous and attend their regular partner usually father

all her offspring (Bull et al. 1998). Mating takes place in late October to early November, and familiar pairs tend to mate sooner than unfamiliar pairs (Leu et al. 2015), after which the pair separates (Bull and Burzacott 2002), but remain in social contact by occupying overlapping core areas of their home ranges and sharing refuges (Leu et al. 2011b).

Competition

Members of a particular species compete with other individuals of the same species (intra-species competition) as well as with those of other species (inter-species competition) for resources within an ecosystem. Inter-species competition can have significant influences on populations and communities (Sih et al. 1985). Considering the spatial use of shelters by sleepy lizards and the fact that they are primarily herbivorous, makes competition among them and a variety of other taxa likely. However, this aspect of the natural history of sleepy lizards has received relatively little attention. Climate profiles suggest that *T. rugosa* and *Tiliqua scincoides* have very similar temperature requirements but the former is more adapted for drier conditions (Hancock and Thompson 1997). A study that examined the resource partitioning by *T. rugosa* and two other congeneric blue-tongue species, *Tiliqua nigrolutea* and *T. scincoides*, at a locality in Cape Jaffa in South Australia found that there are substantial overlaps among the species with regards to their utilisation of heat, space, cover, time and food resources (Yeatman 1988). Still, it was noted that sleepy lizards consume more plant material than the other two blue-tongue species, and that *T. rugosa* more frequently utilise open habitats than *T. nigrolutea* and *T. scincoides* do (Yeatman 1988). Another study compared the thermal ecology and diet of several xerophilous lizards, and found that the surface activity of *T. rugosa* is more restricted by temperature extremes than *Pogona vitticeps* is, and that the former are more generalists in what they eat than the latter are (MacMillen et al. 1989).

Sleepy lizards tend to flourish in some urban localities, such as in Perth, while in others their occurrence is very limited, even when related blue-tongue species do occur. The role that competition and habitat requirements plays in the distribution of sleepy lizards require further study.

Predators

As in competition, predation can have significant influences on communities and therefore ecosystems (Sih et al. 1985). Known and possible predators of sleepy lizards include aboriginal

people, dingos (*Canis lupus dingo*), feral cats (*Felis catus*), foxes (*Vulpes vulpes*), carpet pythons (*Morelia spilota*), dugites (*Pseudonaja affinis*), peninsula brown snakes (*Pseudonaja inframacula*), western brown snakes (*Pseudonaja nuchalis*), common brown snakes (*Pseudonaja textilis*) (Fig. 1.13), and wedge-tailed eagles (*Aquila audax*) (Shine 1989, Roberts and Mirtschin 1991, Bull and Pamula 1998, Metcalfe and Driver 2008, Norval et al. 2018). However, the impact of these predators on sleepy lizard populations has not been studied.



Figure 1.13. As in most other species, sleepy lizards (*Tiliqua rugosa*) are more at risk to predation while they are immature, such as this juvenile that was caught by a common eastern brown snake (*Pseudonaja textilis*).

Parasites and Pathogens

The long-term research on *T. rugosa* in South Australia did not start out as an investigation into the natural history of these lizards, but rather as studies into explaining the parapatric boundaries of the reptile ticks, *Amblyomma albolimbatum*, *Amblyomma limbatum* and *Bothriocroton hydrosauri*, that often infest them. All three species parasitize a variety of reptile hosts, but are most commonly found on *T. rugosa* (Andrews and Petney 1981, Sharrad and King 1981). In addition to these ticks *T. rugosa* has been reported as a host of a variety of other parasites (Table 1.3), some of which have only been recorded from *T. rugosa*, while others are known to parasitize a variety of hosts.

Parasites can negatively influence host survival and or fecundity and therefore directly or indirectly play important roles in host population dynamics (Scott and Dobson 1989, Tompkins and Begon 1999, Valkiūnas 2001). Long-term research on the ticks *A. limbatum* and *B. hydrosauri* and their infestation of sleepy lizards initially suggested that tick infestation intensity (i.e. the number of ticks that infest the host) do not adversely affect survival, growth

Table 1.3. Organisms that have been reported as parasites of *Tiliqua rugosa*.

Class	Species	Reference
Gammaproteobacteria	<i>Escherichia coli</i>	Gordon and Cowling (2003)
	<i>Salmonella enterica</i>	Bull et al. (2012)
Ciliata	<i>Nyctotherus trachysauri</i>	Johnston (1932)
Coccidia	<i>Eimeria</i> sp.	O'Donoghue (1998)
	<i>Eimeria tiliquae</i>	Yang et al. (2013)
	<i>Hemolivia mariae</i>	Smallridge and Bull (2000)
	<i>Schellackia</i> sp.	O'Donoghue (1998)
Flagellata	<i>Bodo</i> sp.	Johnston (1932)
	<i>Copromonas</i> sp.	Johnston (1932)
	<i>Trichomastix</i> or <i>Trichomonas</i> sp.	Johnston (1932)
	<i>Endamoeba</i> sp.	Johnston (1932)
Cestoda	<i>Oochoristica trachysauri</i>	MacCallum (1921)
Nematoda	<i>Abbreviata antarctica</i>	Jones (1992)
	<i>Oxyuris</i> sp.	Thapar (1925)
	<i>Thelandros trachysauri</i>	Johnston and Mawson (1947)
	<i>Veversia tuberculata</i>	von Linstow (1904)
Trematoda	<i>Brachylaima cribbi</i>	Butcher and Grove (2005)
	<i>Microphallus</i> sp.	Angel and Mawson (1968)
	<i>Paradistomum crucifer</i>	MacCallum (1921)
Arachnida	<i>Amblyomma albolimbatum</i>	Neumann (1907)
	<i>Amblyomma limbatum</i>	Sharrad and King (1981)
	<i>Amblyomma moreliae</i>	Roberts (1964)
	<i>Amblyomma triguttatum</i> <i>triguttatum</i>	Petney et al. (2008)
	<i>Amblyomma vikirri</i>	Keirans et al. (1996)
	<i>Bothriocroton hydrosauri</i>	Ferguson (1925)
	<i>Ornithodoros gurneyi</i>	Sharrad and King (1981)

or reproduction in *T. rugosa* (Bull and Burzacott 1993), but the ticks reduce the running speed of juvenile lizards and decrease the general activity levels and the distances that sub-adult and adult lizards travel each day (Main and Bull 2000). Subsequent analysis of the long-term collected data found that infestation by *B. hydrosauri* reduces lizard survival (Jones et al. 2016). Through experimental increases of tick infestation it was also found that high tick loads have a negative influence on the retention of long-term partnerships in *T. rugosa*, most likely by impeding the activity and endurance of the male and thus his ability to attend the female (Bull and Burzacott 2006). The ticks were also identified as vectors for the haemogregarine blood parasite, *Hemolivia mariae* (Smallridge and Bull 1999), which also inhibits the activity of infected lizards (Bouma et al. 2007), which has been suggested to negatively impact the survival of juvenile lizards (Jones et al. 2016).

The long-term research into the social networks of sleepy lizards at the site near Mt. Mary also provided some insight into the transmission of pathogens. It was determined that the asynchronous use of refuges aligns with the life history traits of the tick *A. limbatum* and therefore facilitates the transmission of the tick through the sleepy lizard population (Leu et al. 2010b), and that personalities of the lizards, their social networks, and movement ecology, determined the infestation intensity by the tick *B. hydrosauri*, which is dispersed by the movements of sleepy lizards (Bull 1978, Sih et al. 2018).

Studies were not only limited to sleepy lizards and their ticks. It was also determined that the sleepy lizards in this population are often infected with up to three of the *Salmonella enterica* subspecies (*Salmonella enterica* subsp. *diarizonae*, *S. enterica* subsp. *enterica*, *S. enterica* subsp. *houtenae*, and *S. enterica* subsp. *salamae*) known to infect these lizards in this locality (Bull et al. 2012). Additionally the physical characteristics of their home ranges and their social networks were found to influence which *S. enterica* subspecies they were infected with (Bull et al. 2012, Parsons et al. 2015).

Apart from the results of these studies little is known about the influences of the various parasites on *T. rugosa*. As illustrated by the recently identified nidovirus, which causes a respiratory disease in these lizards, and often results in mortality if left untreated (O’Dea et al. 2016), our understanding of the parasites and pathogens in sleepy lizards is incomplete and warrant additional empirical investigations.

Conservation

Sleepy lizards are often attracted to roads by roadside vegetation and puddles that form on road surfaces during rains (e.g. Sadleir 1958). Due to their slow movement and tendency to stop moving when approached, they are often killed (Fig. 1.14) by passing vehicles (Henle 1990, Bull 1995, Sharrad et al. 1995, Brooker 2016). Eliminating this cause of mortality is likely impossible. A more realistic approach would be to determine which areas have higher numbers of lizards and to take intervention measures in these localities. Investigations into the feasibility of intervention measures such as the construction of fauna underpasses (e.g. Chambers and Bencini 2015) and/or the erection of signs, appealing to drivers to be mindful of lizards crossing the roads (as done for other wildlife), in combination with low barriers, in areas where lizards and other wildlife frequently cross roads are warranted. Studies should also look into the effectiveness of other possible intervention measures, such as the creation of drinking puddles and feeding and basking areas away from roads, which will attract lizards away from roads.



Figure 1.14. Some sleepy lizards (*Tiliqua rugosa*) are accidentally killed as they cross roads. This one however was killed at the side of the road, indicating the driver most likely intentionally aimed for it.

Other anthropogenic activities, such as the erection of fences, can also cause mortality in sleepy lizard populations (e.g. Waite 1925, How and Bull 1998, Ferronato et al. 2014). Possible changes in such structures, to minimize mortality in non-target species should be explored. Urban development causes habitat destruction, one of the most serious threats to wildlife.

Tiliqua rugosa is a fairly common species throughout its range in suitable habitats, and since this species can be human-commensal under some conditions it can even sometimes occur in suburban areas (Shea 1998). Its abundance and current conservation status does not mean that conservation actions are not necessary. Actions should be taken before a species becomes threatened, not when it is endangered. There is therefore a need to determine under what conditions this species can persist in suburban areas. Such information could aid in designing urban and suburban developments that are more lizard-friendly, which could lessen the impact of anthropogenic activities on this and other related species.

Conclusion

Tiliqua rugosa has been studied more extensively than most lizard species in Australia, but the impressive body of information that this work has produced still does not provide a complete understanding of its natural history and ecology. Most of the studies on this species were conducted in South Australia, and more specifically near Mt. Mary in the Mid North of South Australia. The species *T. rugosa* consists of four subspecies, *Tiliqua rugosa asper* Gray 1845 (eastern Australia), *Tiliqua rugosa konowi* Mertens 1958 (Rottnest Island, Western Australia), *Tiliqua rugosa rugosa* Gray 1825 (south-western Australia), and *Tiliqua rugosa palarra* Shea 2000 (mid-west coast of Western Australia), and most of what is known of this species is from the long-term research on the subspecies *T. r. rugosa* from a relatively small part of its distribution range. As indicated throughout this review, various aspects of the natural history warrant additional long and short-term empirical studies to fill the gaps in our understanding of the ecology of this species throughout its distribution range. In localities other than the site near Mt. Mary, the methodology of past studies can be replicated for comparison purposes. Such information would highlight likely regional variations in the ecology of sleepy lizards, which could be crucial for possible management and conservation actions.

The species also provides an excellent model to examine major evolutionary innovations such as the evolution of monogamy, viviparity and adaptations to differing environmental conditions. The solid foundation laid down by the late Michael Bull will undoubtedly be used as a platform for exploring these innovations using new genomic, statistical and other similarly powerful techniques. A truly lasting legacy.

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**Taking stock: a review of the known parasites of the sleepy lizard,
Tiliqua rugosa (Gray, 1825), a common lizard endemic to
Australia**

Parasitism is a very common life strategy and although it results in harm to the host, it plays a vital ecological role in host population and community dynamics. The sleepy lizard, *Tiliqua rugosa* (Gray, 1825), is one of the best studied lizard species in Australia, to a large extent due to studies involving ticks that infest these lizards. In spite of this, little is known about most of the parasites that are known to infect sleepy lizards. The purpose of this review is to provide a synopsis of the species that have been reported as parasites of *T. rugosa* as a foundation for future studies.

Introduction

Parasitism is one of the most common life strategies and consists of one organism, the parasite, which obtains sustenance, and often a living environment, from another living organism or organisms, and thus survives at the expense of the hosts (Fripp 1979, Hudson 2005, Roberts and Janovy 2005). By doing so, all parasites harm the host due to the damage caused to the tissue of the host as they bore through it or feed on it; by stimulating damaging inflammatory and/or immune responses in a host; by robbing the host of its nutrition; and/or by manipulating the host's natural behaviour (Lapage 1963, Poulin et al. 1994, Casadevall and Pirofski 2003, Roberts and Janovy 2005, Poulin 2013).

Although parasitic infections have a negative effect on their hosts, ecologically parasites are very important, because parasites can regulate host populations by often negatively influencing host survival and/or fecundity (Scott and Dobson 1989, Tompkins and Begon 1999, Valkiūnas 2001, Dobson et al. 2008). By doing so, parasites play important role in host population dynamics and influence communities by altering interspecific competition (Hatcher et al. 2008). Parasites sometimes infect more than one host species, and their life cycle may involve host species at different trophic levels. This attribute means parasites can have an influence on energy flow in food chains and ecosystems by influencing prey (the intermediate host of the parasite) in such a way that they are more accessible to predators (the definitive host of the parasite) (Lafferty 1992, Hudson et al. 2006).

In spite of the ecological value of parasites, they tend to be relatively poorly studied organisms and in many instances, even basic information such as parasite-host lists are incomplete. Most studies of parasites tend to be focussed on species that are of economic or human health importance. However, since studies into parasites and their natural history can have far-reaching implications. For example, since parasites have shorter life cycles than their hosts, they tend to evolve faster and can thus be inferential tools for the study of the evolution of their hosts (Whiteman and Parker 2005). Studies into parasite infections in wildlife and their roles in ecosystems can also be used for proactive measures in managing existing and emerging diseases (Spratt 2005, Brooks et al. 2014).

Despite parasite infections often being able to be confirmed via non-invasive methods, such as the examination of faecal samples or some molecular techniques, parasitological studies

usually entail the sacrifice of some host specimens. In addition, studying the natural history of endoparasites often means it is necessary to determine which organs they infect. As a result, common species, rather than rare species, are usually used in parasitological studies.

The sleepy lizard

The sleepy lizard, *Tiliqua rugosa* (Gray 1825), formerly classified as *Trachydosaurus rugosus*, is a relatively large (total length ca. 40 cm) skink species (Gray 1825, Shea 1990, Bull and Pamula 1996, Cogger 2014), endemic to the semiarid parts of southern and eastern Australia (Cogger 2014, Parsons et al. 2015). Throughout its distribution range this species is usually fairly common, so sleepy lizards have been used in numerous parasitological studies. The purpose of this review is to provide a synopsis of the species that have been reported as parasites of these lizards as a foundation for future studies.

Ticks

Tiliqua rugosa is one of the best studied lizard species in Australia, primarily as a result of long-term studies that were initiated by the late Professor C. Michael Bull in South Australia into the parapatric boundary of hard ticks (Family: Ixodidae) *Amblyomma limbatum* Neumann, 1899, and *Bothriocroton hydrosauri* (Denny, 1843) that infest these lizards (Godfrey and Gardner 2017). Ticks are very common ectoparasites of most vertebrates, so it is not surprising that in addition to the above-mentioned hard ticks, an additional four species of hard ticks and one species of soft tick (Family: Argasidae) have been recorded as parasites of sleepy lizards.

Five of the recorded hard tick species, *Amblyomma albolimbatum* Neumann, 1907, *A. limbatum*, *Amblyomma moreliae* (Koch, 1867), *Amblyomma vikirri* Keirans, Bull and Duffield, 1996, and *B. hydrosauri*, mainly parasitize reptiles (Table 1.4), although *A. moreliae* and *B. hydrosauri* have also been reported parasitizing some mammals (Roberts 1964a, 1970, Keirans et al. 1996). The sixth hard tick, *Amblyomma triguttatum triguttatum* Koch, 1844 primarily infests mammals, but a female of this tick has been collected from *T. rugosa* in the wild (Roberts 1970, Waudby et al. 2007, Petney et al. 2008). However, since no additional samples were obtained, this was very likely an instance of an accidental choice of host (Petney et al. 2008). The soft tick *Ornithodoros gurneyi* Warburton, 1926, is a relatively poorly studied species, but it is believed to predominantly infest mammals and will only opportunistically parasitize lizards such as *Pogona barbata* and *T. rugosa* (Doubé 1975, Sharrad and King 1981,

Barker and Walker 2014).

All of the above-mentioned tick species are three-host ticks, which means the different life stages detach to moult and then have to find a new host (Barker and Walker 2014). The larvae, nymphs and adults of the hard ticks that infest sleepy lizards can sometimes be found on a single host (Roberts 1964a, 1969, 1970, Keirans et al. 1996). As for the soft tick, little information is available concerning the behaviour of the different life stages of *O. gurneyi* on hosts under natural conditions (Barker and Walker 2014), and the records of these ticks parasitizing *T. rugosa* did not state the life stages (Sharrad and King 1981).

All ticks are obligate haematophagous arthropods, and in addition to anemia they may also cause other diseases (e.g. dermatosis, tick paralysis and/or otoacariasis) in their hosts (Lapage 1963, Roberts and Janovy 2005). Most studies of the effects of ticks on their hosts tend to focus on species that are of human health and/or economical (i.e. agricultural and veterinary) importance. As for the species that infest sleepy lizards, only the impacts of *A. limbatum* and *B. hydrosauri* (Fig. 1.15) on these lizards have been studied. It was found that infestation by these ticks does not cause mortality in sleepy lizards (Bull and Burzacott 1993), but they reduce the activity of infested lizards (Main and Bull 2000). In male *T. rugosa* this may inhibit his capability to attend the female and thus impede his ability to retain a long-term partnership (Bull and Burzacott 2006). It would be interesting to know what impacts the other tick species have on *T. rugosa* and other hosts, so additional research is encouraged.



Figure 1.15. In some localities sleepy lizard (*Tiliqua rugosa*) can be infested by more than one species of tick; this one is being parasitised by the males of *Amblyomma limbatum* (red arrow) and *Bothriocroton hydrosauri* (blue arrows).

Table 1.4. Tick species that have been reported to infest *Tiliqua rugosa*, and the other reptiles they have been recorded from in Australia. (WA – Western Australia; NT – Northern Territory; QLD – Queensland; NSW – New South Wales; VIC – Victoria; SA – South Australia; TAS – Tasmania)

Species	Host type	Hosts	Locality	Reference
<i>Amblyomma albolimbatum</i>	Chelonian	<i>Pseudemydura umbrina</i>	WA, SA, VIC	Roberts (1964a)
	Lizard	<i>Egernia multiscutata</i>	WA	Sharrad and King (1981)
		<i>Egernia stokesii</i>	WA	Sharrad and King (1981)
		<i>Tiliqua multifasciata</i>	WA	Sharrad and King (1981)
		<i>Tiliqua occipitalis</i>	WA, SA, VIC	Sharrad and King (1981)
		<i>Tiliqua rugosa</i>	WA, SA, VIC	Robinson (1926), Roberts (1964a, 1969)
	Snake	<i>Morelia spilota</i>	WA, SA, VIC	Roberts (1964a)
		<i>Notechis scutatus</i>	WA, SA, VIC	Ferguson (1925), Roberts (1964a)
		<i>Pseudonaja affinis</i>	WA	Sharrad and King (1981)
		<i>Pseudonaja textilis</i>	WA, SA, VIC	Roberts (1964a)
	<i>Amblyomma limbatum</i>	Lizard	<i>Ctenophorus decresii</i>	SA
<i>Ctenophorus fionni</i>			SA	Sharrad (1979)
<i>Pogona vitticeps</i>			SA	Chilton et al. (1992)
<i>Tiliqua multifasciata</i>			WA	Sharrad and King (1981)
<i>Tiliqua rugosa</i>			WA, SA, VIC, NT, TAS	Robinson (1926), Roberts (1964a, 1969)

Table 1.4. continued

	<i>Tiliqua scincoides</i>	WA	Sharrad and King (1981)
	<i>Underwoodisaurus mii</i>	SA	Sharrad (1979)
	<i>Varanus acanthurus</i>	WA	Roberts (1964a)
	<i>Varanus glebopalma</i>	WA	Sharrad and King (1981)
	<i>Varanus gouldii</i>	WA	Roberts (1964a)
	<i>Varanus mitchelli</i>	WA	Sharrad and King (1981)
	<i>Varanus panoptes</i>	WA	Sharrad and King (1981)
	<i>Varanus sp.</i>	WA, SA, VIC, NT, TAS	Roberts (1964a)
	<i>Varanus timorensis</i>	WA	Sharrad and King (1981)
	<i>Varanus tristis</i>	WA	Sharrad and King (1981)
Snake	<i>Acanthophis praelongus</i>	WA	Sharrad and King (1981)
	<i>Aspidites melanocephalus</i>	WA	Sharrad and King (1981)
	<i>Aspidites ramsayi</i>	WA, SA, VIC, NT, TAS	Roberts (1964a)
	<i>Boiga irregularis</i>	WA	Sharrad and King (1981)
	<i>Dendrelaphis punctulatus</i>	WA	Sharrad and King (1981)
	<i>Liasis childreni</i>	WA	Sharrad and King (1981)
	<i>Liasis olivaceus</i>	WA	Sharrad and King (1981)
	<i>Morelia spilota</i>	WA	Robinson (1926)
	<i>Pseudechis australis</i>	WA	Roberts (1964a)
	<i>Pseudechis porphyriacus</i>	WA, SA, VIC, NT, TAS	Roberts (1964a)

Table 1.4. continued

<i>Amblyomma moreliae</i>	Lizard	<i>Egernia</i> sp.	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Egernia striolata</i>	NSW, VIC, QLD, NT	Roberts (1970)
		<i>Physignathus lesueurii</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Tiliqua nigrolutea</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Tiliqua rugosa</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Tiliqua scincoides</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Varanus</i> sp.	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Varanus tristis orientalis</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Varanus varius</i>	NSW, VIC, QLD, NT	Roberts (1964a)
	Snake	<i>Aspidites melanocephalus</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Dendrelaphis calligaster</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Morelia spilota</i>	NSW, VIC, QLD, NT	Koch (1867)
		<i>Morelia spilota variegata</i>	NSW, VIC, QLD, NT	Roberts (1964a)
		<i>Notechis scutatus</i>	NSW, VIC, QLD, NT	Roberts (1964a)
	<i>Oxyuranus scutellatus</i>	NSW, VIC, QLD, NT	Roberts (1964a)	
	<i>Pseudechis australis</i>	NSW, VIC, QLD, NT	Roberts (1970)	
	<i>Pseudechis porphyriacus</i>	NSW, VIC, QLD, NT	Roberts (1964a, 1969)	
	<i>Pseudonaja textilis</i>	NSW, VIC, QLD, NT	Roberts (1964a)	
<i>Amblyomma triguttatum triguttatum</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Petney et al. (2008)
<i>Amblyomma vikirri</i>	Lizard	<i>Egernia stokesii</i>	SA	Keirans et al. (1996)

Table 1.4. continued

		<i>Tiliqua rugosa</i>	SA	Keirans et al. (1996)
<i>Bothriocroton hydrosauri</i>	Chelonian	<i>Chelodina longicollis</i>	SA, NSW, TAS	Roberts (1964b, 1970)
	Lizard	<i>Liopholis whitii</i>	SA	Smyth (1973)
		<i>Niveoscincus metallicus</i>	SA	Smyth (1973)
		<i>Pogona barbata</i>	SA, NSW	Roberts (1953)
		<i>Rankinia diemensis</i>	NSW, TAS	Roberts (1964b, 1970)
		<i>Tiliqua adelaidensis</i>	SA	Fenner and Bull (2007)
		<i>Tiliqua nigrolutea</i>	SA, NSW, VIC, QLD, TAS	Roberts (1964b, 1970), Shea (1983)
		<i>Tiliqua occipitalis</i>	SA	Smyth (1973)
		<i>Tiliqua rugosa</i>	WA, SA, TAS	Roberts (1953, 1964b), Sharrad and King (1981)
		<i>Tiliqua scincoides</i>	SA, NSW, TAS	Roberts (1964a, 1964b)
		<i>Varanus gouldii</i>	TAS	Roberts (1964a)
	Snake	<i>Acanthophis antarcticus</i>	SA	Smyth (1973)
		<i>Austrelaps superbus</i>	TAS	Roberts (1953, 1964a, 1964b), Smyth (1973)
		<i>Drysdalia coronoides</i>	TAS	Roberts (1970)
		<i>Notechis</i> sp.	SA	Smyth (1973)
		<i>Notechis scutatus</i>	TAS	Roberts (1953, 1964b)

Table 1.4. continued

		<i>Pseudonaja textilis</i>	SA	Roberts (1970), Smyth (1973)
<i>Ornithodoros gurneyi</i>	Lizard	<i>Pogona barbata</i>	–	Doube (1975)
		<i>Tiliqua rugosa</i>	WA	Sharrad and King (1981)

Ticks are extremely important as vectors for various bacterial, filarial, fungal, protozoan, rickettsial, spirochete, and viral pathogens (Lapage 1963, Hoogstraal 1985, Roberts and Janovy 2005). Studies of the ticks that parasitise *Tiliqua rugosa* have identified *A. limbatum* and *B. hydrosauri* as vectors for the haemogregarine parasite, *Hemolivia mariae* Smallridge and Paperna, 1997 (Smallridge and Paperna 1997). To the best of our knowledge no other tick-borne pathogens of *T. rugosa* have been reported, but it is highly likely that there are others. For example, while *B. hydrosauri* is most likely one of the best studied reptile ticks in the world (Barker and Walker 2014), only recently has an undescribed *Rickettsia* species been detected in *B. hydrosauri* on sleepy lizards (Whiley et al. 2016). Additionally, the bacterium *Borrelia queenslandica* has also been detected in *O. gurneyi* (Pope and Carley 1956, Carley and Pope 1962) but not from ticks taken from sleepy lizards. However, it is likely that they and/or other pathogens are present in *T. rugosa* and their tick parasites and that other tick-borne pathogens simply have not been detected yet. Studies into the potential pathogens harboured by the above mentioned ticks are therefore warranted.

Bacteria

Various species of bacteria have been reported as primary pathogens and secondary invaders in reptiles (Cooper 1981). However, the only bacteria to have been reported from *T. rugosa* to date are *Escherichia coli* (Migula 1895), *Salmonella* sp., and *Salmonella enterica* (ex Kauffmann and Edwards 1952) Le Minor and Popoff 1987 (Family: Enterobacteriaceae), which have also been recorded from various vertebrates worldwide, including several other reptile species from Australia (Table 1.5). It must be taken into consideration that although *E. coli* and *Salmonella* sp. have been implicated in mortality and diseases in reptiles, these organisms appear to be part of the normal gastrointestinal flora of many species, , there are non-pathogenic strains of both *E. coli* and *Salmonella* (Blount 2015, Public Health England 2015), or may only become pathogens when the immune system of the animal is compromised by stress and/or other diseases (Hoff and Hoff 1984, Jacobson 2007). Although neither of these organisms appear to have a negative influence on *T. rugosa* in the wild, their role in the ecology of this lizard is not fully understood and requires additional research.

Protozoans

As with most other parasites, under natural conditions almost all reptile species harbour some protozoan species, although not all are necessarily pathogenic (Keymer 1981). Still, in spite of

Table 1.5. Bacteria species that have been reported to infect *Tiliqua rugosa*, and the other reptiles they have been recorded from in Australia. (WA – Western Australia; QLD – Queensland; NSW – New South Wales; VIC – Victoria; SA – South Australia)

Species	Host type	Hosts	Locality	Reference
<i>Escherichia coli</i>	Chelonian	<i>Elseya dentata</i>	–	Gordon and Cowling (2003)
	Crocodilians	<i>Crocodylus johnstoni</i>	–	Gordon and Cowling (2003)
		<i>Crocodylus porosus</i>	–	Gordon and Cowling (2003)
	Lizards	<i>Egernia saxatilis</i>	–	Gordon and Cowling (2003)
		<i>Lampropholis delicata</i>	NSW	Gordon and Cowling (2003)
		<i>Lampropholis guichenoti</i>	NSW	Gordon and Cowling (2003)
			<i>Tiliqua rugosa</i>	–
<i>Salmonella</i> sp.	Chelolians	<i>Chelodina oblonga</i>	WA	Iveson et al. (1969)
		<i>Emydura victoriae</i>	WA	Iveson et al. (1969)
	Crocodilian	<i>Crocodylus porosus</i>	WA, QLD	Iveson et al. (1969), Thomas et al. (2001)
	Lizards	<i>Chlamydosaurus kingii</i>	WA	Lee and Mackerras (1955)
		<i>Ctenophorus ornatus</i>	WA	Iveson et al. (1969)
		<i>Cyclodomorphus branchialis</i>	WA	Iveson et al. (1969)
		<i>Diporiphora bilineata</i>	WA	Iveson et al. (1969)
		<i>Intellagama lesueurii</i>	WA, QLD	Lee and Mackerras (1955), Iveson et al. (1969)
	<i>Lialis burtonis</i>	WA	Iveson et al. (1969)	

Table 1.5. continued

	<i>Moloch horridus</i>	WA	Iveson et al. (1969)
	<i>Pogona barbata</i>	WA, VIC, NSW	Lee and Mackerras (1955), Iveson et al. (1969)
	<i>Tiliqua multifasciata</i>	WA	Iveson et al. (1969)
	<i>Tiliqua occipitalis</i>	WA	Iveson et al. (1969)
	<i>Tiliqua rugosa</i>	WA, VIC	Iveson et al. (1969)
	<i>Tiliqua scincoides</i>	WA, VIC, QLD	Iveson et al. (1969), Thomas et al. (2001)
	<i>Varanus tristis</i>	WA	Iveson et al. (1969)
	<i>Varanus varius</i>	SA, VIC, NSW, QLD	Iveson et al. (1969)
Snakes	<i>Antaresia childreni</i>	QLD	Iveson et al. (1969)
	<i>Aspidites melanocephalus</i>	WA	Iveson et al. (1969)
	<i>Austrelaps ramsayi</i>	VIC	Iveson et al. (1969)
	<i>Demansia olivacea</i>	VIC	Iveson et al. (1969)
	<i>Dendrelaphis punctulatus</i>	QLD	Iveson et al. (1969)
	<i>Morelia spilota spilota</i>	QLD	Iveson et al. (1969)
	<i>Morelia spilota variegata</i>	QLD	Iveson et al. (1969)
	<i>Notechis scutatus</i>	VIC	Iveson et al. (1969)
	<i>Oxyuranus microlepidotus</i>	QLD	Thomas et al. (2001)

Table 1.5. continued

		<i>Pseudonaja affinis</i>	WA	Iveson et al. (1969)
		<i>Simalia amethystina</i>	QLD	Thomas et al. (2001)
<i>Salmonella enterica</i> subsp. <i>diarizonae</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Parsons et al. (2015)
<i>Salmonella enterica</i> subsp. <i>enetricea</i>	Crocodilian	<i>Crocodylus porosus</i>	–	Parsons et al. (2011)
	Lizard	<i>Ctenophorus isolepis</i>	–	Parsons et al. (2011)
		<i>Ctenophorus reticulatus</i>	–	Parsons et al. (2011)
		<i>Ctenotus inornatus</i>	–	Parsons et al. (2011)
		<i>Ctenotus pantherinus</i>	–	Parsons et al. (2011)
		<i>Delma pax</i>	–	Parsons et al. (2011)
		<i>Diporiphora winneckeii</i>	–	Parsons et al. (2011)
		<i>Eremiascincus fasciolatus</i>	–	Parsons et al. (2011)
		<i>Liopholis whitii</i>	–	Parsons et al. (2011)
		<i>Pogona minor</i>	–	Parsons et al. (2011)
		<i>Pseudemoia entrecasteauxii</i>	–	Parsons et al. (2011)
		<i>Tiliqua multifasciata</i>	–	Parsons et al. (2011)
		<i>Tiliqua rugosa</i>	SA	Parsons et al. (2011, 2015)
		<i>Varanus acanthurus</i>	–	Parsons et al. (2011)
		<i>Varanus brevicaudus</i>	–	Parsons et al. (2011)
		<i>Varanus gouldii</i>	–	Parsons et al. (2011)
<i>Salmonella enterica</i> subsp. <i>houtenae</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Parsons et al. (2015)

Table 1.5. continued

<i>Salmonella enterica</i> subsp. <i>salamae</i>	Lizard	<i>Tiliqua rogosa</i>	SA	Parsons et al. (2015)
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their size, some pathogenic protozoans can affect the host to such an extent that they control the host's populations (Bennett et al. 1976). As stated earlier, the ticks *A. limbatum* and *B. hydrosauri* are vectors for the coccidian protozoan (Family: Karyolysidae) *H. mariae*, and sleepy lizards become infected by this blood parasite when they ingest infected ticks, which in turn become infected if they feed as larvae or nymphs on infected lizards (Smallridge and Bull 1999, Smallridge and Bull 2001a).

Amblyomma limbatum larvae and nymphs are more susceptible to becoming infected by *H. mariae* than those of *B. hydrosauri*, and the former species is also more efficient at transmitting *H. mariae* to lizard hosts (Smallridge and Bull 1999). Although the prevalence of *H. mariae* in *A. limbatum* is fairly high, the infections do not appear to have negative effects on the survival of the ticks, who also do not avoid feeding on infected lizards (Smallridge and Bull 2001b). As for the lizards though, the infection prevalence and intensity are relatively low, and juveniles and sub-adults rarely become infected (Smallridge and Bull 2000), however infections may persist for several months, although usually at decreasing intensities (Smallridge and Bull 2001a). It was also found that individuals with larger home ranges were more likely to become infected, most probably due to a greater potential for encountering and ingesting infected ticks within the home range of other infected lizards (Bouma et al. 2007). But when a sleepy lizard becomes infected by *H. mariae* there is a reduction of the home range of the lizard (Bouma et al. 2007). It is not clear whether this home range reduction is due to changes in the fitness of the lizard, a defensive response of the host to prevent further infections, or a combination of the effect and response (Bouma et al. 2007). In an experimental infection of the lizards *Heremites vittatus* (reported as *Mabuya vittata*) and *Stellagama stellio* (reported as *Agama stellio*) with *H. mariae*, it was found that the developmental schedule of the parasite was retarded (Paperna and Smallridge 2001), which may suggest a degree of host specificity.

Another potentially host-specific protozoan parasite of sleepy lizards is *Eimeria tiliquae* Yang, Brice, Ryan and Bennett, 2013 (Family: Eimeriidae), which was found in the faeces of *Tiliqua rugosa* specimens from Western Australia (Yang et al. 2013). *Eimeria* infections are usually via the oral-faecal route, and although most of these protozoans are host specific, a particular host may be infected by multiple species (Rose 1985). In a survey of *T. rugosa* specimens that were admitted to a rehabilitation centre in Western Australia, it was found that about 21% were infected by *Eimeria* sp., and although some of the infected lizards had symptoms of upper

respiratory tract infections, none had signs of gastrointestinal infections (Yang et al. 2013).

The only other protozoan species recorded from sleepy lizards and identified to the species level is *Nyctotherus trachysauri* Johnston, 1932, (Family: Nyctotheridae), and like *E. tiliquae*, to date this protozoan has only been collected from *T. rugosa* (O'Donoghue and Adlard 2000). Johnston (1932) stated that these protozoans are relatively large, can be rare to common in the rectums of *T. rugosa*, and specimens of this protozoan remained alive for three days at room temperature in a container containing faeces in water.

Several other undetermined protozoan species have been reported from sleepy lizards. An undetermined species of *Schellackia* sp. (Family: Lankesterellidae) was recorded from two *T. rugosa* specimens from south-east Queensland (O'Donoghue 1998), while *Trichomonas* spp. (Family: Trichomonadidae) and *Balantidium* spp. (Family: Balantiididae) trophozoites were recorded from sleepy lizards from Western Australia (Yang et al. 2013). Johnston (1932) also recorded *Trichomonas* sp., in addition to *Bodo* sp. (Family: Bodonidae), *Copromonas* sp. (Family: Peranemataceae), and *Endamoeba* sp. (Family: Entamoebidae) from sleepy lizards from an unstated location.

Cestodes

Cestodes (tapeworms) are fairly common endoparasites of reptiles, and under some conditions can reach high infection intensities (Klingenberg 1993, Lane and Mader 1996). The only cestode reported from sleepy lizards to date is *Oochoristica trachysauri* (MacCallum, 1921) (Family: Listowiidae). This appears to be a common parasite of *T. rugosa* in South Australia, and the drier parts of New South Wales, Victoria, and southern Queensland (Johnston 1932). In a study by Gyawali et al. (2013) it was found that ca. 15% (19% of the males and 11.5% of the females) of the *T. rugosa* at Bunday Bore Station in Mid North District of South Australia were infected by these parasites. The parasite lives in the intestines of its host, and infection intensities can be so high that the lumen of the host's intestine is filled with the worms (MacCallum 1921, Johnston 1932). Apart from this, nothing else appears to be known about this tapeworm, and its impact on the host requires empirical study.

Nematodes

Nematodes are very common parasites and a great variety is known to infect reptiles

(Klingenberg 1993, Lane and Mader 1996). Nematodes from the families Oxyuridae and Pharyngodonidae have been reported as parasites of *T. rugosa*. Infection by species from both families is usually via an oral-fecal route (Anderson 2000), which is also most likely the way sleepy lizards are infected.

The only oxyurid species reported from *T. rugosa*, which has been described to the species level, is *V. tuberculata* (Linstow, 1904). Another oxyurid species, that could only be assigned to the common genus *Oxyuris*, due to the absence of male specimens, has also been reported from *T. rugosa* (Thapar 1925). *Veversia tuberculata* is reported to usually infect the large intestines of sleepy lizards (von Linstow 1904), and according to Johnston (1932) is fairly common in sleepy lizards from South Australia, Victoria, New South Wales, and southern Queensland.

Two species of pharyngodonid nematodes, *Abbreviata antarctica* (Linstow, 1899) and *Thelandros trachysauri* Johnston and Mawson, 1947, are known to infect *T. rugosa*, as well as a variety of other lizard and snake species from several states in Australia (Table 1.6). According to Angel and Mawson (1968), Jones (1992), and Gyawali et al. (2013) *Th. trachysauri* is a very common parasite of *T. rugosa*, but no other information appears to be available.

Trematodes

Trematodes (flukes) are also fairly common endoparasites of reptiles (Lane and Mader 1996), and three species of trematodes, *Brachylaima cribbi* Butcher, 2003 (Family: Brachylaimidae), *Paradistomum crucifer* (Nicoll, 1914) (Family: Dicrocoeliidae), and *Microphallus* sp. (Family: Microphallidae) have been reported to infect *T. rugosa* (Angel and Mawson 1968, Butcher and Grove 2005). Of these, the life cycle of *B. cribbi* appears to be the best studied (Butcher and Grove 2001, Butcher 2003, Butcher and Grove 2003, 2005), most likely since it can infect humans. A variety of terrestrial snail species have been recorded as first and second intermediate hosts of *B. cribbi*, and the known definitive hosts are primarily birds (Butcher and Grove 2003, 2005). Still, *T. rugosa* (i.e. the only reptile) and a few mammals, including *Homo sapiens*, have also been recorded as definitive hosts (Butcher and Grove 2005). Since the life cycle of species of *Brachylaima* usually entails infection of the same or different terrestrial snail species as the first and second intermediate hosts and either a bird or a mammal as the

Table 1.6. The helminth species that have been reported to infect *Tiliqua rugosa*, and the other reptiles they have been recorded from in Australia. (WA – Western Australia; NT – Northern Territory; QLD – Queensland; NSW – New South Wales; VIC – Victoria; SA – South Australia; TAS – Tasmania)

Species	Host type	Hosts	Locality	Reference
CESTODES				
<i>Oochoristica trachysauri</i>	Lizard	<i>Tiliqua rugosa</i>	SA, WA	MacCallum (1921), Johnston (1932)
NEMATODES				
<i>Abbreviata antarctica</i>	Lizard	<i>Pogona microlepidota</i>	WA	Jones (1986)
		<i>Pogona minima</i>	WA	Jones (1986)
		<i>Pogona minor</i>	WA	Jones (1986)
		<i>Pogona nullarbor</i>	WA	Jones (1986)
		<i>Tiliqua multifasciata</i>	WA	Jones (1992)
		<i>Tiliqua nigrolutea</i>	NSW, QLD	Johnston and Mawson (1947)
		<i>Tiliqua occipitalis</i>	SA, WA	von Linstow (1899), Jones (1995)
		<i>Tiliqua rugosa</i>	WA	Jones (1992)
		<i>Tiliqua scincoides</i>	NSW, QLD	Irwin-Smith (1922), Johnston and Mawson (1947)
		<i>Varanus acanthurus</i>	WA, NT	Jones (1983a)
		<i>Varanus giganteus</i>	WA	Jones (1985)
		<i>Varanus glebopalma</i>	WA	Jones (1988)
		<i>Varanus gouldii</i>	VIC, QLD, SA, WA	Johnston and Mawson (1947, 1951), Jones (1983b)
		<i>Varanus rosenbergi</i>	SA, WA	Johnston and Mawson (1941a), Jones (1983b)

Table 1.6. continued

		<i>Varanus tristis</i>	WA, SA	Jones (1995)
		<i>Varanus varius</i>	QLD, SA	Irwin-Smith (1922), Ortlepp (1922), Johnston and Mawson (1941b, 1947)
	Snake	<i>Acanthophis antarcticus</i>	SA	von Linstow (1899), Johnston (1911)
		<i>Austrelaps superbus</i>	TAS	Jones (2003)
		<i>Echiopsis curta</i>	WA	Jones (1978)
		<i>Elapognathus coronatus</i>	WA	Jones (1978)
		<i>Morelia spilota</i>	–	Ortlepp (1922)
		<i>Notechis scutatus</i>	WA	Jones (1978)
		<i>Pseudechis australis</i>	WA	Jones (1978)
		<i>Suta fasciata</i>	WA	Jones (1978)
<i>Oxyuris</i> sp.	Lizard	<i>Pseudemoia entrecasteauxii</i>	SA	Irwin-Smith (1922), Pichelin et al. (1999)
		<i>Tiliqua rugosa</i>	–	Thapar (1925)
<i>Thelandros trachysauri</i>	Lizard	<i>Egernia stokesii</i>	SA	Hallas et al. (2005)
		<i>Tiliqua occipitalis</i>	WA	Jones (1995)
		<i>Tiliqua rugosa</i>	WA, SA	Johnston and Mawson (1947), Jones (1992)

Table 1.6. continued

<i>Veversia tuberculata</i>	Lizard	<i>Tiliqua rugosa</i>	SA, VIC, NSW, QLD	von Linstow (1904), Johnston (1932)
TREMATODES				
<i>Brachylaima cribbi</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Butcher and Grove (2005)
<i>Microphallus sp.</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Angel and Mawson (1968)
<i>Paradistomum crucifer</i>	Lizard	<i>Christinus marmoratus</i>	SA	Mawson (1971)
		<i>Delma fraseri</i>	WA	Nicoll (1914)
		<i>Hemiergus peronii</i>	SA	Angel and Mawson (1968)
		<i>Tiliqua rugosa</i>	SA	Angel and Mawson (1968)
		<i>Tiliqua scincoides</i>	WA	Angel and Mawson (1968)
		<i>Varanus varius</i>	WA	Angel and Mawson (1968)

definitive host (Toledo et al. 2014), it is likely that sleepy lizards are accidental hosts of this intestinal fluke. *B. cribbi* has been recorded from various localities in the southern parts of Western Australia, South Australia, and Victoria (Butcher and Grove 2003). Many of the intermediate and definitive hosts are introduced species in Australia, so it is not clear whether *B. cribbi* is an introduced or native species in Australia (Butcher and Grove 2005).

As for the other trematode, *P. crucifer*, little information exists other than its definitive hosts and site of infection (i.e. bile and pancreatic ducts and liver). It is worth noting however, that first intermediate hosts of species from the family Dicrocoeliidae, are predominantly terrestrial snails, while certain species of ants are the second intermediate hosts (Manga-González and Ferreras 2014). It also appears that lizards are the definitive hosts of *P. crucifer* because this parasite has not been recorded from any other vertebrates (Table 1.6).

Angel and Mawson (1968) recorded one *Microphallus* sp. specimen from in the upper small intestines of a *T. rugosa* specimen from Middle Beach, South Australia. Aquatic snails and crustaceans are usually the first and second intermediate hosts of species from the family Microphallidae, while the definitive hosts are primarily birds although some mammal species have also been recorded (Kudlai et al. 2015). As in *B. cribbi*, it is therefore likely that sleepy lizards are accidental hosts of this fluke.

Viruses

Viruses are pathogens of all vertebrates, but due to the associated costs involved, investigations into viral infections tend to be limited to avian, fish and mammalian species of economic or human health importance, and as a result reptiles as hosts of viruses are very poorly studied (Clark and Lunger 1981). It is therefore not surprising that the most recently recorded microbe to infect *T. rugosa* is a nidovirus associated with a respiratory disease in these lizards (O’Dea et al. 2016), the first nidovirus recorded for lizards. Since mortality rates are high in the absence of appropriate supportive treatment (O’Dea et al. 2016), this disease is of conservation concern. The distribution of this virus and its transmission dynamics are not known, and additional research is urgently needed to address these questions.

Discussion and conclusion

Parasites are ubiquitous in the environment, yet we know very little about those that infect

wildlife (Gómez and Nichols 2013). We know much more about one particular host species, in a large part due to the pioneering work of Professor Michael Bull (Godfrey and Gardner 2017). This review not only summarises the known parasites of *T. rugosa*, but also indicates that additional research is needed to enhance our understanding of the natural history of sleepy lizards and their parasites. Based on the number of incomplete described parasitic species recorded from *T. rugosa*, it is likely that the understanding of which species are true parasites of these lizards is incomplete. In addition to that, some of the current listed parasites, such as *B. cribbi*, may in fact be accidental parasites and therefore not play a significant role in the natural history of these lizards.

It is likely that the taxonomy of the parasites reported require revision. For example, some records, for example *Bodo* sp. and *Endamoeba* sp. (Johnston 1932), seem doubtful since no other records of these protozoan species appear to have been reported from Australia (O'Donoghue and Adlard 2000). Others, such as *V. tuberculata* and *Oxyuris* sp. share some morphological characters with the genera *Pharyngodon*, *Tachygonetria* and/or *Thelandros* (Thapar 1925), and the validity of these classifications need to be verified.

This in turn raises the question of cryptic species. For instance, both the nematode families Oxyuridae and Pharyngodonidae belong to the order Oxyurida, and members of this order are usually considered strictly monoxenous (Anderson 2000), i.e. they infect a single host species. Yet, the species from these families that have been reported as parasites of *T. rugosa* have also been recorded from other lizards and snakes (Table 1.6).

Parasites, like all other organisms, have limiting factors that determine their distribution, and as a result there are often spatial variations in the parasite communities of a species. The sleepy lizard is a habitat generalist and can inhabit a variety of habitat types (Cogger 2014), so its parasites can be expected to vary, even on a relatively regional scale. A clear example of this is a study into the spatial variations of the subspecies of *S. enterica* at a study site near Mt. Mary in the Mid North region of South Australia. Four subspecies of this bacteria (*S. enterica* subsp. *diarizonae*, *S. enterica* subsp. *enterica*, *S. enterica* subsp. *houtenae*, and *S. enterica* subsp. *salamae*) were detected in *T. rugosa* at this locality (Parsons et al. 2015). It was also found that although individual hosts were infected with up to three of the *Salmonella enterica* subspecies, their distributions were not random, and was most likely as a result of variations in their abilities to survive in the abiotic environment, and the social networks of the lizards

determined which *S. enterica* subspecies they were infected with (Bull et al. 2012, Parsons et al. 2015). In addition to that, at the same locality, the distribution of the ticks *A. limbatum* and *B. hydrosauri* vary across a climate-induced ecological gradient (Bull and Burzacott 2001).

Distribution surveys from throughout the distribution range of *T. rugosa* are needed to determine the distribution range of the various parasite species. These surveys should also aim to verify the parasite species as parasites of these lizards. In order to rule out the likelihood of cryptic species, the parasite identification should ideally incorporate morphological, allometric, and molecular approaches as done in some other parasite studies (e.g. Kelehear et al. 2011, Tan et al. 2012, Scholz et al. 2014).

Apart from the studies of the impacts of *A. limbatum*, *B. hydrosauri*, and *H. mariae* on *T. rugosa* (Bull and Burzacott 1993, Main and Bull 2000, Bull and Burzacott 2006, Bouma et al. 2007), very little is actually known about the influence of the various parasites on *Tiliqua rugosa*, even though the available information suggest that other species likely also influence these lizards. For example, in a study in which nematodes (*Pharyngodon tiliquae* and *Th. trachysauri*) infections in *Egernia stokesii* were manipulated with antihelminthic drugs, it was found that dewormed lizards spent more time basking and moved about for longer times during observation sessions, which suggests that the nematode infection alters the behaviour of the host and reduces its fitness (Fenner and Bull 2008). *Th. trachysauri* also infects *T. rugosa*, and studies are needed to determine the influences of these and other parasites on this lizard species. Since influences of the parasites may vary due to spatial and temporal variations in intensity, as well as density-independent and density-dependent factors (Scott and Dobson 1989), future studies should be performed at various localities throughout the distribution ranges of *T. rugosa*.

Studies of the parasites of *T. rugosa* will contribute to the understanding of the natural history of the parasites as well as their host. Such information can be the basis of future studies of the influences of the parasites on sleepy lizards and their other hosts. The use of common species such as sleepy lizards as a model in parasitological studies is instrumental in elucidating general parasite infection and transmission dynamics, which can be an important management tool for proactive actions in the management of diseases in a variety of taxa.

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Research aims and chapter outline

The parasite communities of host species that are not very habitat specific and/or has a large geographic distribution can spatially vary, and these variations in the parasite community can be explored by examining the parasite communities of the host at localities that span across ecological gradients. In this thesis I contribute to the understanding of the parasite community of *T. rugosa* inhabiting the precipitation-induced ecological gradient at the Bunday Bore study site. My specific aims therefore are to:

1. Determine which ectoparasites, gastrointestinal helminths and haemoparasites infest or infect sleepy lizards at the study site.
2. Examine the distribution of the kangaroo soft tick at the study site.
3. Determine if sleepy lizards are a competent host for the kangaroo soft tick.
4. To test for an association between the recorded parasites and certain environmental conditions as indicated by plant communities.



Figure 1.8. The kangaroo soft tick (*Ornithodoros gurneyi*) is a rarely encountered mammal tick adapted to xeric habitats so occurs in arid and semi-arid habitats of Australia.

In Chapter 2, I report on the gastrointestinal helminths that I collected from sleepy lizards and a few other reptiles in the vicinity of the study site. Endoparasites such as gastrointestinal helminths can not always reliably be identified based on the eggs that are found in faecal examinations. It is therefore essential to determine which endoparasites are present in particular hosts within an area through other means against which eggs that are found during faecal

examinations can be cross-referenced. The most reliable method for obtaining adult gastrointestinal helminths for species identification purposes is via the dissection of potential hosts. I therefore dissected reptiles that were found dead on roads in the vicinity of the study site to examine their digestive tracts for the presence of gastrointestinal helminths. I also dissected the parts of the digestive tracts of lizards that were humanely killed as part of an unrelated study and donated to me. I collaborated with Prof. Charles Bursey (Pennsylvania State University, Pennsylvania, USA) and Prof. Stephen Goldberg (Whittier College, California, USA) in identifying the helminths that I collected.

In 2017 during the course of an unrelated study at the Bunday Bore study site, researchers from the University of California at Davis (California, USA) collected unknown arthropods from a sleepy lizard. With the assistance of Dr. Bruce Halliday (CSIRO Australian National Insect Collection), some were identified as exotic snake mite (*Ophionyssus natricis*). To determine whether this was just an isolated incidence, I conducted surveys in 2019 and 2020. In Chapter 3, I report on the findings in two publications, which is, to the best of our knowledge, the first record of snake mites parasitizing free-living sleepy lizards in South Australia. I also include a review of the distribution of this mite in Australia and a summary of the hosts they have been collected from. I then focus on sleepy lizards and present an argument that these lizards are very suitable hosts of this mite.

Sharrad (1979) recorded a few instances of the kangaroo soft tick parasitising sleepy lizards near Mt. Mary, a few kilometres south-east of the Bunday Bore study site, which is further south than what is believed to be the southern limit of the distribution of this tick. In 2017 I also recorded a few instances of these ticks parasitising sleepy lizards at the Bunday Bore study site. To determine if the low prevalence were due to limitations in the distribution of this tick I incorporated surveys from the environment with the surveys done on the lizards from late 2017 to 2019. I also incorporated data that I collected from lizards that were caught in 2020 and 2021 as part of an unrelated study. Ticks sometimes try to feed on atypical hosts, and since this may result in the death of the tick, it was necessary to determine if sleepy lizards are competent hosts of this parasite. I therefore used engorged ticks from the wild and captivity to examine whether they will survive and moult after feeding on the blood of sleepy lizards. In Chapter 4, I present the results of these studies in the form of a submitted manuscript. In it I describe the distribution range of this tick in the vicinity of the Bunday Bore study site, and report on the prevalence of parasitism by these ticks on sleepy lizards. I also present the result

of the survival experiment and show that sleepy lizards are competent hosts of this tick.

The Bunday Bore study site and the surrounding region has a semi-arid climate, with an average annual rainfall of less than 250 mm, of which most usually fall during the austral winter (Smyth 1973). The habitats in this region consist of a mosaic of areas that have been cleared for agriculture and grazing, interspersed with fragments of the arid mallee scrub and chenopod shrublands (Fig. 1.9) that used to dominate the south-western and north-eastern side of the study site respectively (Jessup 1948, Smyth 1973). For this thesis I utilized established transects (dirt roads), associated with a long-term monitoring of the sleepy lizards and their ticks. Variations in precipitation in an area can influence plant communities in such a way that it creates ecological gradients. To explore this in relation to the Bunday Bore study site I selected

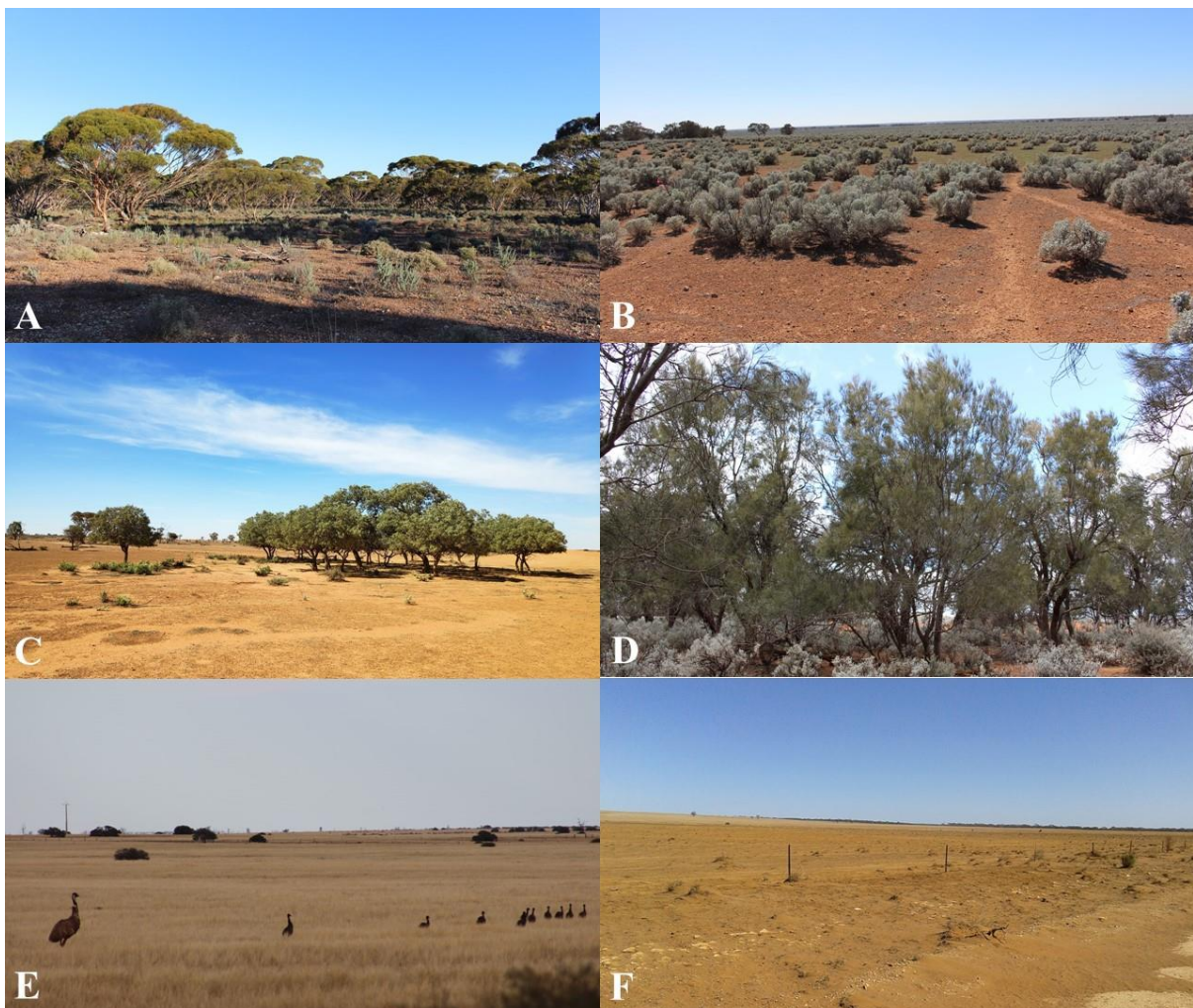


Figure 1.9. The habitats at the Bunday Bore study site and the surrounding area is a mosaic of fragmented remnants of mallee scrublands (A) and chenopod shrublands (B), often with isolated stands of trees such as *Alectryon oleifolius* (C) or *Casuarina pauper* (D). Some parts have been cleared to create grassy paddocks (E), while certain areas are almost bare due to bad land management practices (F).

two transects that span across the transition from chenopod shrubland to mallee scrubland. I collected vegetation data according to the Braun-Blanquet cover abundance scale seasonally from survey plots along the transects. In collaboration with Prof. Leslie Brown (Applied Behavioural Ecology and Ecosystem Research Unit, University of South Africa) I analysed the floristic data. In Chapter 5, I report on the plant species recorded during the surveys and describe the roadside plant communities that were identified. I then identify species that are indicators of particular environmental conditions along the transects.

Species that can inhabit a variety of habitat types often occur throughout an ecological gradient, whereas those that are more habitat specific tend to have a more restricted distribution (Pianka 1966, Sutherst and Maywald 1985, Lynch and Gabriel 1987). This applies to free-living species as well as parasites. In Chapter 6, I report on the prevalence of the various parasite species I recorded in the years 2017 to 2019, as well as their distribution in relation to the study site. I then use the data collected from lizards that were caught along Bunday Church Road and Salford Road in 2018 to explore which species are drivers in the differences in the parasite community and whether there is an association between these parasites and the environmental conditions as indicated by different plant communities along two transects.

In the final chapter I discuss the collective results from the study into the parasites at the Bunday Bore study site. I also highlight gaps in our knowledge for future studies and provide food for thought on the conservation of the parasites of sleepy lizards.

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Chapter 2

New host and locality records for gastrointestinal helminths of five reptile species from the Mid North region of South Australia

Note to examiners

Helminths are a large often neglected group of parasites and our understanding of their infections in *Tiliqua rugosa* is limited. Here I increase knowledge on the species and the geographic distribution and prevalence of several helminth species that infect reptiles from the Mid North region of South Australia. This chapter consist of observations on the gastrointestinal helminths from dissected specimens of *Tiliqua rugosa* and four other common reptile species from the study site and surrounding areas. This was done because intestinal parasites can not always be reliably identified based on their eggs from faecal examinations. The adult stages should therefore be obtained for verification. This chapter has been published in the journal *Transactions of the Royal Society of South Australia* and is therefore formatted according to the specifications of that journal (e.g. referencing and indicating the authority and publication date when a species is first mentioned in the text), except for the referencing style, which has been changed to make it consistent with that of the other parts of the thesis. The numbering of the figure and tables were also changed to make them pertaining to this chapter.

Abstract

Parasites in wildlife tend to be poorly studied in spite of the ecological roles they play, and even basic information such as the geographic distribution of the parasites and the hosts they infect or infest are not well known. For parasites that live within the host, part of the reason for this lack of information is that the hosts must be killed and dissected to determine which endoparasites they harbour, which has legal and/or ethical implications and limitations. These obstacles can be overcome by collecting and examining potential hosts that are found dead on roads or that are sacrificed as part of other non-related studies. In this study our objective was to expand on our understanding of the gastrointestinal parasites of some common reptiles from an area in the vicinity of a study site in the Mid North region of South Australia by making use of snakes and large lizards that were found dead on roads or that were humanely killed for another study. Our study provides new locality and/or host species records for some parasite species.

Introduction

Parasitism is a very efficient life strategy, which is evident through the ubiquity of parasites and because many species are parasitic in at least part of their life cycle (Windsor 1998). Through the negative influences of parasites on their hosts, parasites play various ecological roles such as the regulation of host populations, increasing energy flow and/or influencing host communities via parasite-mediated competition (Hudson and Greenman 1998, Thomas et al. 1999, Valkiūnas 2001, Wood and Johnson 2015). Although the influence of parasites is generally considered to be negative, this is not necessarily to a substantial degree. For instance, studies on the endangered woylie (*Bettongia penicillate* Gray, 1837) have found that these marsupials are parasitized upon by a variety of ecto- and endoparasites, some of which are host specific (Thompson et al. 2018). Yet, only *Trypanosoma copemani* Austen, Jefferies, Friend, Ryan, Adams and Reid, 2009 has been determined to be pathogenic (Thompson et al. 2014), which only appears to be so when there is a lack of competitive interaction from other trypanosome species (Thompson et al. 2018). This highlights the importance of an understanding of all the parasite species of a particular host. However the parasite fauna of most wildlife species are poorly documented (Poulin and Leung 2010), and in spite of their ecological importance, many parasites are undiscovered or described, most described parasite species are relatively poorly studied, and as a group parasites are generally overlooked in conservation and management plans and biodiversity monitoring (Windsor 1995, Gómez and Nichols 2013, Carlson et al. 2020, Kwak et al. 2020). Carlson et al. (2020) outlined a global parasite conservation plan, focussing on data collection and synthesis, risk assessment and prioritizing, conservation practice, and outreach and education. A key component of this conservation plan is fundamental information such as the host species of the various life stages and the geographic distribution of a particular parasite to develop a better understanding of its ecological roles and to determine its conservation status.

Molecular techniques and the examination of scat samples can determine the presence of a parasite species but do not indicate the life stage of the parasite, which organs of the host are infected, or the level of infection. Due to these shortcomings, studies to determine the host species and geographic distribution of endoparasites often require the killing and dissection of sampled specimens of the host species, which have legal and/or ethical implications and limitations. The use of animals that are found dead on roads (roadkill) and specimens that are sacrificed as part of unrelated studies can therefore be utilised instead of collecting and

sacrificing specimens specifically for endoparasite host and geographic distribution studies (Carlson et al. 2020). This investigation made use of such specimens that were collected on an *ad hoc* basis with the aim to expand on the understanding of the gastrointestinal parasites of reptiles in the Mid North region of South Australia, as part of an expansion on the ongoing long-term study initiated by the late Prof. Michael Bull on sleepy lizards and their ticks (Godfrey and Gardner 2017).

Material and methods

During the months August to December in the years 2017 to 2019, we collected reptiles that were found dead on roads leading to and within the Bunday Bore Station study site in the Mid North region of South Australia, ca. 160 km north of Adelaide, as well as from neighbouring areas (Fig. 2.1). Specimens that were in an advanced stage of decomposition or very damaged were not collected. In addition, we collected a juvenile sleepy lizard, *Tiliqua rugosa* (Gray 1825) that was killed by an eastern brown snake, *Pseudonaja textilis* (Duméril, Bibron, & Duméril, 1854) but not ingested (Norval et al. 2018). The snout-vent length (SVL) of all the hosts was determined as follows: when the SVL of the specimen was shorter than 40 cm, this body length measurement was measured with a transparent plastic ruler to the nearest mm; if it exceeded 40 cm it was measured to the nearest mm with a steel measuring tape. If the head of the animal was crushed the SVL was not recorded. The carcasses were examined fresh and each was dissected by a longitudinal incision from the throat to the rectum, and the gastrointestinal tract was removed by cutting all the connective tissue and across the oesophagus and the rectum. The oesophagus, stomach, and small and large intestines were slit open longitudinally and separately searched for helminths under a dissecting microscope at $\times 10$ to $\times 50$ magnifications. The inner and outer wall of the digestive tract and body cavity were also examined for free-moving helminths and cysts containing larvae. Helminths were collected and preserved in 70% ethanol.

We also dissected and examined parts of the digestive tracts of 22 *T. rugosa* and nine eastern blue tongue skinks, *Tiliqua scincoides* (White 1790) that were preserved in 70% ethanol and donated to us for examination after they were humanly killed as part of an unrelated project (Animal Welfare Committee of Flinders University approval No. E481/19) and the Department of Environment, Water and Natural Resources of the Government of South Australia permit A23436). Fourteen of the *T. rugosa* and five *T. scincoides* were from known localities in the

Mid North of South Australia, while the remainder of the specimens were from unknown localities within the same geographic region. The digestive tracts were dissected in the same manner as the other specimens and helminths that were found were collected and preserved in 70% ethanol.

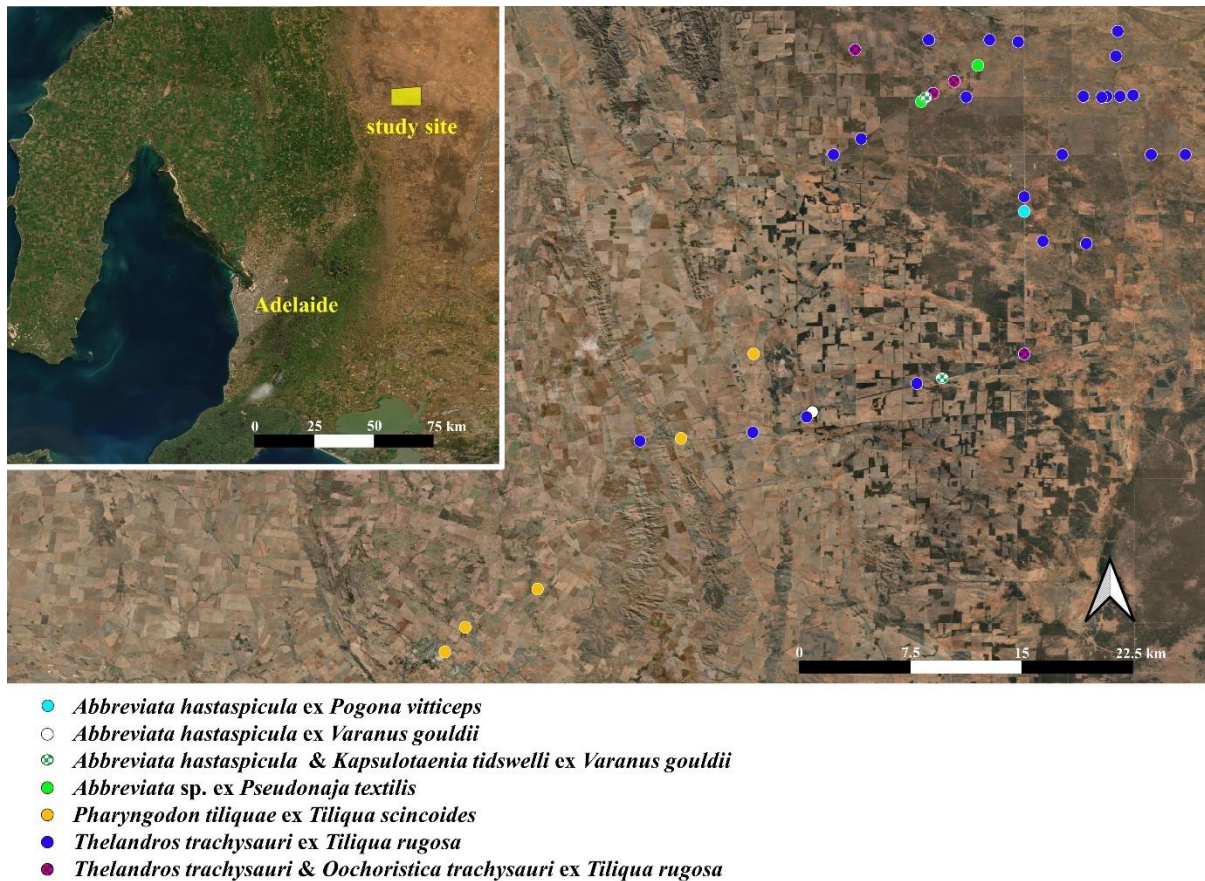


Figure 2.1. The position of the study site in relation to Adelaide, South Australia (inset), and the distribution of the gastrointestinal helminths recorded in this study from hosts of known collection localities.

For study, helminths were removed from the 70% ethanol. Nematodes were cleared in a drop of undiluted glycerol on a glass slide and identified from these temporary preparations (Magath 1916). Cestodes and digeneans were regressively stained with Harris' hematoxylin, dehydrated in an alcohol series, cleared in xylene, whole-mounted in Canada balsam, and examined by compound microscopy (Sepulveda and Kinsella 2013). Identification of the digenean is based upon the descriptions in Butcher et al. (2002); cestodes upon the keys of Khalil et al. (1994) and original descriptions; and nematodes are based upon the reference keys of Anderson et al. (1974) and original descriptions. It was not possible to determine the infection intensity for most of the examined specimens since the specimens were often damaged and only parts of the digestive tract could be thoroughly searched. The prevalence of each parasite species in each

host species was determined as a percentage of the number of specimens of each host species infected by the respective parasites (Bush et al. 1997). Selected helminths were deposited in the Australian Helminthological Collection (AHC) of the South Australian Museum as *Oochoristica trachysauri* ex *Tiliqua rugosa* AHC 36686 to AHC 36692, and in the Harold W. Manter Laboratory (HWML), University of Nebraska, Lincoln, USA, as *Abbreviata hastaspicula* ex *Pogona vitticeps* HWML 112109; *Abbreviata hastaspicula* ex *Varanus gouldii* HWML 112114; *Abbreviata* sp. ex *Pseudonaja textilis* HWML 112110; *Brachylaima cribbi* ex *Tiliqua rugosa* HWML 216366; *Kapsullotaenia tidswelli* ex *Varanus gouldii* HWML 112115; *Pharyngodon tiliquae* ex *Tiliqua scincoides* HWML 112113; *Thelandros trachysauri* ex *Tiliqua rugosa* HWML 112111; and *Thelandros trachysauri* ex *Tiliqua scincoides* HWML 112112. The open software QGIS (Version 3.16.3; available at <http://qgis.osgeo.org>) was used for creating the study site and parasite species distribution map.

Results

We examined 59 reptiles from five species belonging to the squamate families Agamidae, Elapidae and Scincidae (Table 2.1). Gastrointestinal parasites were recorded in 55 (93.2%) of the examined reptiles and they harboured one or two species (mean \pm SD = 1.1 \pm 0.4) of parasites. All the recorded parasites were helminths and belonged to the phyla Nematoda and Platyhelminthes (Table 2.1). Nematodes were the most numerous parasites recorded, and were identified as *Abbreviata hastaspicula* Jones, 1979, *Abbreviata* sp. (fourth stage), *Pharyngodon tiliquae* Baylis, 1930 and *Thelandros trachysauri* Johnston and Mawson, 1947. The recorded platyhelminths were the trematode *Brachylaima cribbi* Butcher and Grove, 2001, and the eucestodes *Kapsulotaenia tidswelli* (Johnston 1909) and *Oochoristica trachysauri* (MacCallum 1921). *Kapsulotaenia tidswelli*, *O. trachysauri* and *P. tiliquae* infected only one host species, while *A. hastaspicula*, *B. cribbi* and *T. trachysauri* were recorded from two host species (Table 2.1). *Abbreviata hastaspicula* and fourth stage *Abbreviata* sp. larvae were found in the digestive tract as well as occasionally within the body cavity of the host. *Brachylaima cribbi* specimens were collected from the intestines. *Kapsulotaenia tidswelli* and *O. trachysauri* were only found in the small intestines, while the nematodes *P. tiliquae* and *T. trachysauri* were only recorded from the large intestines of their hosts.

Table 2.1. The numbers and the ranges of snout-vent lengths (SVL), with means and standard deviations in parenthesis, of the reptiles examined in this study, and the families, species and infection prevalence of the helminths that were collected from them.

Species	Number	SVL (mm)	Family	Parasite	Prevalence
LIZARDS					
<i>Pogona vitticeps</i>	2	176 – 197 (186 ± 10)	Physalopteridae	<i>Abbreviata hastaspicula</i>	50%
<i>Tiliqua rugosa</i>	38	173 – 351 (269.9 ± 30.3)*	Brachylaimidae	<i>Brachylaima cribbi</i>	2.6%
			Linstowiidae	<i>Oochoristica trachysauri</i>	10.5%
			Pharyngodonidae	<i>Thelandros trachysauri</i>	97.4%
<i>Tiliqua scincoides</i>	12	105 – 300 (243 ± 62.3)*	Brachylaimidae	<i>Brachylaima cribbi</i>	8.3%
			Pharyngodonidae	<i>Pharyngodon tiliquae</i>	83.3%
			Pharyngodonidae	<i>Thelandros trachysauri</i>	16.7%
<i>Varanus gouldii</i>	3	274 – 362 (327 ± 38.1)	Physalopteridae	<i>Abbreviata hastaspicula</i>	100%
			Proteocephalidae	<i>Kapsulotaenia tidswelli</i>	66.7%
SNAKES					
<i>Pseudonaja textilis</i>	4	797 – 1140 (973 ± 140.2)*	Physalopteridae	<i>Abbreviata</i> sp. (4th stage)	50%

* The SVLs of the following numbers of specimens were not recorded so were excluded from the SVL analyses: *Tiliqua rugosa* = 3; *Tiliqua scincoides* = 1; *Pseudonaja textilis* = 1.

Discussion

All of the parasite species recorded in this study, except for the trematode *B. cribbi* that has only been reported from South Australia, have been reported from more than one state, indicating that they have a wide geographic distribution in Australia (Table 2.2), but our records are new locality records for some of the parasite species. Also, with the exception of *O. trachysauri* that has only been recorded from *T. rugosa*, all the recorded parasite species have been recorded from more than one host species (Table 2.2), suggesting that they are either generalist parasites or host specificity is limited to related species. However, we present new host records for some of the parasite species reported on herein.

Adult worms of physalopteran nematodes of the genus *Abbreviata* are needed for species identification, so it is not possible to determine which species had infected the *P. textilis* reported on herein. *Abbreviata hastaspicula* has previously been reported from the sand goanna, *Varanus gouldii* (Gray 1838), which is its predominant host (King and Jones 2016), as well as various other varanid species and even a black-headed python, *Aspidites melanocephalus* Krefft, 1864 (Table 2.2). However, to the best of our knowledge our records are new distribution records and the central bearded dragon, *Pogona vitticeps* (Ahl 1926) is a new host record. As in other physalopterans *A. hastaspicula* requires an arthropod intermediate host to complete its life cycle (Anderson 2000, King and Jones 2016) and its definitive hosts are medium-sized varanids inhabiting xeric habitats (Jones 2014). If an arthropod that is infected by a third larval stage (the infective stage) of a physalopteran is consumed by an atypical vertebrate host (paratenic host) the infective stage of the parasite persists without essential development and growth (Anderson et al. 1974, Anderson 2000). Our observation therefore indicates that large agamids, such as *P. vitticeps*, can be definitive hosts for this nematode. The natural intermediate hosts of *A. hastaspicula* is not known (King et al. 2013). Empirical studies are therefore needed to determine the natural intermediate hosts of this parasite as well as its prevalence in *P. vitticeps*, a common but poorly studied lizard species, in order to gain a better understanding of the natural history of *A. hastaspicula* and its hosts.

The digenean trematode *B. cribbi* has previously been reported from *T. rugosa* (Butcher and Grove 2005), but as far as we can determine *T. scincoides* is a new host record. Even though this parasite has only been reported from the southern Australian states South Australia and Victoria, it is suspected to be of European origin due to its high prevalence in European helcid

Table 2.2. A summary of the reported Australian hosts of the helminth species recorded in this study (WA – Western Australia; NT – Northern Territory; QLD – Queensland; NSW – New South Wales; V – Victoria; SA – South Australia; Unk – Unknown).

Species	Host type	Hosts	Locality	Reference
CESTODES				
<i>Kapsulotaenia tidswelli</i>	Lizard	<i>Varanus giganteus</i>	WA	Thompson et al. (2020)
		<i>Varanus gouldii</i>	QLD, SA, WA	Johnston (1912a); Thompson et al. (1980); Thompson et al. (2020); herein
		<i>Varanus varius</i>	NSW, QLD	Irwin-Smith (1922); Johnston (1909); Johnston (1912b); Johnston (1913); Jones (2007)
<i>Oochoristica trachysauri</i>	Lizard	<i>Tiliqua rugosa</i>	NSW, QLD, SA, VIC, WA	MacCallum (1921); Johnston (1932); Angel and Mawson (1968); herein
NEMATODES				
<i>Abbreviata hastaspicula</i>	Lizard	<i>Pogona vitticeps</i>	SA	herein
		<i>Varanus acanthus</i>	NT, WA	Jones (1983b)
		<i>Varanus giganteus</i>	WA	Jones (1985)
		<i>Varanus gouldii</i>	NT, SA, WA	Jones (1983a); Jones (1986); Jones (1988); Jones (1995); Barton and Jones (2018); herein
		<i>Varanus mertensi</i>	Northern Australia	Jones (1988)
		<i>Varanus mitchelli</i>	Northern Australia	Jones (1988)
		<i>Varanus panoptes</i>	NT, WA	Jones (1983a); Jones (1988)

Table 2.2 continued

		<i>Varanus spenceri</i>	NT	Barton and Jones (2018)
		<i>Varanus storri</i>	WA	Jones (1988)
		<i>Varanus tristis</i>	WA, Northern Australia	Jones (1986); Jones (1988); Jones (1995)
	Snake	<i>Aspidites melanocephalus</i>	QLD	Jones (1979)
		<i>Aspidites ramsayi</i>	NT	Barton and Jones (2018)
		<i>Stegonotus cucullatus</i>	NT	Barton and Jones (2018)
		<i>Tropidonophis mairii</i>	NT	Barton and Jones (2018)
<i>Abbreviata</i> sp.	Lizard	<i>Ctenophorus nuchalis</i>	WA	Jones (1995)
		<i>Ctenotus regius</i>	SA	Goldberg and Bursey (1995)
		<i>Ctenotus schomburgkii</i>	SA	Goldberg and Bursey (1995)
		<i>Intellagama lesueurii</i>	NT	Barton and Jones (2018)
		<i>Moloch horridus</i>	WA	Bursey et al. (1996)
		<i>Nephrurus laevis</i>	WA	Bursey and Goldberg (1999)
		<i>Nephrurus levis</i>	NT, WA	Bursey and Goldberg (1999)
		<i>Varanus acanthus</i>	WA	Jones (1983b)
		<i>Varanus giganteus</i>	WA	Jones (1985)
		<i>Varanus glauerti</i>	Northern Australia	Jones (1988)
		<i>Varanus glebopalma</i>	Northern Australia	Jones (1988)
		<i>Varanus gouldii</i>	WA, NT, Northern Australia	Jones (1983a); Jones (1988); Barton and Jones (2018)

Table 2.2 continued

	<i>Varanus mertensi</i>	Northern Australia	Jones (1988)
	<i>Varanus mitchelli</i>	Northern Australia	Jones (1988)
	<i>Varanus panoptes</i>	Northern Australia	Jones (1988)
	<i>Varanus scalaris</i>	Northern Australia	Jones (1988)
	<i>Varanus storri</i>	Northern Australia	Jones (1988)
	<i>Varanus tristis</i>	Northern Australia	Jones (1988)
Snake	<i>Acanthophis antarcticus</i>	WA	Jones (1978)
	<i>Acanthophis pyrrhus</i>	WA	Jones (1978)
	<i>Aspidites melanocephalus</i>	Unk	Barton and Jones (2018)
	<i>Aspidites ramsayi</i>	NT, QLD	Jones (1978); Barton and Jones (2018)
	<i>Demansia vestigiata</i>	QLD	Barton and Jones (2018)
	<i>Denisonia fasciata</i>	WA	Jones (1978)
	<i>Denisonia punctata</i>	WA	Jones (1978)
	<i>Drysdalia coronata</i>	WA	Jones (1978)
	<i>Echiopsis curta</i>	WA	Jones (1978)
	<i>Liasis olivaceus</i>	WA	Jones (1978)
	<i>Notechis ater</i>	WA	Jones (1978)
	<i>Pseudechis australis</i>	WA	Jones (1978); Barton and Jones (2018)
	<i>Pseudechis porphyriacus</i>	QLD	Barton and Jones (2018)
	<i>Pseudonaja affinis</i>	WA	Jones (1978)

Table 2.2 continued

		<i>Pseudonaja modesta</i>	WA	Jones (1978)
		<i>Pseudonaja nuchalis</i>	WA	Jones (1978)
		<i>Pseudonaja textilis</i>	SA, WA	Jones (1978); herein
		<i>Simoselaps bertholdi</i>	WA	Jones (1978)
		<i>Stegonotus cucullatus</i>	NT	Barton and Jones (2018)
		<i>Tropidonophis mairii</i>	NT, QLD	Barton and Jones (2018)
<i>Pharyngodon tiliquae</i>	Lizard	<i>Cyclodomorphus melanops</i>	WA	Jones (1992)
		<i>Egernia cunninghamii</i>	NSW	Johnston and Mawson (1947); Adamson (1984)
		<i>Liopholis kintorei</i>	SA	Adamson (1984)
		<i>Egernia stokesii</i>	SA	Hallas et al. (2005)
		<i>Egernia striolata</i>	NSW, SA	Johnston and Mawson (1947); Adamson (1984); Norval et al. (2021)
		<i>Egernia whitii</i>	NSW	Johnston and Mawson (1947); Adamson (1984)
		<i>Nephrurus vertebralis</i>	WA	Bursey and Goldberg (1999)
		<i>Tiliqua multifasciata</i>	WA	Jones (1992)
		<i>Tiliqua occipitalis</i>	WA	Jones (1992)
		<i>Tiliqua scincoides</i>	QLD, SA	Baylis (1930); Johnston and Mawson (1947); Adamson (1984); herein

Table 2.2 continued

<i>Thelandros trachysauri</i>	Lizard	<i>Egernia stokesii</i>	SA	Hallas et al. (2005)
		<i>Tiliqua occipitalis</i>	WA	Jones (1995)
		<i>Tiliqua rugosa</i>	SA, WA	Johnston and Mawson (1947); Jones (1992)
		<i>Tiliqua scincoides</i>	SA	herein
TREMATODES				
<i>Brachylaima cribbi</i>	Lizard	<i>Tiliqua rugosa</i>	SA	Butcher and Grove (2005); herein
		<i>Tiliqua scincoides</i>	SA	herein

snail and bird species that have been introduced into Australia from Europe (Butcher and Grove 2001, 2003, 2005). The snails are intermediate hosts of this parasite and the birds, mammals and reptiles are the definitive hosts, which are infected when they ingest infected snails (Butcher and Grove 2001, 2003, 2005). Snails are common food items of *T. rugosa* and *T. scincoides* (Satrawaha and Bull 1981, Yeatman 1988, Dubas and Bull 1991, Koenig et al. 2001, Turner 2010), but these lizards are most likely incidental hosts.

The eucestode *K. tidswelli* has previously been recorded as a parasite of *V. gouldii*, however our record appears to be a new distribution record. The life cycle of only a few proteocephalid species are fully known, but it can be assumed that the life cycle of most species is probably similar to some extent (Frank 1981). The eggs, which are excreted along with the faeces of the vertebrate definitive host, are ingested by a small crustacean (Copepoda) (Frank 1981). The definitive host becomes infected when it ingests an infected copepod or an infected paratenic vertebrate host, which can be an amphibian, fish or reptile (Frank 1981). Based on the available information, the adults of *K. tidswelli* are host specific to some extent, only infecting medium-sized varanids (Table 2.2). In addition, the *V. gouldii* used in our study were collected in arid habitats, with intermittent water sources, suggesting that the intermediate host of *K. tidswelli* is very likely a terrestrial arthropod. Nothing else is known about host specificity or the life cycle of *K. tidswelli* and additional studies are required to determine the arthropod intermediate hosts and possible vertebrate paratenic hosts.

Oochoristica trachysauri is a common parasite of *T. rugosa* in South Australia, New South Wales, Victoria, and southern Queensland (Johnston 1932), and appears to be definitive host specific since it has not been reported from any other vertebrates in spite of its broad geographic distribution. The intermediate hosts of *O. trachysauri* are not known. Studies on the development and/or lifecycle of *Oochoristica anolis* Harwood, 1932, *Oochoristica javaensis* Kennedy, Killick, & Beverley-Burton, 1982, *Oochoristica osheroffi* Meggitt, 1934 and *Oochoristica vacuolata* Hickman, 1954 identified coleopterans and/or orthopterans as suitable intermediate hosts (Hickman 1963, Widmer and Olsen 1967, Conn 1985, Criscione and Font 2001). Several coleopterans and orthopterans have been recorded as food items of sleepy lizards (Norval and Gardner 2020), and additional empirical studies are needed to determine whether any of them are the intermediate hosts of *O. trachysauri*.

The pinworms (Order: Oxyurida) *P. tiliquae* and *T. trachysauri* have previously been reported

from several lizard species from various states in Australia (Table 2.2). Interestingly, with the exception of the midline knob-tailed gecko, *Nephrurus vertebralis* Storr, 1963 (Family: Carphodactylidae), all the known hosts are skinks (Family: Scincidae) from the subfamily Egerniinae, which may reflect the research concentration on this sub-family. Some of these species, such as *Egernia stokesii* (Gray 1845), are social and form stable family groups that use the same refuges (Chapple 2003, Gardner et al. 2007). Others, such as *T. rugosa*, are more solitary and only associate with conspecifics during the breeding season (Bull 1988). Oxyurids have a direct life cycle and infection is via the faecal-oral route (Telford 1971, Frank 1981) so infection by *P. tiliquae* and *T. trachysauri* most likely take place when lizards use tongue flicks to investigate chemical cues from scats of conspecifics. Hallas and Bull (2006) noted that both these nematode species can be sympatric in *E. stokesii* and that the eggs of *P. tiliquae* is more sensitive to desiccation than that of *T. trachysauri*. Jones (1992), however found that in *Tiliqua occipitalis* (Peters 1863) coinfections were rare and statistically insignificant. In our study we found that even though *T. rugosa* infested by *T. trachysauri* and *T. scincoides* infested by *P. tiliquae* are sympatric in parts of the study site (Fig. 2.1), no instances of coinfection were noted. A possible explanation is the exclusion of the one species by another through interference competition as seen in studies on oxyurids of some insects (Adamson 1990). Another explanation could be that there are differences in the gut environment requirements of *P. tiliquae* and *T. trachysauri*. All the known hosts of *P. tiliquae* are either insectivorous or omnivorous (Table 2.3). As for *T. trachysauri*, all the known hosts are omnivorous and the diet of adults tend to consist primarily of plant material. Nothing is known about the differences in the gut environment of these species. The role that the gut environment of the host and/or interference competition play in the niche partitioning of *P. tiliquae* and *T. trachysauri* needs to be verified through empirical studies.

This study expanded our understanding of the geographic distribution and the host species of several endoparasites from Australia. It also highlighted the need for additional empirical studies to elucidate the life cycles of many of the parasites whose natural history have not been studied.

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Table 2.3. The foraging strategy of the known hosts of the oxyurid nematodes *Pharyngodon tiliquae* and *Thelandros trachysauri* (* species that exhibit an ontogenetic shift from a more insectivorous to a more herbivorous foraging strategy as they mature).

Species	Foraging strategy	Reference
<i>Cyclodomorphus melanops</i>	Insectivorous	Pianka (2011)
<i>Egernia cunninghamii</i> *	Omnivorous	Brown (1991)
<i>Egernia stokesii</i> *	Omnivorous	Duffield and Bull (1998)
<i>Egernia striolata</i>	Omnivorous	Bustard (1970)
<i>Egernia whitii</i>	Omnivorous	Brown (1991); Hickman (1960)
<i>Liopholis kintorei</i>	Omnivorous	McAlpin (2001)
<i>Nephrurus vertebralis</i>	Insectivorous	How et al. (1990); Pianka and Pianka (1976)
<i>Tiliqua multifasciata</i>	Omnivorous	Pianka (2011)
<i>Tiliqua occipitalis</i>	Omnivorous	Shea (2006)
<i>Tiliqua rugosa</i>	Omnivorous	Norval and Gardner (2020)
<i>Tiliqua scincoides</i>	Omnivorous	Yeatman (1988)

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Chapter 3

The snake mite (*Ophionyssus natricis*) a neglected introduced ectoparasite of sleepy lizards

Note to examiners

The snake mite (*Ophionyssus natricis*) is an exotic invasive parasite, usually associated with lizards and snakes in captivity. This species was formerly believed to be restricted to urban and suburban environments along the eastern and southern coastal regions of Australia. In 2017, we found some specimens by chance in our study site in the Mid North region of South Australia. As this mite had not been formally recorded as a parasite of *T. rugosa* in our study site, this prompted a review of the distribution of this mite in Australia and an investigation to verify if this mite occurs in the study site, which forms the first part of this chapter. In 2020 more instances were found of snake mite parasitising sleepy lizards in areas surrounding the study site, as well as in a suburban conservation park in Adelaide. A report on these distribution records and a discussion on the suitability of sleepy lizards as hosts of the snake mite forms the second part of this chapter. The first and second part of this chapter were published in the journals *Acarologia* and *Transactions of the Royal Society of South Australia* respectively. They are therefore formatted according to the specifications of the respective journal (e.g. *Transactions of the Royal Society of South Australia* requires referencing and indicating the authority and publication date when a species is first mentioned in the text). Please also note that in the report published in *Acarologia* some of the generic names were abbreviated with two letters as opposed to one to avoid confusion and they remain in that format in the relevant part herein. The referencing style of both reports were however changed to make them consistent with that of the other parts of my thesis. The numbering of the figures and tables were changed for clarification and so that they appear chronologically in the chapter. Unless stated otherwise, I took the photographs and micrographs and therefore own the copyright.

Occurrence of the introduced snake mite, *Ophionyssus natricis* (Gervais, 1844) in the wild in Australia

Abstract

The snake mite, *Ophionyssus natricis* (Acari: Macronyssidae), occurs in many parts of the world and is of animal and human health significance. In Australia *Op. natricis* is considered an introduced species in parts of coastal eastern and southern Australia and is thought to be absent in the wild. Herein we report on the occurrence of *Op. natricis* in Adelaide and a rural locality in South Australia on lizards from the genus *Tiliqua*. We also review previous records of this mite species based on published reports and the examination of museum specimens. The results show that the snake mite has been collected on wild hosts at several localities in at least two states and is therefore not restricted to reptiles in captivity.

Introduction

Of all the mite species that are known to infest reptiles, one of the most problematic is the snake mite, *Ophionyssus natricis* (Gervais 1844) because it is of animal and human health significance. Snakes and lizards infested by this mite may suffer from abnormal shedding (i.e. dysecdysis), and heavy infestation can cause anemia and even result in mortality (Frank 1981, Lane and Mader 1996, Šlapeta et al. 2018). This mite has also been identified as a possible vector of several blood-borne bacterial, filariid and viral pathogens of snakes (Wozniak and DeNardo 2000), and has also been implicated in instances of dermatitis in humans (Schultz 1975, Beck 1996, Amanatfard et al. 2014).

The native range of *Op. natricis* is uncertain. *Ophionyssus natricis* was originally described from specimens taken from snakes in a zoo in Europe and were subsequently also collected from areas around Paris and Rome (Gervais 1844). However, it has subsequently been reported in many parts of the world from captive snakes and lizards in zoo collections and the pet trade, and even a few instances from reptiles in the wild (Yunker 1956, Goldberg and Bursey 1991, Miranda et al. 2017), which indicates that it is easily spread via the movement of animals. In Australia *Op. natricis* is considered an introduced species and has been recorded in parts of coastal eastern and southern Australia (Domrow 1988). Herein we report infestation by *Op. natricis* of three skinks (family: Scincidae) from three localities in South Australia, with comments on the distribution of this parasite in Australia based on museum specimens and published records.

Observation

Case 1. On 19 September 2017, seven (four female and three male) sleepy lizards (*Tiliqua rugosa*) were collected from a study site surrounding the Bunday Lutheran church ruins, ca. 130 km north-northeast of Adelaide City, in a pastoral district in the mid north region of South Australia. The habitat of this study site is a chenopod shrubland, dominated by the shrubs *Atriplex vesicaria*, *Maireana pyramidata* and *Maireana sedifolia*, with scattered stands of the tree *Casuarina pauper*. The lizards were collected as part of an ongoing study into the roles of personality and sociality in the spread of parasites in these lizards (Sih et al. 2018), and were kept individually overnight in sterilised calico bags. The following morning (20 September 2019) after the lizards had been released back into the wild, 14 tiny (ca. 1 mm in length) arthropods were collected from inside one of the fabric bags. The arthropods were preserved

in 95% ethanol, and were later determined to be one larva of the ixodid tick *Bothriocroton hydrosauri*, four larvae of the argasid tick *Ornithodoros gurneyi*, and nine females of *Op. natricis*. In a subsequent follow-up investigation between 17 August 2019 and 18 November 2019, 26 *T. rugosa* were caught in the vicinity of the abovementioned study site and four were caught in the vicinity of an earth dam located ca. 3.5 km southwest of the study site. An additional three *T. rugosa* were caught in the outskirts of Eudunda, the nearest town, ca. 35 km southwest of the study site. The lizards were held within a plastic container and brushed with a fine paintbrush (Figure 3.1) prior to being released at the point of capture. The container was then rinsed out with 70% ethanol, that was poured through a fine (0.25 x 0.25 mm mesh) polyester fabric. The fabric was subsequently examined under a dissection microscope for the presence of *Op. natricis*. None of the examined lizards were found to be infested by snake mite except for one that was collected on 17 August 2019 inside a dried-up earth dam (Figure 3.2; S33.88845 E139.31065; datum WGS84) at the study site surrounding the Bunday Lutheran Church ruins. Four females of *Op. natricis* (Figure 3.3) were collected from the lizard, but when it was subsequently recaptured on 18 November 2019 less than 50 m from where it was initially



Figure 3.1. The sleepy lizards (*Tiliqua rugosa*) were checked for mite by holding them within a plastic container and brushing the areas under their scales with a fine paintbrush (photographed by Jessica Clayton).

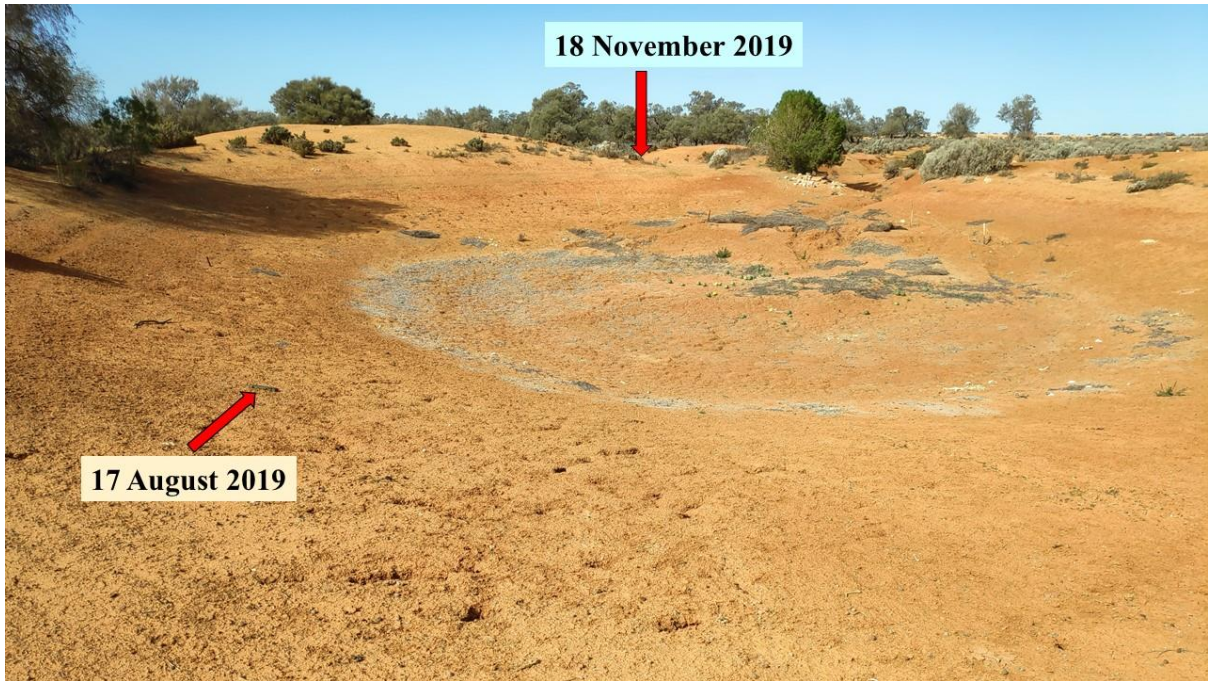


Figure 3.2. The locality inside the dried-up earth dam where the infested lizard was caught and where it was recaptured four months later (photographed by Gerrut Norval).



Figure 3.3. One of the four engorged females of *Ophionyssus natricis* that were collected from a sleepy lizard (*Tiliqua rugosa*) on 17 August 2019 at the study site surrounding the Bunday Lutheran Church ruins in the Mid North of South Australia (photographed by Gerrut Norval).

captured (Figure 3.2), no *Op. natricis* were found. None of the other lizards were captured more than once. The *Op. natricis* specimens were deposited in the Australian National Insect Collection (ANIC), CSIRO, Canberra (voucher numbers ANIC 51-006454 to ANIC 51-006462).

Case 2. On June 12, 2018, a juvenile eastern bluetongue lizard (*Tiliqua scincoides*) that had been injured by a domestic cat (*Felis catus*), was collected by a wildlife rescuer from the garden of a private residence on Beach Road in Hackham, a suburban neighborhood of Adelaide, and taken to a veterinarian. Several suspected mites were collected, preserved in 95% ethanol, and donated to us to determine the species. These mites were determined to be five females and two males of *Op. natricis*, which were also subsequently deposited in ANIC (females - ANIC 51-0006463 to ANIC 51-006467; males - ANIC 51-006468 and ANIC 51-006469)

Case 3. On August 04, 2019, another *T. scincoides* that had been injured by a domestic cat, was collected by a wildlife rescuer from the garden of a private residence in Morphett Vale, a suburban neighborhood of Adelaide, and taken to a veterinarian. The lizard was suspected to be infested by *Op. natricis* and was treated accordingly. The kitchen tissue towels that were used as a substrate in the container the lizard was transported in were examined under a dissection microscope and one female of *Op. natricis*, was found and subsequently deposited in ANIC (ANIC 51-006513)

Summary of museum records and literature review. We found 42 records of *Op. natricis* specimens collected in Australia from the Northern Territory, Queensland, Victoria and South Australia (Table 3.1). Six lizard and 13 snake species have been reported as hosts of *Op. natricis* in Australia. Of the 17 records of lizards as hosts of this parasite 52.9% were animals in captivity and of the 24 records of snakes as hosts, 62.5% were animals in captivity. One record was of mite that were collected from the environment.

Discussion

Our record of *Op. natricis* from the mid-north of South Australia shows that this parasite is present on wild populations of *T. rugosa* and that it has become naturalized in the wild in Australia. *Ophionyssus natricis* was first reported from Australia by Womersley (1956), on the basis of specimens from snakes at Sydney, New South Wales, and Malanda, Queensland.

Domrow (1985) recorded it on the European green lizard, *Lacerta viridis*, in Melbourne, which suggests a possible pathway for the introduction of the mite into Australia. Domrow (1988) summarised previous records of *Op. natricis* on several species of snakes in Australia and pointed out that the records often came from hosts in captivity. Our summary of known hosts of this mite in Australia supports this conclusion (Table 3.1). In addition, most records of these

Table 3.1. Records of the snake mite (*Ophionyssus natricis*) from Australia, with notes on the record date, locality and the host (* explicitly stated that the specimens came from hosts in captivity; ** uncatalogued specimens; † the specimens are from the wild). ANIC – Australian National Insect Collection; QM – Queensland Museum; SAMA – South Australian Museum.

Host	Locality	Date	Reference
Lizards			
<i>Amphibolurus muricatus</i> *	South Australia Museum, Adelaide, SA	Aug. 1973	SAMA**
<i>Carlia longipes</i> , listed as <i>Leiolopisma fusca</i> *	Arnhem Land, NT	1948	SAMA**
<i>Chlamydosaurus kingii</i> *	Taronga Zoo, Sydney, NSW	3 Dec. 1999	ANIC 1133/1
<i>Lacerta viridis</i> *	Royal Melbourne Zoo, VIC	Sep. 1982	QM 93971 – 93980; Domrow (1985)
<i>Tiliqua rugosa</i> *	Adelaide University, SA	2 Aug. 1979	SAMA**
<i>Tiliqua rugosa</i> *	Lone Pine Koala Sanctuary, Brisbane, QLD	13 Oct. 1995	QM**; Walter and Shaw (2002)
<i>Tiliqua rugosa</i> *	Warrawong Wildlife Sanctuary, Mylor, SA	23 May 2007	SAMA**
<i>Tiliqua scincoides</i>	St. Albans, VIC	1 Jul. 1971	QM 93993
<i>Tiliqua scincoides</i>	Campbellfield, Melbourne, VIC		Hoser and Valentic (1996)
<i>Tiliqua scincoides</i>	Melbourne, VIC	Jul. 1999 – Apr. 2000	Watharow and Reid (2002)
<i>Tiliqua scincoides</i>	Capel Sound, Mornington Peninsula, VIC	1995	Hoser and Valentic (1996)
<i>Tiliqua scincoides</i>	Werribee, Skeleton Coast, VIC	15 May. 1999	QM**

Table 3.1 continued

<i>Tiliqua scincoides</i>	Campbellfield, Ivanhoe, Rockbank, Sydenham, Templestowe and Werribee, VIC	Jul. – Sep. 1999	Walter and Shaw (2002)
<i>Tiliqua scincoides</i> *	Largs North, SA	2 Aug. 1978	SAMA**
<i>Tiliqua scincoides</i>	Melbourne, VIC	1990 – 1993	Turner (2010)
Unspecified lizards *	Adelaide College of Advanced Education, SA	30 Apr. 1980	SAMA**
Unspecified lizards and snakes	Adelaide, SA	12 Jul. 1964	QM 93960 - 93970
Snakes			
<i>Acanthophis antarcticus</i> *	Adelaide, SA	12 Aug. 1971	Domrow (1985)
<i>Acanthophis antarcticus</i> *	Bundaberg, QLD	Jun. 1989	QM 93986 - 93990
<i>Antaresia children</i> , listed as <i>Liasis childreni</i>	Townsville, QLD	24 Jul. 1966	QM 93957 - 93959
<i>Antaresia children</i> , listed as <i>Liasis childreni</i>	Townsville, QLD	Dec. 1982	QM 93991
<i>Antaresia children</i> , listed as <i>Liasis children</i> *	Royal Melbourne Zoo, VIC		QM 93992
<i>Aspidites ramsayi</i> *	South Australia Museum, Adelaide, SA	Dec. 1980	SAMA**
<i>Hoplocephalus bitorquatus</i> ¹	Namoi River, NSW	Nov. 2001 – Mar. 2002	Fitzgerald et al. (2010)
<i>Hoplocephalus stephensii</i> *	Brisbane, QLD		ANIC**
<i>Liasis fuscus</i> *	Lone Pine Koala Sanctuary, Brisbane, QLD	13 Oct. 1995	QM**
<i>Morelia viridis</i> *	Royal Melbourne Zoo, VIC	21 Jun. 1971	QM**
<i>Morelia viridis</i> *	Mareeba, Far North, QLD	3 Jan. 1999	QM**
<i>Morelia spilota metcalfei</i> *	Warrawong Wildlife Sanctuary, Mylor, SA	23 May 2007	SAMA**

Table 3.1 continued

<i>Morelia spilota variegata</i>	Malanda, QLD	27 Oct. 1952	SAMA J16096
<i>Morelia spilota variegata</i> *	Royal Melbourne Zoo, VIC	Jun. 1983	QM 93997
<i>Notechis scutatus</i>	Campbellfield, Melbourne, VIC	1996	Hoser and Valentic (1996)
<i>Pseudechis porphyriacus</i> *	Brisbane, QLD	1982	Domrow (1985)
<i>Pseudonaja textilis</i> , listed as <i>Demansia textilis</i>	Sydney, NSW	Dec. 1952	SAMA J16086 - J16092
<i>Python molurus bivittatus</i> *		26 Dec. 1956	SAMA J16098 – J16110
<i>Tropidechis carinatus</i>	North Coast, NSW		QM 93981 - 93985
Unspecified snake *	Westmead, NSW	1 Sep. 2006	ANIC**
Unspecified snake *	Warrnambool, VIC	Oct. 1974	QM 93996
Unspecified snake *	Adelaide		SAMA J16093 - J16094
Unspecified snake	Cairns, QLD	Oct. 1952	SAMA J16095
Unspecified snake skin *	Queensland Institute of Medical Research, QLD	Jun. 1967	QM 939954- 93995
Environment			
Moss ¹	Mt. Remarkable, Melrose, SA	2 Sep. 1960	SAMA J16083 - J16085

mites on free-living reptiles involve animals that were found in the proximity of suburban areas, so the likelihood of these reptiles being escaped pets or that the mite was spread by escaped pets cannot be excluded. Wildlife Health Australia (2018) stated that “there are no confirmed reports of snake mite in free-ranging reptiles in Australia”. The nearest human settlement to our study site in the mid north of South Australia is a small village (Robertstown, 2006 population 318), ca. 17 km away and the nearest town (Eudunda, 2006 population 640) is ca. 35 km away, so the presence of the mite in the study site is unlikely due to a close proximity to pet reptiles. In addition, the likelihood that the infested lizard was a released pet can be ruled out since the lizards we collected are marked with unique toe-clips and have been used in consecutive years. Our conclusion that this mite has become naturalised in Australia is further supported by the mite specimens that were collected from moss on Mt. Remarkable near Melrose in South Australia (Table 3.1), and the instances of infestation of the endangered pale-headed snake (*Hoplocephalus bitorquatus*) in a pastoral area near Pillaga in New South Wales (Fitzgerald et al. 2010).

Ophionyssus natricis can readily be accidentally introduced into new localities along with their hosts. The larvae of *Op. natricis* are fairly sedentary, while the protonymph, deutonymph and adult (male and female) life stages are more active (Camin 1953). Only the protonymph and adult life stages are parasitic and as they are photophobic tend to attach under scales of the host (Camin 1953, Wozniak and DeNardo 2000). They can be easily overlooked, particularly if they are un-engorged. Case in point, the first record of *Op. natricis* in New Zealand was from dead juvenile eastern blue-tongued lizards that were obtained from Melbourne Zoo by Wellington Zoo in New Zealand (Heath 1986). It is therefore imperative that wildlife rescuers in areas where this mite is present should take this into consideration when reptiles are relocated, and ideally, such reptiles should be treated for possible mite infestation prior to release.

Interestingly, our observations in the mid-north of South Australia were from a xeric environment, which indicates that the distribution of *Op. natricis* can extend beyond high rainfall parts of Australia. *Ophionyssus natricis* is generally considered sensitive to desiccation and eggs and larvae suffer high mortality in conditions with a relative humidity lower than 75% (Camin 1953, Wozniak and DeNardo 2000). The distribution in drier areas is of concern regarding the potential for spread of this invasive species.

Our findings also highlight the importance of continued monitoring of *Op. natricis* in the wild.

During the 2017 field season it was noted that what appeared to be snake mite were quite common on sleepy lizards at the study site surrounding the Bunday Lutheran church ruins (Sih 2017, *personal observation*). The study site typically experiences rainfall of less than 300 mm during late winter to early summer (August to December) (Kerr and Bull 2004) and the only record of *Op. natricis* at the study site in 2019 was in mid-August when the study site was experiencing a severe drought, which lasted throughout the time the study was conducted. It is therefore possible that no other instances of mite infestation were found at the study site because the mite population in this area may have been drastically reduced or may even have died out due to desiccation stress. It is therefore recommended that follow up investigations be done during non-drought periods to verify the status and distribution of this snake mite population.

Herein we describe instances of *Op. natricis* parasitising skinks from South Australia and summarised the known hosts and distribution records of this mite in Australia. Our observation from the mid-north of South Australia is a new distribution record for *Op. natricis* in Australia and appears to be the first confirmed instance of these mite infesting a sleepy lizard in the wild. Additional surveys are needed to confirm the extent of the distribution of this mite in Australia, and we encourage the submittal of mite specimens for species verification and the reporting of the host species and localities where *Op. natricis* are found in Australia.

Disclosures

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Additional instances of snake mite (*Ophionyssus natricis*) parasitism on sleepy lizards (*Tiliqua rugosa*) in South Australia

Abstract

The snake mite, *Ophionyssus natricis* (Acari: Macronyssidae) is an ectoparasite of veterinary significance because infestations can cause abnormal shedding and anaemia in its hosts. The snake mite has also been implicated in the transmission of pathogens. In this study 59 sleepy lizards (*Tiliqua rugosa*) were caught at a study site in the vicinity of Mt. Mary in the Mid North region of South Australia and Hallett Cove Conservation Park in Adelaide and examined for the presence of snake mite. About 10% of the lizard were found to be infested by snake mite. This study confirms that sleepy lizards are competent hosts of this ectoparasite and presents additional distribution records for South Australia.

Introduction

The snake mite, *Ophionyssus natricis* (Gervais 1844), was originally described from specimens collected in Europe, and subsequently has been reported from parts of Africa, the Americas, Asia, and Oceania (Gervais 1844, Yunker 1956, Miranda et al. 2017). This parasite is usually associated with captive snakes and lizards, both in the pet trade and for institutions such as zoological gardens (Frank 1981, Wozniak and DeNardo 2000). However, specimens have been collected from free-living reptiles in parts of Africa (Yunker 1956, Till 1957), Asia (Simonov and Zinchenko 2010), Australia (Domrow 1985, Walter and Shaw 2002, Norval et al. 2020), Europe (Hassl 2016, Mendoza-Roldan et al. 2019) and North America (Goldberg and Bursey 1991).

Heavy infestation by *O. natricis* can cause abnormal shedding and/or anaemia in its host (Frank 1981, Lane and Mader 1996, Wozniak and DeNardo 2000), and because these mite have been implicated in the spread of pathogens, such as the causative agent of body inclusion disease in boids (Chang and Jacobson 2010, Simard et al. 2020), which can be fatal to the host, the presence of this mite in the wild outside of its native range is of significant conservation and veterinary concern, and their occurrence should be monitored and reported. In a previous study we showed that this exotic parasite species also occurs in the wild in parts of Australia (Norval et al. 2020). We now report on additional instances of *O. natricis* parasitism on sleepy lizards, *Tiliqua rugosa* (Gray 1825), in the wild in South Australia.

Materials and methods

In the period October 2020 to January 2021, 61 *T. rugosa* were collected on an *ad hoc* basis from the area surrounding a study site near Mt. Mary in the Mid North region of South Australia (33°55'40.7"S 139°17'07.8"E), about 150 km northeast from Adelaide (hereinafter referred to as site 1), as well as from Hallett Cove Conservation Park (35°04'23.9"S 138°29'55.4"E), situated about 22 kilometres south of the centre of Adelaide City (hereinafter referred to as site 2; Fig. 3.4). The snout-vent length (measured from the tip of the snout to the posterior margin of the cloaca) of every lizard was measured with a transparent plastic ruler to the nearest mm. Each lizard was then examined for the presence of *O. natricis*. The lizards collected in early October and early November 2020 were examined for *O. natricis* with a fine paintbrush as described in Norval et al. (2020). All the other lizards were examined in a similar manner, but a soft toothbrush was used instead of a paintbrush, and the ethanol that was used for rinsing

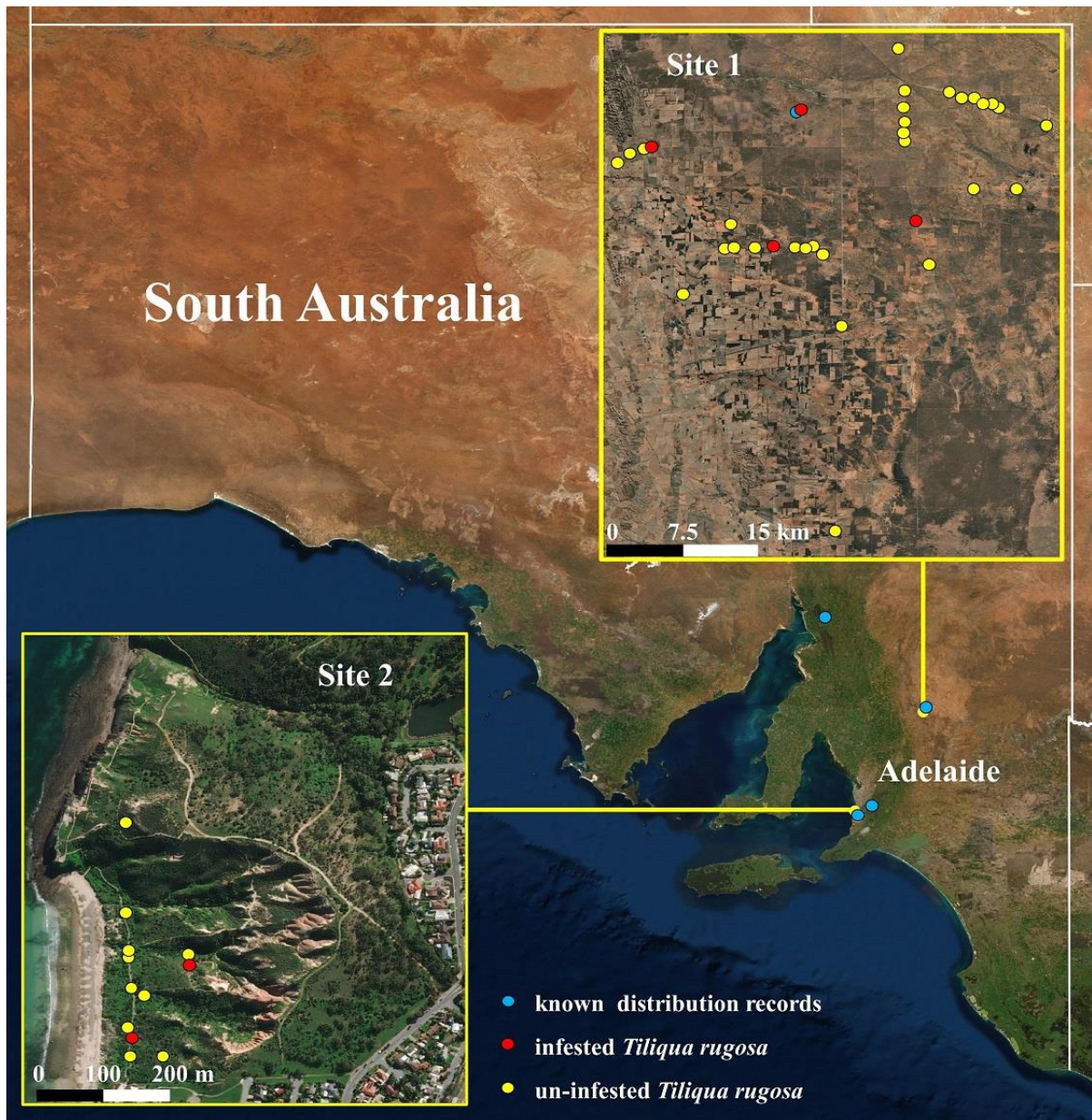


Figure 3.4. The known distribution records of the snake mite (*Ophionyssus natricis*) from the wild in South Australia, and the additional records from the two sites used in this study.

out the plastic container was collected in a 50 ml plastic jar instead of pouring it through a fine fabric filter. The collected ethanol was then subsequently poured into a petri dish and examined under a dissection microscope. The lizards collected in the period November 2020 to January 2021 were kept in captivity as part of an unrelated study and were returned to the wild at the point of capture in March 2021. All the other lizards were released back into the wild after morphometric measurements were taken and the lizards were examined for snake mite.

Mites were identified according to the descriptions in Camin (1953), Domrow (1988) and Moraza et al. (2009). The diagnostic character states in the female of *O. natricis* are the

fragmentation of the dorsal shield to form a large anterior (podonotal) shield with 10 pairs of setae, two pairs of small mesonotal plates immediately behind the podonotal shield, and a minute pygidial shield on the posterior dorsal idiosoma. The pygidial shield is irregular in shape and usually aseptose but sometimes carries one or two setae, the peritreme reaches forward to the posterior margin of coxa II, and the genital setae are inserted in soft integument, not on the genital shield. The male of *O. natricis* has a large dorsal shield with three successively smaller lobes, covering almost all of the podosoma and most of the opisthosoma, with 11 to 17 pairs of setae.

We used the statistics software package Prism 8 (Graphpad Software, San Diego, California, USA) for statistical analyses. Data transformation did not correct the distribution of the data from site 2, so the Mann-Whitney U test, with $\alpha = 0.05$, was used for comparing the SVLs of the lizards from the two sites.

Results

The 46 sleepy lizards collected at site 1 ranged in SVL from 265 mm to 347 mm, with a mean and standard deviation of 303.2 mm and 19.6 mm respectively. The SVL of the 15 sleepy lizards collected at site 2 ranged from 184 mm to 267 mm, with a mean and standard deviation of 242.3 mm and 23.1 mm respectively. Even though *T. rugosa* from southern regions of their range tend to be smaller than conspecifics from more northern regions (Sharrad and King 1981), all the lizards caught at site 2 were considered juveniles (SVL < 200 mm) and subadults (SVL < 270 mm) (Bull 1995), and were significantly smaller than those caught at site 1 ($U = 1, P < 0.0001$).

Four (8.7%) of the lizards caught at site 1 and two (13.3%) caught at site 2 were found to be infested by *O. natricis*. The infested lizards from site 1 ranged in SVL from 265 mm to 314 mm, with a mean and standard deviation of 286.5 mm and 20.34 mm respectively (Table 3.2). Females, males, protonymphs and a deutonymph of *O. natricis* were collected (Table 3.2). Details of the specimens collected, and their collection registration numbers, are presented in Table 3.3.

Discussion and conclusion

This study expanded the known geographic distribution of *O. natricis* in South Australia, and

presented additional instances of *O. natricis* parasitism on *T. rugosa* in suburban and rural habitats. The presence of this parasite in Hallet Cove Conservation Park, which is within a suburban neighbourhood, is not completely unexpected. Formerly *O. natricis* has been found parasitizing lizards and snakes in localities neighbouring suburban parts of Melbourne in Victoria, Australia (Watharow and Reid 2002, Turner 2010). *Ophionyssus natricis* is commonly associated with pet reptiles and when infested pet lizards and snakes escaped or are released into the wild they can transmit this parasite to free-living reptiles. Infested terraria substrates and/or furnishing that is not sterilized before being discarded is another potential source for transmitting this mite. The occurrence of *O. natricis* in the wild in arid parts of South Australia is more unusual since the localities where they have been found are not in the proximity of large towns or cities (Norval et al. 2020). As these areas are mainly used for agricultural activities it is possible that the snake mite was introduced along with lizards and/or snakes that were either intentionally or accidentally taken to this area. Our observations of *O. natricis* infesting sleepy lizards that were caught in the vicinity of the Bunday Lutheran Church ruins in 2017, 2019 and 2020 indicates that it is possible for this mite to persist for multiple years in some arid conditions (Norval et al. 2020; herein).

Table 3.2. The collection date, collection locality, and snout-vent length of the sleepy lizards (*Tiliqua rugosa*) that were infested by snake mite (*Ophionyssus natricis*), and the numbers of female, male, deutonymph and protonymphs of this mite that were collected.

Date	Locality	SVL	Female	Male	Deutonymph	Protonymph
2 Oct. 2020	Study site 1	282				1
2 Nov. 2020	Study site 2	255				1
17 Nov. 2020	Study site 1	314	3	3	1	1
30 Nov. 2020	Study site 1	265				1
23 Dec. 2020	Study site 2	249		1		1
8 Jan. 2021	Study site 1	285	1			

Ophionyssus natricis is a typical nest parasite, and although the protonymphs, deutonymphs and adults are mobile, they tend to be nidicolous due to their need for humid off-host microhabitats to avoid desiccation and being negatively phototactic (Camin 1953). The importance of the behaviour of the host in the maintenance of a nest parasite is illustrated by the biology of kangaroo soft tick, *Ornithodoros gurneyi* Warburton, 1926, and the behaviour of red kangaroos, *Osphranter rufus* (Desmarest 1822) and common wallaroos, *Osphranter*

Table 3.3. Records of the snake mite (*Ophionyssus natricis*) from free-living reptiles and the environment in South Australia, with notes on the record date, locality and the host [* - herein; ** - unreported new record; * - reported on in Norval et al. (2020)]. ANIC – Australian National Insect Collection; SAMA – South Australian Museum.**

Host	Locality	Date	Reference
Lizards			
<i>Tiliqua scincoides</i> ***	Hackham, Adelaide	12 Jun. 2018	ANIC 51-0006463 to ANIC 51-006469
<i>Tiliqua scincoides</i> ***	Morphett Vale, Adelaide	4 Aug. 2019	ANIC 51-006513
<i>Tiliqua rugosa</i> ***	Bundey, Mid North	19 Sep. 2017	ANIC 51-006454 to ANIC 51-006462
<i>Tiliqua rugosa</i> *	Bundey, Mid North	2 Oct. 2020	ANIC 51-006530
<i>Tiliqua rugosa</i> *	Hallett Cove, Adelaide	2 Nov. 2020	ANIC 51-006531
<i>Tiliqua rugosa</i> *	Beatty, Mid North	17 Nov. 2020	ANIC 51-006520 to ANIC 51-006527
<i>Tiliqua rugosa</i> *	Bright, Mid North	30 Nov. 2020	ANIC 51-006528
<i>Tiliqua rugosa</i> *	Hallett Cove, Adelaide	23 Dec. 2020	ANIC 51-006566 and ANIC 51-006567
<i>Tiliqua rugosa</i> *	Bower, Mid North	8 Jan. 2021	ANIC 51-006529
Snakes			
<i>Pseudonaja textilis</i> **	Upper Sturt, Adelaide	20 Dec. 2020	ANIC 51-006532 to ANIC 51-006565
Environment			
Moss ***	Mt. Remarkable, Melrose	2 Sep. 1960	SAMA J16083 to J16085

robustus (Gould 1841), its primary hosts. Both these macropods have a very wide distribution in Australia, but the tick's distribution is limited to open arid regions where the kangaroo hosts have to make frequent use of the same resting places due to limited numbers of suitable trees, shrubs and caves, thereby ensuring a frequent enough blood meal to sustain the ticks (Browning 1962, Doube 1972). Yunker (1956) hypothesised that competent hosts for *O. natricis* are expected to be reptile species that are abundant or gregarious, and that occupy small home ranges and often utilise the same refuges. Sleepy lizards would therefore be suitable hosts. They occupy stable overlapping home ranges that are relatively small compared to those of similar-sized carnivorous lizards (Bull 1978, Satrawaha and Bull 1981, Bull 1987, Bull and

Freake 1999). Within their home ranges, sleepy lizards utilise numerous refuges (Leu et al. 2010, 2011) and neighbours may periodically utilise the same refuges asynchronously (Spiegel et al. 2015). During the mating season males and females form stable pair bonds (Fig. 3.5), and usually shelter together within the same refuge (Kerr et al. 2003). Neonates remain within the territory of the mother (Bull and Baghurst 1998) and in some instances may share the same refuge with her (Fig. 3.6). Multiple individuals may also hibernate together (Fig. 3.6). The social behaviour of this lizard is therefore very suitable for the transmission of *O. natricis* to additional potential hosts and different localities. Studies involving the reptile ticks *Amblyomma limbatum* Neumann 1899, and *Bothriocroton hydrosauri* (Denny 1843) demonstrated that the refuge use of sleepy lizards play an important role in the maintenance of the ticks and their dispersal (Leu et al. 2010). The role that refuge use by sleepy lizards, or other reptilian hosts, play in maintenance and spread of *O. natricis* is poorly understood and warrants empirical studies.



Figure 3.5. During the late austral spring to early summer sleepy lizards (*Tiliqua rugosa*) form pairs that usually remain in close proximity with each other until after mating.

If sleepy lizards are such suitable hosts, why are not more of them found to be infested by *O. natricis*? As with other ectoparasites, the survival of *O. natricis* depends not only on finding competent hosts, but also on the off-host environment. The eggs, larvae, replete protonymphs, deutonymphs and egg-laden females of *O. natricis* require dark humid refuges in which moulting and/or oviposition can occur, and high temperatures and low relative humidity (< 75%) causes high mortality in the eggs and larvae of this mite (Camin 1953, Wozniak and DeNardo 2000). Our records from Australia are not the only instances of this mite occurring in arid habitats. Domrow (1974) collected females of *O. natricis* from an *Antaresia childreni* (Gray 1842) (reported as *Liasis childreni*) that was caught in the wild in Townsville in northeastern Queensland, Australia, and Yunker (1956) reported *O. natricis* parasitism on



Figure 3.6. (A) Three adult sleepy lizards (*Tiliqua rugosa*) were found on 8 June 2019 hibernating together under a discarded sheet of corrugated iron, (B) and on 26 September 2019 an adult female and a juvenile (most likely her offspring) were found sheltering for the night under the same sheet of corrugated iron.

several snake species in Egypt. This indicates that at least in some arid environments the off-host environment is suitable for the survival of *O. natricis*. A possible determining factor is the

proximity to a water source. In all the instances in our study where *O. natricis* was recorded in the arid environment of site 1, the lizard was caught in the vicinity (< 100 m) of a man-made earth dam. The potential roles that proximity to water sources plays in the distribution of *O. natricis* in arid environments therefore needs to be verified through empirical studies and should include research into the more sensitive to desiccation off-host stages and the environmental conditions that enable their persistence in arid regions.

Ophionyssus natricis is negatively phototactic, and as a result they tend to attach and feed while concealed under the scales of their host (Camin 1953, Wozniak and DeNardo 2000). The dorsal and lateral scales of a sleepy lizard's body are large and posteriorly raised (Andrews and Petney 1981), so is a more suitable host for this mite than other Australian skinks with smaller and tighter packed scales. In our study we only record instances of *O. natricis* parasitizing adults and subadults of *T. rugosa* and we did not find any on the small number of juveniles that we examined. This is likely due to our small sample sizes rather than the lack of suitable attachment sites on the smaller lizards. Case in point, a female sleepy lizard was collected on 30 November 2020 and while in captivity, she gave birth to two neonates on 25 February 2021. On the same day the young were removed from the mother's enclosure and housed individually in their own enclosures until 4 March 2021 when they were prepared to be released back into the wild. Several adults of *O. natricis* were collected from both the neonates, and since the one lizard was atypically small (Fig. 3.7), this observation indicates that *O. natricis* can attach and feed on sleepy lizard neonates and juveniles. As *O. natricis* can attach and feed under the large scales of sleepy lizards of all age classes, this lizard species can be an important reservoir for the maintenance of this mite in suitable habitats.

Ophionyssus natricis is a possible vector of several pathogens of snakes, and squamates infested by large numbers of this mite may suffer from abnormal shedding and/or anaemia, which can result in mortality (Frank 1981, Lane and Mader 1996, Wozniak and DeNardo 2000, Šlapeta et al. 2018). The physiological impact of *O. natricis* on *T. rugosa* is not known. Studies involving ticks found that high infestations reduce the running speed of juvenile sleepy lizards and decrease the general activity levels of subadults and adults (Main and Bull 2000), and reduce lizard survival (Jones et al. 2016). If *O. natricis* can be maintained under the scales of sleepy lizards, it is possible that this mite may have similar effects on these lizards, especially in juveniles.



Figure 3.7. A typically sized neonate of a sleepy lizard (*Tiliqua rugosa*) and an atypically small sibling born in captivity on 25 February 2021. Both these lizards became infested by snake mite (*Ophionyssus natricis*) from the mother.

Tiliqua rugosa is commonly parasitised by the reptile ticks *Amblyomma albolimbatum* Neumann, 1907, *A. limbatum* and *B. hydrosauri*, and these tick species have been found to be infected by pathogens such as the haemogregarine blood parasite *Hemolivia mariae* Smallridge and Paperna, 1997 and several *Rickettsia* species, including the causative agent of Flinders Island spotted fever, *Rickettsia honei* Stenos, Roux, Walker, and Raoult, 1998 (Smallridge and Paperna 1997, Stenos et al. 2003, Staines et al. 2020, Tadepalli et al. 2021). The possibility of *O. natricis* becoming a vector for these and other pathogens, and the potential role of this mite in the transmission of diseases in wildlife warrants further study.

In this study we reported on instances of snake mites infesting free-living sleepy lizards in South Australia, and present new distribution records in this state. These records indicate that additional surveys are needed in suitable habitats to determine the extent of the distribution of this exotic invasive parasite in South Australia. It is possible that the collection technique we used failed to recover all the mites that may have been present, and the numbers of infested individuals may therefore be an underestimate. If possible, future surveys should therefore utilize other techniques, such as thorough rinsing with detergent, to increase the accuracy and effectiveness of detecting the presence of these mite. Because this mite and the pathogens it may harbour can be of veterinary importance, we also recommend that studies be undertaken

to determine what risks this mite present to native Australian wildlife.

Disclosures

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Chapter 4

A mammal tick with a taste for lizard blood: parasitism by the kangaroo soft tick, (*Ornithodoros gurneyi*) on sleepy lizards

(*Tiliqua rugosa*)

Note to examiners

The kangaroo soft tick (*Ornithodoros gurneyi*) is a tick species that is generally restricted to arid habitats and as a result little is known about its natural hosts. Earlier records of this tick from Mt. Mary presented an opportunity to address a few unanswered questions pertaining to this tick species. Since this locality is situated further south than what is currently considered to be the southern distribution range limit of this tick, does it occur at the Bunday Bore study site, and if so, to what extent? The kangaroo soft tick is considered a mammal tick, so another question is whether these ticks can moult after feeding on sleepy lizards. In Chapter 4, I report on the results of studies I did to determine the distribution of this tick in the vicinity of the Bunday Bore study site, the dynamics of its parasitism on sleepy lizards, and whether sleepy lizards were competent hosts of this parasite. This chapter is a manuscript that was published in the journal *Ticks and Tick-borne Diseases* and it is therefore formatted according to the specifications of the journal. The referencing style was however changed to make it consistent with that of the other parts of my thesis. The numbering of the figures and tables were changed for clarification and so that they appear chronologically in the chapter. I took the photographs and micrographs and therefore own the copyright.

Abstract

The kangaroo soft tick (*Ornithodoros gurneyi*) inhabits xeric habitats of Australia and as a result little is known about the natural hosts of the different instar stages of this tick. Large macropods are inferred as the kangaroo soft tick's principal hosts since all life stage instars inhabit the loose soil of kangaroo wallows. These ticks have been reported to parasitise bearded dragons (*Pogona barbata*) and sleepy lizards (*Tiliqua rugosa*), however it is not known whether these lizards are competent hosts of this tick. In this study the prevalence and intensity of *O. gurneyi* parasitism on *T. rugosa* was examined by sampling lizards at a long-term study site spanning a precipitation-induced ecological gradient. The competence of *T. rugosa* as a host for *O. gurneyi* was explored by keeping larvae and nymphs of *O. gurneyi*, that had fed on *T. rugosa*, in captivity to determine if they will survive and moult. We found that *O. gurneyi* occurs throughout the study site, but that they were seldom recorded to parasitise *T. rugosa*. Still, it was noted that *T. rugosa* is a competent host of *O. gurneyi*. The results of this study indicate that kangaroos are not the only competent hosts of *O. gurneyi* and raises the question of whether *T. rugosa* and other lizards may play a role in the dispersal of these ticks.

Introduction

The ecology of soft ticks (Argasidae) is generally not as well studied as those of the better known hard ticks (Ixodidae), which often inhibits their inclusion in discussions on tick ecology (Randolph 2004). This lack of information is in part due to the fact that these ticks are seldom observed on their hosts because they usually inhabit the nests, roosts or other resting places of their hosts and feed rapidly while the host is resting, after which they detach and seek shelter (Hoogstraal 1985). This is most certainly the case for the kangaroo soft tick, *Ornithodoros gurneyi* Warburton, 1926 an argasid that occurs in xeric parts of Australia, and is therefore rarely encountered and as a result its natural history is not well-documented (Doube 1972, Barker and Walker 2014).

The geographic distribution of a tick species is limited by the abundance of competent host species and a suitable off-host environment (Léger et al. 2013, Barker and Walker 2014). *Ornithodoros gurneyi* has not been found on any macropods, but it has been inferred that red kangaroos (*Osphranter rufus*) and common wallaroos (*Osphranter robustus*) are their primary hosts since all post-embryonic instars (i.e. larvae, nymphs and adults) of this tick can be found in the loose soil of shaded wallows (also called kangaroo camps or scrapes; Fig. 4.1) and caves frequented by these marsupials during the hottest part of the day (Doube 1975a, Barker and Walker 2014). Browning (1962) and Doube (1972) found that *O. gurneyi* was present in localities where kangaroos have to make frequent use of the same wallows due to limited numbers of suitable trees and shrubs, and that the ticks are absent in more wooded areas in the same localities where potential hosts infrequently rested due to an excess of suitable sites. As a result the distribution of *O. gurneyi* is believed to be limited to the xeric open inland plains and hills of Australia (Browning 1962, Doube 1972).

Ticks are usually morphologically and behaviourally adapted to locate and feed efficiently from specific hosts, resulting in degrees of host specificity, especially in adult instars (Oliver 1989, McCoy et al. 2013). A fundamental aspect of the ecology of a tick is therefore an understanding of which species the different instars (i.e. life stages) of the tick parasitize. What natural hosts *O. gurneyi* infest is poorly understood. It is inferred that all postembryonic instars of *O. gurneyi* parasitise large macropods, but to date no specimens of this tick have been found attached to these hosts in the wild (Doube 1975a, Barker and Walker 2014). The kangaroo soft tick has been reported to opportunistically bite humans and domestic animals such as cattle



Figure 4.1. A kangaroo wallow under an oilbush (*Geijera linearifolia*). Note the loose soil and evidence of how the kangaroo excavates a hollow area to lay in.

(*Bos taurus*), dogs (*Canis familiaris*), horses (*Equus caballus*) and pigs (*Sus scrofa*) (Henry 1938, Wilkinson 1958, Roberts 1970, Doube 1975a, Norval et al. 2021), but to the best of our knowledge the only confirmed instances of *O. gurneyi* parasitism on native mammals in the wild are the larvae that were collected from a greater bilby (*Macrotis lagotis*) in Davenport Downs, Queensland, and nymphs that were taken from a short-beaked echidna (*Tachyglossus aculeatus*) on Kangaroo Island, South Australia (Australian National Insect Collection; ANIC 48 000 205 and ANIC 48 000 292). In addition, instances of parasitism on the bearded dragon (*Pogona barbata*, reported as *Amphibolurus barbatus*) and the sleepy lizard (*Tiliqua rugosa*) have also been reported (Doube 1975a, Sharrad and King 1981). However, it is not known if *O. gurneyi* can moult to the next life stage after feeding on reptilian hosts (i.e. whether reptiles are competent hosts).

In the early 1970's Sharrad (1979) recorded six instances of immature instars of *O. gurneyi* parasitizing sleepy lizards that were caught in the vicinity of Mt. Mary in the Mid North region of South Australia. This locality is south of what is considered the southern distribution limit of *O. gurneyi* on mainland Australia (Fig. 4.2). The purpose of this study was therefore to investigate the extent of the distribution of *O. gurneyi* at a study site that is situated ca. 5 km northwest of Mt. Mary. We also aimed to determine the prevalence of *O. gurneyi* as a parasite of sleepy lizards at this study site and to establish if sleepy lizards are competent hosts of this

tick.

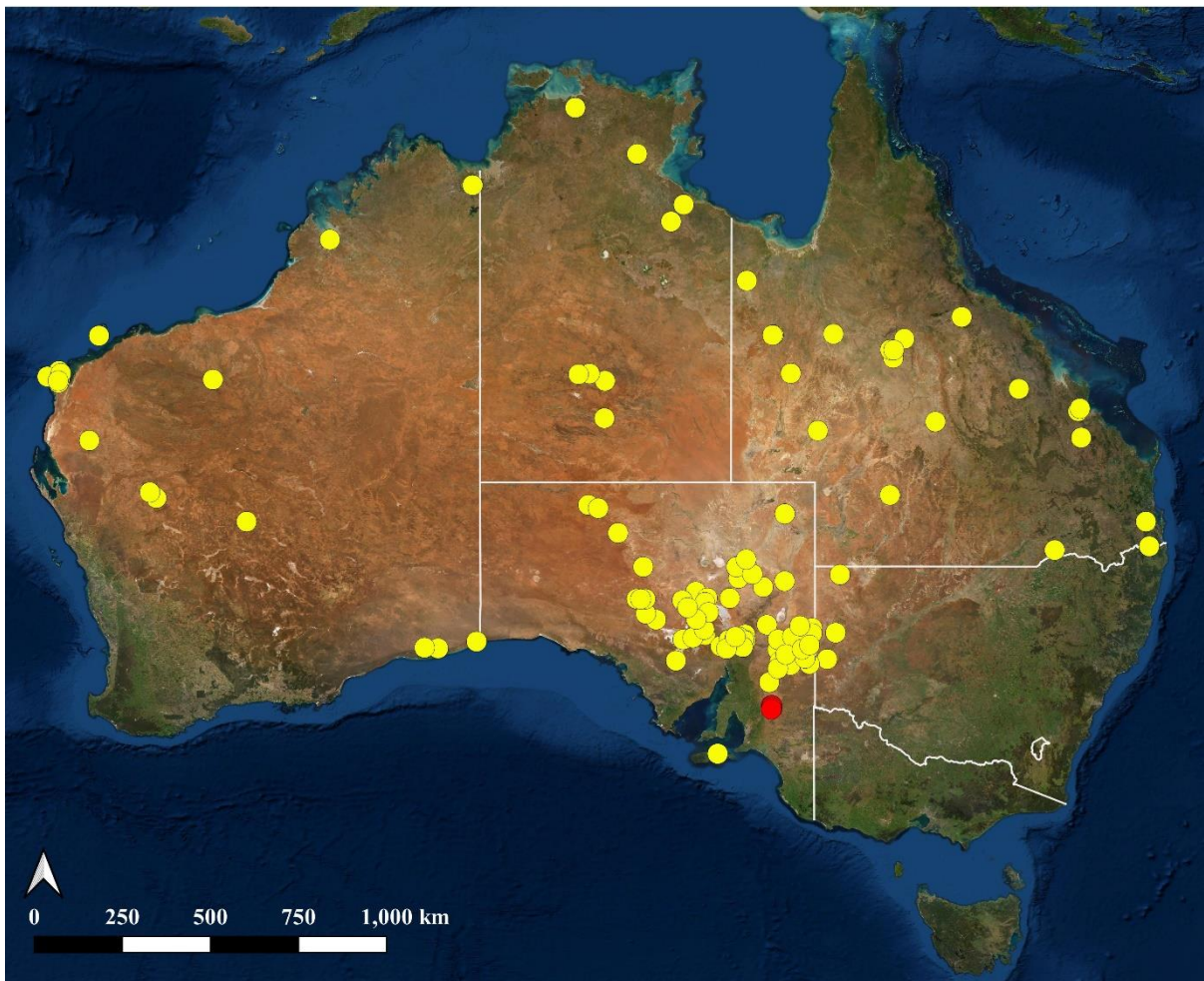


Figure 4.2. Localities where kangaroo soft ticks (*Ornithodoros gurneyi*) have been recorded at in Australia (yellow dots) and where this species was found near Mt. Mary in the Mid North region of South Australia (red dot).

Material and methods

Study site: The study was conducted at the Bunday Bore Station study site, ca. 160 km north of Adelaide, in the Mid North region South Australia, as well as some of the surrounding areas as part of an ongoing long-term study on sleepy lizards and their ticks, initiated by the late Prof. Michael Bull (Godfrey and Gardner 2017). The climate of this region is semi-arid, with an average annual rainfall of less than 250 mm, of which most usually fall during the austral winter (Smyth 1973). The habitat of the study site is a mosaic of large areas that have been cleared for agriculture and grazing, interspersed by remnant fragments of the original grassy arid mallee (*Eucalyptus* spp.) scrub and *Maireana sedifolia* shrubland that historically dominated the south-western and north-eastern parts respectively (Jessup 1948, Smyth 1973).

Tick description and identification: All the ticks were identified as *O. gurneyi* based on the following morphological features; the nymphs resemble the adults but lack a genital aperture (Roberts 1970). The body is oval, bluntly attenuated anteriorly and broadly rounded posteriorly (Warburton 1926, Roberts 1970). The integument mammillate, with dorsal depressions and deep ventral grooves (Warburton 1926, Roberts 1970). Eyes absent and numerous short white hairs scattered over the integument, and ventrally extending to the camerostome and coxae, where they are long and curved (Warburton 1926, Roberts 1970). The camerostome is deep and the hood distinct, separated from the cheeks by a furrow (Warburton 1926, Roberts 1970). The cheeks are large and mammilate basally, and divided into a number of long finger-like processes (Warburton 1926, Roberts 1970, Barker and Walker 2014). The hypostome almost devoid of teeth. All the legs with prominent subapical dorsal protuberance and dorsal humps, which are mild on tarsus 3 and restricted to one on leg 4 on the metatarsus (Warburton 1926, Roberts 1970, Barker and Walker 2014). The dorsal depressions and deep ventral grooves are present in the larvae.

Distribution of *Ornithodoros gurneyi* in and around the study site: We searched for soft ticks in kangaroo wallows as previously done by Browning (1962) and Doube (1975b) because searching for soft ticks off the host in the natural environment is a more reliable method than searching on potential hosts for determining their distribution. The kangaroo wallows were located on an *ad hoc* basis while traveling along the roads in and around the study site. Only wallows with a loose substrate (i.e. uncompacted) were investigated. First, the GPS location of the wallow was recorded with a Garmin eTrex ® 10 GPS receiver and notes were made of the species of shrub, tree or manufactured structure that overshadowed it. Next, a 395 mm diameter garden sieve (5 mm × 5 mm mesh size) was placed into a 400 mm diameter water tank screen (1 mm × 1 mm mesh size). The garden sieve was then filled with soil from inside the kangaroo wallow and then lifted to approximately the height of the brim of the tank screen and shaken until only stones and other large objects remained inside the sieve. The sieve was then visually examined for any ticks after which the content was returned to the wallow. Next, the tank screen was held over a plastic basin and shaken to separate fine and coarse soil particles. These particles were then separately visually inspected for active ticks. When ticks were found, they were collected and placed inside empty 5 ml plastic vials. At the end of the day the ticks were either transferred to aerated jars, with 6 cm deep fine sand inside them, for future studies or preserved in 5 ml plastic vials filled with 95% ethanol. The open software QGIS (Version 3.16.3; available at <http://qgis.osgeo.org>) was used for creating the study site and distribution

maps.

***Ornithodoros gurneyi* parasitism on *Tiliqua rugosa*:** To investigate the prevalence of *O. gurneyi* parasitism on *T. rugosa*, lizards were caught along roads in and surrounding the study site during the period late August 2017 to early January 2021, primarily in the period late August to mid-December when they are expected to be most active (Bull 1987, Bull et al. 1991, Firth and Belan 1998). The lizards were located by driving slowly along the dirt roads (i.e. established transects) passing through the study site and searching for lizards crossing the road or actively walking in open areas between vegetation at the sides of the road (Bull 1987). On cooler days and periods when the lizards were likely to be inactive, we searched for lizards by looking under discarded sheets of corrugated iron that can be found at some localities in the study site. The GPS location of each capture locality was recorded with a Garmin Etrex 10 GPS receiver. The lizards were captured by hand and individual lizards were identified by their unique toe-clips, and newly captured lizards were individually marked by toe-clipping as part of the ongoing long-term study on this species initiated in 1982 at this study site (Bull 1987, Godfrey and Gardner 2017). The snout-vent length (SVL) and of every captured lizard was measured (to the nearest mm) with a transparent plastic ruler, and the body mass (BM) was then measured (to the nearest 50 g) with a Pesola® Macro-Line spring scale. The area around the cloaca, in the ear and nasal openings, and the body of the animal, were then visually examined for attached soft ticks before releasing the lizard back into the wild at the point of capture. If present, the ticks were counted and their attachment sites were noted, after which they were removed with a pair of fine forceps. With the exception of a few *O. gurneyi* specimens that were to be used in moulting experiments, all the collected tick specimens were preserved in 95% molecular grade ethanol. The keys in Roberts (1970) were used to verify the species of the sampled ticks.

Moulting experiment: To determine whether *O. gurneyi* that had fed on lizard blood can survive and moult, on 12 October 2019 and 04 December 2020 we collected larvae and 1st nymphs of *O. gurneyi* from inside a calico bag that housed an infested sleepy lizard overnight, having been caught the previous day. For the ticks sampled on 12 October 2019 a 220 ml plastic jar was filled with ca. 5 cm deep layer of fine sand, and the engorged ticks were placed in the jar on top of the sand. Holes for ventilation were made in the lid of the jar and the mouth of the jar was covered with fine polyester gauze netting before securing the lid. The jar was then placed in a 210 mm glass desiccator, with ca. 700 ml of a saturated potassium chloride (KCl)

solution in its base, in a room where it would not be exposed to direct sunlight. Doube (1972) studied larval and nymphal instar development in cages that were placed in the natural environment, and his samples that were placed in the cages in October were checked 85 days later. We therefore decided to check whether the ticks had moulted 100 days after they had been placed into the jar since the aim of our experiment was to determine whether *O. gurneyi* that had fed on sleepy lizard blood can moult and not a study into the development time of the different immature instars, and because the ticks were being held indoors, which is cooler than under natural conditions. The ticks sampled on 04 December 2020 were treated differently to determine the development time of larvae and nymphs at a constant temperature. To do so, the ticks were randomly assigned to one of three treatments; (1) inside an empty sterile 50 ml plastic jar, or (2) inside one of two sterile 50 ml plastic jars filled with a 5 cm deep layer of fine sand. If the ticks in the empty jar survived and moulted, the status of the ticks in the jars that were filled with 5 cm deep layer of sand were checked on the same day that moulting was observed in the empty jar. But if the ticks in the empty jar died, the status of the ticks in one of the jars that was filled with 5 cm deep layer of sand were checked at 30 days and those of the other at 45 days. The centre of the lid of each jar was cut out, and the mouth of the jar was covered with fine polyester gauze netting before securing the lid. We also placed these jars into a desiccator, but instead of keeping it at room temperature, it was placed into a Laboro benchtop incubator, set to maintain a constant temperature of 28°C. We determined the moulting success rate of the ticks in jars filled with sand by sieving through the sand with a fine tea sieve for the ticks and/or their moults.

In order to obtain later instar nymphs of the kangaroo soft tick that had engorged on sleepy lizard blood, we created a fine sand layer, ca. 5-10 mm deep, in a plastic tub with a length, width and height of 28 cm, 19 cm and 10 cm respectively. At about midday on 09 December 2019 we placed a sleepy lizard that had been collected in the morning of the same day inside the tub on top of the sand. We then placed nymphs of *O. gurneyi* that had been collected from kangaroo wallows in the study site in June (i.e. 6 months prior) on top of the lizard. The tub was subsequently closed and left undisturbed in a shaded part of the room. At ca. 17:00 the lizard was removed and examined for attached ticks, after which the sand was sieved with a fine tea sieve to collect any ticks that may have buried into the sand. Nymphs that engorged were treated the same way as the conspecifics collected on 12 October 2019, and the lizards were then released at the point of capture.

Statistical analyses: We calculated the body condition index (BCI) of each lizard as the residuals of a linear regression between log-transformed BM and log-transformed SVL to compare un-infested and infested lizards. The SVL, BM and BCI data from un-infested lizards were not normally distributed, so a Mann-Whitney *U* test, with $\alpha = 0.05$, was used for all comparisons, computed using the Prism 8 package (Graphpad Software, San Diego, California, USA).

Results

The distribution of *Ornithodoros gurneyi* in and around the study site: We examined 105 kangaroo wallows from throughout the study site and surrounding areas (Fig. 4.3). The majority were under various trees and shrubs (Table 4.1), and the raised portions of the Morgan-Whyalla pipeline was the only man-made structure in the study site under which we found kangaroo wallows. We found *O. gurneyi* in 59 (56.2%) of the examined kangaroo wallows and most of these were under large shrubs or small trees (Table 4.1). *Ornithodoros gurneyi* was found throughout the study site. Voucher specimens were deposited in the parasites collection of South Australian Museum, Adelaide, South Australia (voucher numbers J 21400 to 214002, and J 24546).

Prevalence of *Ornithodoros gurneyi* parasitism on *Tiliqua rugosa*:

Five hundred and fifty-two uniquely marked lizards were caught during the study period, primarily in the months September to December of each year (Fig 4.4). However, some specimens were caught on an *ad hoc* basis in other months as well (Fig 4.4). Sixty-one of the lizards were recaptured multiple times (range = 1 – 4; mean \pm SD = 1.3 \pm 0.6), and the period between recaptures ranged from 5 to 741 days (mean \pm SD = 230.5 \pm 194.1). Each capture was regarded as an independent event because the lizards were not recaptured on the same day and any *O. gurneyi* that were found were removed. In total, there were 633 lizard captures.

For the entire study period, 2.5% of the captured lizards were infested by *O. gurneyi*, with an annual rate in 2017 to 2020 of 2.3 to 4.11% (mean \pm SD = 2.9 \pm 0.4). Infested lizards were mainly found in the months October to December (Fig 4.4.) and were caught throughout the study site (Fig. 4.5). The infested lizards ranged in SVL and BM from 269 mm to 337 mm (mean \pm SD = 296.5 mm \pm 17.4) and 325 g to 900 g (mean \pm SD = 543.3 g \pm 143.2) respectively.

Table 4.1. A summary of the numbers of kangaroo wallows that were examined for this study, the prevalence of kangaroo soft ticks (*Ornithodoros gurneyi*) in these wallows, and the type of cover they were found under.

Type of cover	Growth form	Number of wallows examined	Prevalence (%)
Plants			
<i>Acacia nyssophylla</i>	Shrub	8	100
<i>Acacia oswaldii</i>	Tree	7	85.7
<i>Alectryon oleifolius</i>	Tree	7	71.4
<i>Arundo donax</i>	Perennial cane	1	100
<i>Casuarina pauper</i>	Tree	9	55.6
<i>Eremophila oppositifolia</i>	Shrub	2	100
<i>Eremophila oppositifolia</i> and <i>Maireana sedifolia</i>	Shrub	1	100
<i>Eucalyptus oleosa</i>	Tree	11	27.3
<i>Exocarpos aphyllus</i>	Shrub	20	50
<i>Exocarpos aphyllus</i> and <i>Geijera linearifolia</i>	Shrub	1	0
<i>Geijera linearifolia</i>	Shrub	14	71.4
<i>Geijera linearifolia</i> and <i>Myoporum platycarpum</i>	Shrub	1	100
<i>Maireana pyramidata</i>	Shrub	4	25
<i>Maireana sedifolia</i>	Shrub	3	66.7
<i>Myoporum platycarpum</i>	Shrub	9	11.1
<i>Pittosporum angustifolium</i>	Tree	1	100
<i>Senna artemisioides</i>	Shrub	2	0
Man-made structures			
Pipeline		4	50

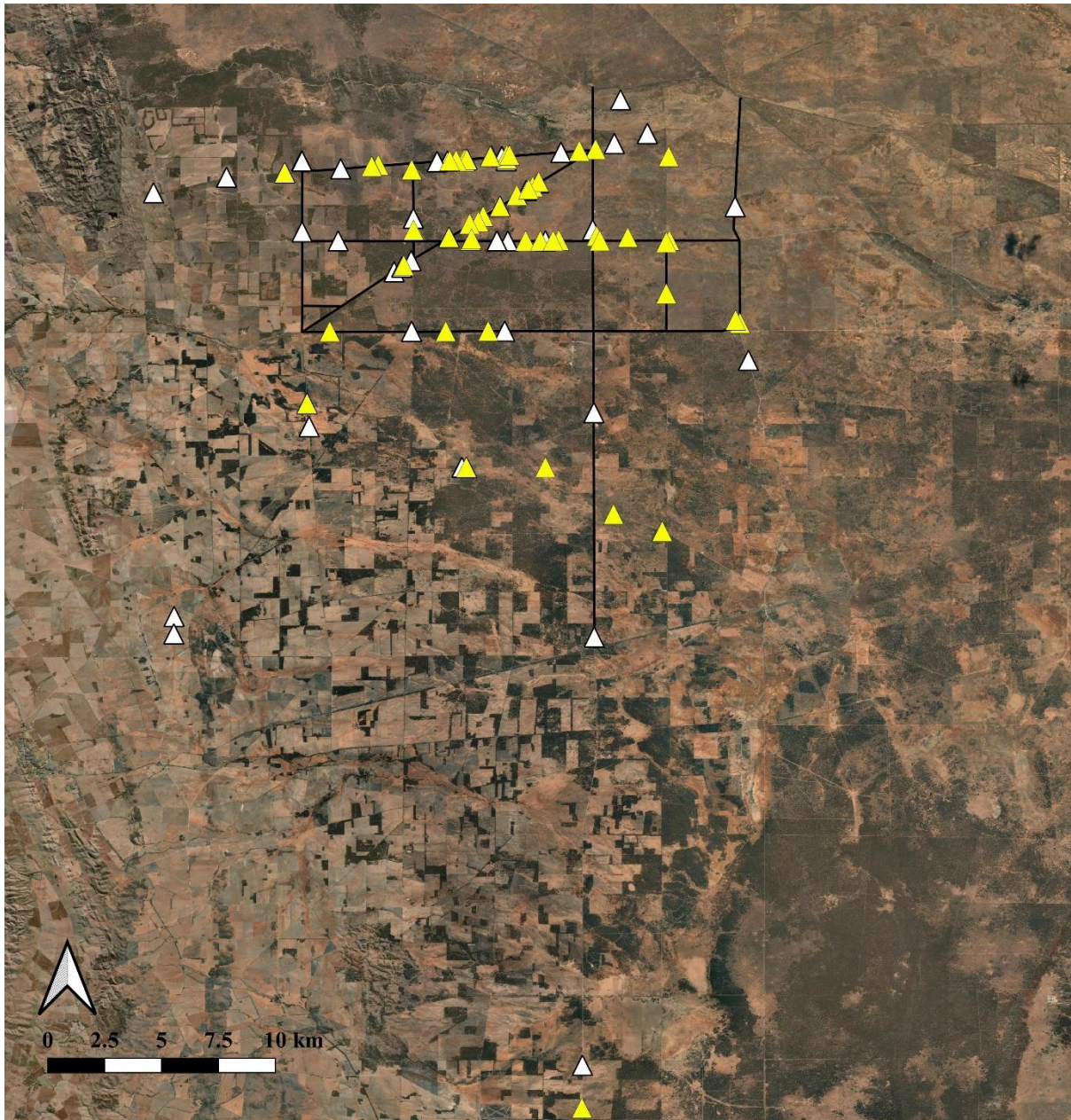


Figure 4.3. The distribution of kangaroo wallows in the study site where kangaroo soft ticks (*Ornithodoros gurneyi*) were found (yellow triangles), as well as where they were not found (white triangles). The black lines are the transects used for the ongoing sleepy lizard (*Tiliqua rugosa*) surveys.

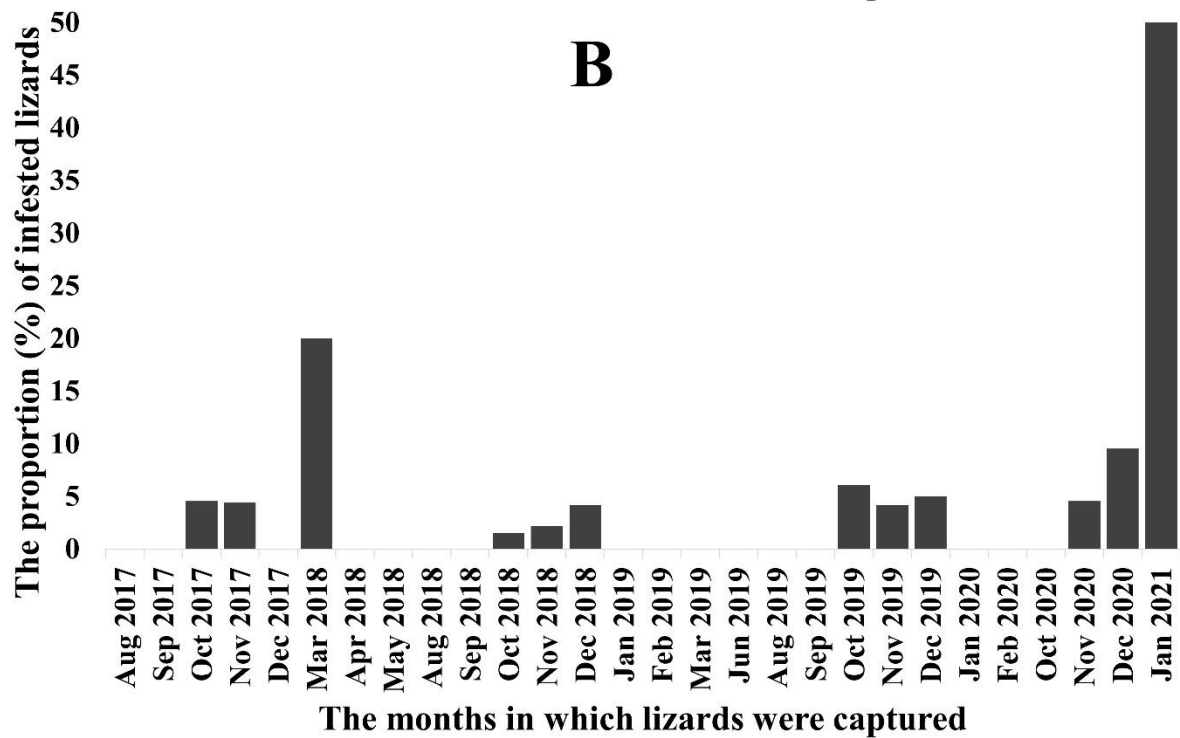
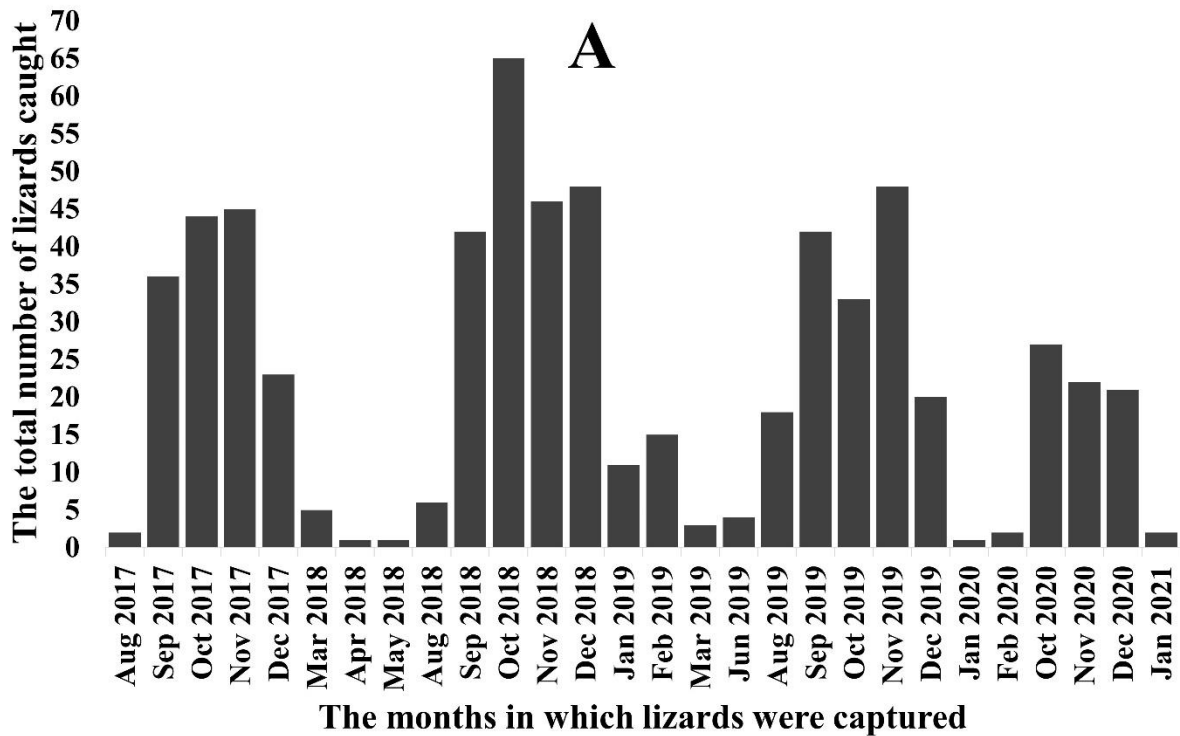


Figure 4.4. (A) The monthly total number of sleepy lizards (*Tiliqua rugosa*) that were caught for this study, and (B) the monthly proportion of those lizards that were infested by kangaroo soft ticks (*Ornithodoros gurneyi*).

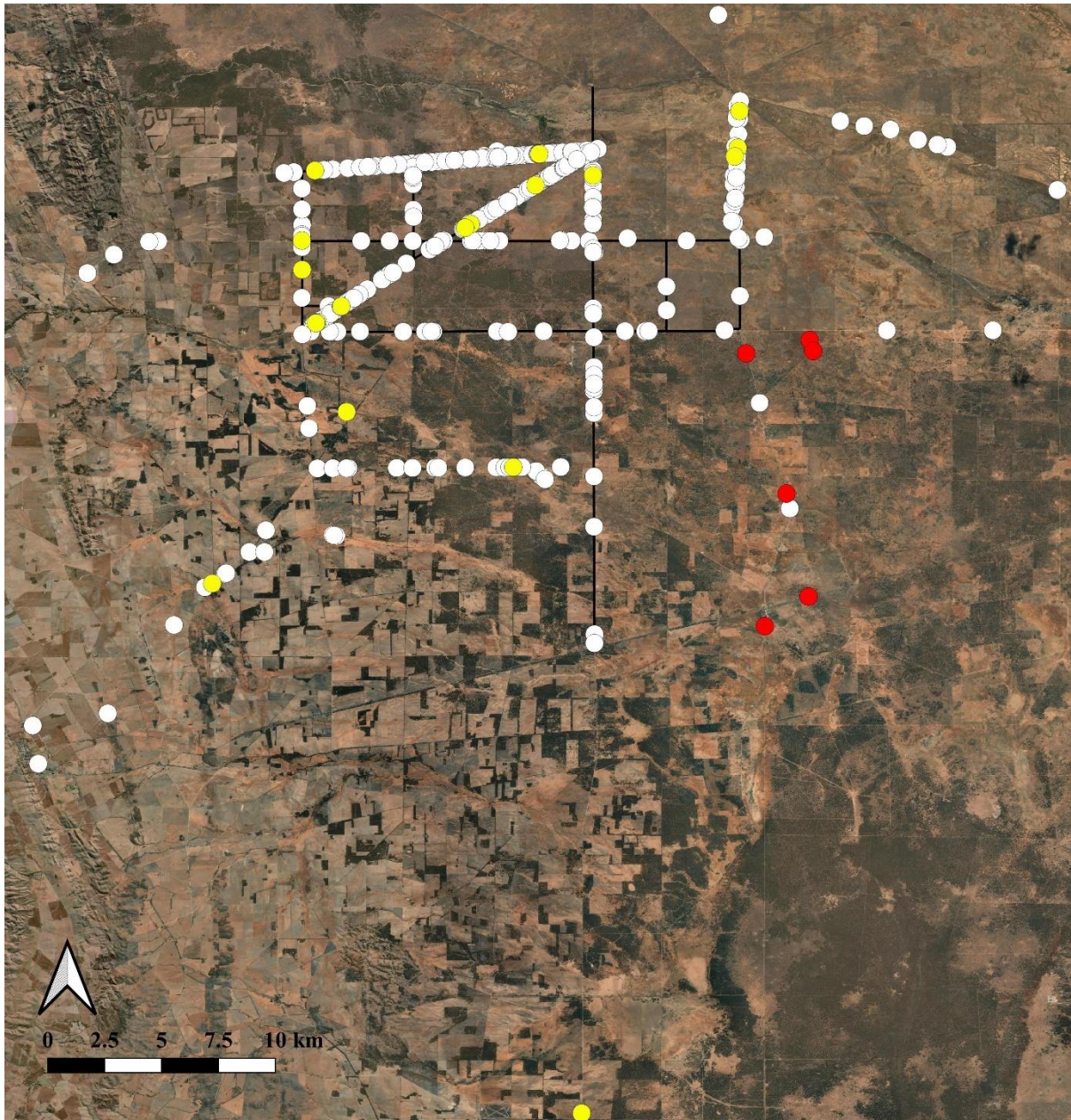


Figure 4.5. The distribution of where the sleepy lizards (*Tiliqua rugosa*) used in this study were caught. The localities where lizards that were infested by kangaroo soft ticks (*Ornithodoros gurneyi*) were caught are indicated with yellow dots, those of un-infested lizards are indicated with white dots, and localities where infested lizards were caught in the 1970's are indicated with red dots, and the black lines are the transects used for the ongoing sleepy lizard surveys.

The SVL of the infested lizards did not differ significantly from those of un-infested conspecifics (range = 177 mm – 347 mm; mean \pm SD = 299.4 mm \pm 25.9) that were caught in the same months ($U = 2626$, $P = 0.0933$), nor did the BCI of infested lizards (range = -0.15174 – 0.06172; mean \pm SD = -0.03126 \pm 0.07411) differ significantly ($U = 2612$, $P = 0.0933$) from those of un-infested conspecifics (range = -0.19899 – 0.17525; mean \pm SD = 0.00158 \pm 0.05656) collected in the same months.

We only found larvae and nymphs of *O. gurneyi* parasitizing the sleepy lizards. No attempts were made to measure engorged nymphs as they can not always reliably be assigned to specific nymphal instars, but we deduced, due to their relatively small size, they were likely first and second nymphal instars. The ticks were usually attached under the large neck and dorsal scales of the lizards, in the folds of the eyelids and inside the ear and nasal openings (Fig. 4.6). A voucher specimen was deposited in the Australian National Insect Collection (ANIC), CSIRO, Canberra, Australian Capital Territory (voucher number ANIC 48 006 559).

Results of the moulting experiment:

The numbers of engorged larvae and nymphs that were used in the moulting experiments and their moulting success rates are summarised in Tables 4.2 and 4.3. All the ticks used in this experiment moulted or died. The majority (82.8%) survived and successfully moulted, and mortality was only noted in the larvae and nymphs (likely 1st instar nymphs due to their small size) that detached from lizards caught in the wild (Table 4.2). None of the nymphs that were placed with a sleepy lizard host in captivity were still attached by the time the lizard was examined ca. four hours after the ticks were placed on it, and although only 28.6% of the nymphs that were placed with a sleepy lizard host in captivity attached and engorged, their survival and moulting rates were 100% (Table 4.3).

Discussion

Our study found that *O. gurneyi* occurs throughout our study site in the Mid North region of South Australia, which is situated further south than what was formerly hypothesised by Browning (1962) and Seddon (1968) to be the southern limit of the extension range of this tick in Australia. This extension of *O. gurneyi*'s distribution is unlikely the result of a recent introduction of this tick species into the area since in the early 1970's Sharrad (1979) also found some specimens parasitizing *T. rugosa* that were caught in the vicinity of our study site. Browning (1962) found that *O. gurneyi* was confined to open arid habitats and absent in more wooded areas in spite of the presence of suitable hosts. It was speculated that this was due to the resting and thermoregulation behaviour of red kangaroos, which make frequent use of the same wallows in open arid habitats due to the limited number of suitable shady shrubs and trees, and thereby ensure the likelihood of the ticks finding a suitable host, but do not utilize the same wallows in wooded habitats (Browning 1962, Doube 1972). Even though red kangaroos are present in our study site, they tend to be rare. Western grey kangaroos (*Macropus*

fuliginosus) on the other hand are quite common in this locality. If the resting and thermoregulation behaviour of western grey kangaroos differ from that of red kangaroos and they utilize the same wallows in spite of the habitat type, it would ensure a frequent host for the ticks and enable their survival in more wooded environments. The unreported *O. gurneyi* specimens from Kangaroo Island (Australian National Insect Collection; ANIC 48 000 205 and ANIC 48 000 292) and our records from the Mid North region of South Australia indicate that the distribution of this tick is more extensive than what is currently believed. The distribution of *O. gurneyi* and the role different kangaroo species play in its distribution requires additional study.

Table 4.2. The collection date and numbers of larvae and nymphal kangaroo soft ticks (*Ornithodoros gurneyi*) that were collected from wild-caught sleepy lizards (*Tiliqua rugosa*), for the moulting trial. Their development time, moulting and mortality rates are also given (* - development was checked after a predetermined period, so the actual development period is not known; (a) in the empty jar; (b) in the sand-filled jar that was to be checked at 30 days; and (c) in the sand-filled jar that was to be checked at 45 days).

Collection date	Life stage	Number of ticks	Development time	Moulting success (%)	Mortality rate (%)
12 October 2019	larva	6	<100 days*	66.7	33.3
4 December 2020	a larva	4	10 days	75	25
	a nymph	1	10 days	–	100
	b larva	4	10 days	100	0
	b nymph	1	12 days	–	100
	c larva	6	10-12 days	83.3	16.7

Table 4.3. The date on which the nymphal kangaroo soft ticks (*Ornithodoros gurneyi*) were placed on a captive sleepy lizard (*Tiliqua rugosa*) in captivity, the number of nymphs placed in the container with the lizard (No. Used), and the number of ticks that attached and engorged (No. Engorged) as part of the moulting experiments. The time at which the development was checked (Duration) after the engorged tick had fed and detached from the lizard, as well as the moulting success rate are also given (* development was checked after a predetermined period, so the actual development period is not known).

Date	No. Used	No. Engorged	Duration	Moulting (%)
9 December 2019	10	1	<100 days*	100
15 December 2019	14	5	<100 days*	100
3 October 2020	4	1	<45 days*	100

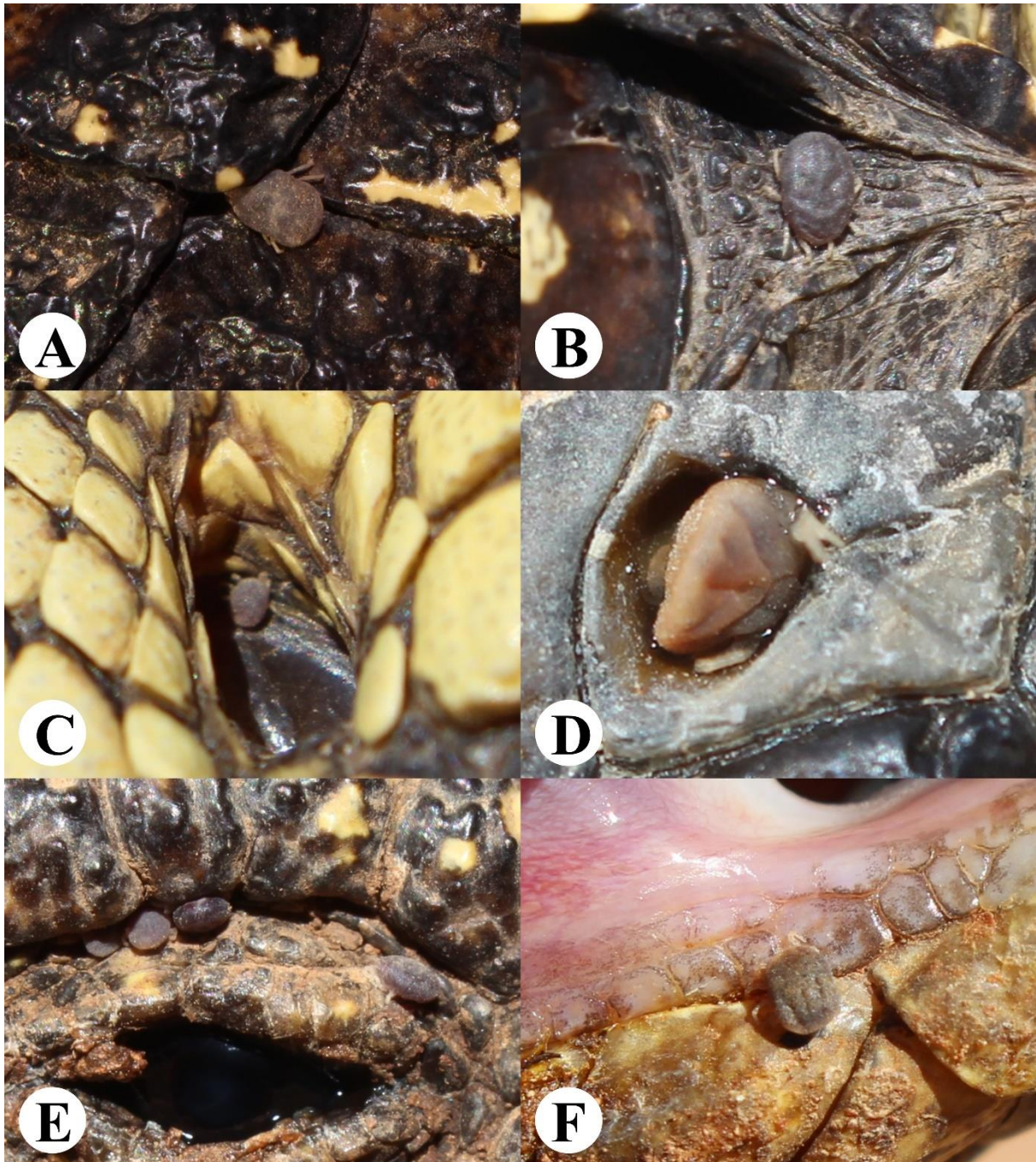


Figure 4.6. The attachment sites of kangaroos soft ticks (*Ornithodoros gurneyi*) were (A) underneath (A) and between (B) large dorsal scales, inside the ear (C) and nasal (D) openings, and in the folds of the eyelids (E) of sleepy lizards (*Tiliqua rugosa*). One nymph was attached between the scales of the lower labials near the corner of the mouth of one of the lizards (F).

In this study we also showed that *O. gurneyi* is a parasite of *T. rugosa* in this locality and that sleepy lizards are competent hosts. Complex interactions take place between feeding ticks and the host and as a result the host can play an important role in the evolution of the tick and its host specificity (Mans et al. 2003). Some *Ornithodoros* species, only parasitize a narrow range of hosts. For instance, *Ornithodoros compactus* has only been recorded from tortoises in southern Africa, and *Ornithodoros darwini* and *Ornithodoros galapagensis* are parasites of

lizards from the Galápagos Islands (Keirans et al. 1980, Schatz 1991, Horak et al. 2006). Others are more opportunistic and may parasitise a variety of hosts. For example, *Ornithodoros puertoricensis* has been recorded to parasitise mammals, amphibians, a bird, mammals and reptiles (Bermúdez et al. 2015). Still, it should be noted that many ticks tend to be host specific to some extent and feeding on atypical hosts could negatively impact survival (Hoogstraal and Aeschlimann 1982). For instance, Andrews et al. (2006) noted that the larvae of the echidna-specializing tick, *Bothriocroton tachyglossi*, do not survive when they were experimentally exposed to *T. rugosa*, because the ticks either failed to attach or when they attached they did not engorge. In our study most of the ticks that fed on sleepy lizards survived and moulted. Also, since mortality was only noted in a small number of larvae and first nymphal instars, it is possible that they died as a result of the handling, rather than due to the source of their blood meal. Tick survival should be explored in future studies, which should also include the use of adult ticks.

If sleepy lizards are competent hosts of *O. gurneyi*, why are they not found more frequently on these lizards? The low observed prevalence of *O. gurneyi* parasitism on *T. rugosa*, is most likely due to the behaviour of the lizards and the ticks. Sleepy lizards are primarily active during the warmer mid part of the day during the late winter to spring period, but during the hot late spring to summer period their activity is limited to the cooler morning and afternoon periods (Firth and Belan 1998, Kerr and Bull 2006). During the hottest part of hot days the lizards seek shelter in the shade of bushes (Firth and Belan 1998, Kerr et al. 2003) and occasionally trees (Fig. 4.7). Sleepy lizards that seek shelter under shrubs and trees with a kangaroo wallow under them may then be parasitised by *O. gurneyi*, explaining why parasitism was only noted during the months October to March, and not during the cooler months of August and September.

Considering that the lizards remain inactive and the ticks feed rapidly, the noted low prevalence of *O. gurneyi* parasitism on *T. rugosa* may be an underestimate. Under laboratory conditions, studies have found that larvae and early-instar nymphs of *O. gurneyi* attached to laboratory rats and rabbits (the experimental hosts) for about three days (up to 12 days) to feed, as opposed to late-instar nymphs and adults that attached and fed for less than three hours (Browning 1962, Doube 1972, 1975a). In our study all the nymphs that attached and engorged on sleepy lizard blood under captive conditions did so in less than four hours (the exact time is not known). It is therefore likely that instances *O. gurneyi* parasitism on *T. rugosa* go unrecorded because early-instar nymphs and even possibly adults of these ticks feed and detach while the lizard

remains inactive, resting under bushes. As a result only the instances involving larvae and early-instar nymphs of *O. gurneyi* that attached for longer periods of time are recorded because during the course of the long-term surveys at the study site lizards are primarily collected when they are active. Additional studies are therefore needed to explore the likelihood of more frequent *O. gurneyi* parasitism on *T. rugosa*.



Figure 4.7. At 11:30 on 16 December 2019 sleepy lizard (*Tiliqua rugosa*) number 4880 was found in the shade under a bullock-bush (*Alectryon oleifolius*). The ground temperature in the shade under the tree was 29.1°C, while the ground temperature out in the open was 52.0°C. Droppings indicated that kangaroos and sheep also sheltered under the tree at some point and several nymphal and adult kangaroo soft ticks (*Ornithodoros gurneyi*) were collected from the soil.

The role that sleepy lizards and other reptiles play in the ecology of *O. gurneyi* is unknown. Apart from being a potential host the lizards may also play a role in the dispersal of these ticks. The dispersal of a species can be a short distance to adjacent environments (diffusion dispersal) or long distance to distant environments (saltation dispersal) (Davis and Thompson 2000). Red kangaroos can travel several kilometres per day in search of water (Croft and Clancy 2008), and are therefore most likely responsible for diffusion and saltation dispersal of the larvae of *O. gurneyi* that attach for several days. Sleepy lizards travel about 200 m per day (Taggart et al. 2018), and as stated earlier, during the hottest part of hot days the lizards will seek shelter in the shade of shrubs and trees, and during extended periods of unfavourable conditions they usually seek shelter in the burrows of other animals (Kerr et al. 2003). Considering that *O. gurneyi* has been collected together with the reptile tick *Amblyomma moreliae* from burrows

near Rockhampton in Queensland, Australia (Wilkinson 1958), it is likely that larvae and early-instar nymphs of *O. gurneyi* that attached for several days can be dispersed to these localities by the lizards, which therefore play a role in the diffusion dispersal of *O. gurneyi*. Empirical studies are needed to examine the role that sleepy lizards play in the ecology of *O. gurneyi* and vice versa.

Conclusions

Our study indicates that the distribution of *O. gurneyi* in Australia is more extensive than what was formerly assumed and that *T. rugosa*, and likely other large reptiles, are competent hosts of these ticks. Surveys are needed to determine the distribution of this tick, and additional empirical studies are required to gain a better understanding of the ecology of this poorly studied tick and its hosts. Given the poorly known argasid fauna of Australia and the extent of the distribution of *O. gurneyi*, further studies, employing more refined morphological and molecular analyses, should be done to confirm the species status of this tick throughout its range.

Disclosures

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Appendix 4.1. A summary of the museum specimens and literature sources that were used for generating the distribution map of the kangaroo soft tick (*Ornithodoros gurneyi*) in Australia.

Host / Habitat	State	Locality	Reference
Not stated	NT	Borrooloola	ANIC 48 000 196, 48 000 197, 48 000 199, and 48 000 214
Not stated	NT	Coniston Station	J7145 and J7146
Not stated	NT	Gibeanie Station	ANIC 48 000 203
Not stated	NT	Kapalga	ANIC 48 000 190 - ANIC 48 000 194
Kangaroo wallow	NT	MacDonnell Ranges	Doube (1975b)
Not stated	NT	McArthur River Station	ANIC 48 000 198
Kangaroo wallow	NT	Reynolds Ranges	Doube (1975b)
	NT	Wongalara Wildlife Sanctuary	WAM 124535
Not stated	QLD	Barcaldine	B3406
Not stated	QLD	Biloela	ANIC 48 000 212
Not stated	QLD	Boulia, Digby Peaks	ANIC 48 000 216
<i>Homo sapiens</i>	QLD	Charters Towers	QM W676
<i>Macrotis lagotis</i>	QLD	Davenport Downs	ANIC 48 000 205
Burrow	QLD	Gracemere	Wilkinson (1958)
Not stated	QLD	Hughenden	Seddon (1968)
Not stated	QLD	Julia Creek	Seddon (1968)
<i>Homo sapiens</i>	QLD	Katandra	ANIC 48 006 150
Cave	QLD	Lawn Hill	Roberts (1936)
<i>Homo sapiens</i>	QLD	Lawnhill Gorge	ANIC 48 000 204
Not stated	QLD	Lords Table	QM S73710
Burrow	QLD	Malchi	Wilkinson (1958)
Not stated	QLD	Mt. Isa	ANIC 48 000 217; QM S29943; Seddon (1968)
Not stated	QLD	Pinkella Opel Mine	QM S35009
Not stated	QLD	Rockhampton	ANIC 48 000 188, 48 000 215 and 48 000 218

Appendix 4.1 continued

Not stated	QLD	Rockhampton	ANIC 48 000 188, 48 000 215 and 48 000 218
Not stated	QLD	Stamford	ANIC 48 000 183, 48 000 200, 48 000 206 - 48 000 211, 48 000 219 and 48 000 220; Seddon (1968)
Not stated	QLD	Taroom	QM S45760
	QLD	Wights Mountain	QM W242
Not stated	QLD	Winton	ANIC 48 000 222
Kangaroo wallow	NSW	Fowlers Gap	Doube (1975b)
Kangaroo wallow	NSW	Pine Creek	Doube (1975b)
Kangaroo wallow	NSW	Tibooburra	Henry (1938)
Kangaroo wallow	SA	Flinders Ranges	Doube (1975b)
Not stated	SA	Hawker	ANIC 48 000 189; QM S29862
Not stated	SA	Kalabity	J7111 - J7126
<i>Tachyglossus aculeatus</i>	SA	Kangaroo Island	ANIC 48 000 292
Kangaroo wallow	SA	Kingoonya	Doube (1975b)
Kangaroo wallow	SA	Lake Gairdner	Petney et al. (2004)
Not stated	SA	Lake Harry	J7128
Kangaroo wallow	SA	Moralana Station	B350; Doube (1975a); J7130
Not stated	SA	Mount Aleck	J7127
	SA	Mt. Mary	Sharrad (1979)
Kangaroo wallow	SA	north of the 10 in. isohyet	Browning (1962)
<i>Homo sapiens</i>	SA	Oakden Hills Station	J7131 - J7144
Not stated	SA	South Gap Station	J7129
	WA	Barrow Island	WAM 57831 and 57832
	WA	Bilarni Cave	WAM 58120
	WA	Cape Range National Park	WAM 58122
	WA	Chichester Range	WAM 110549
Not stated	WA	Derby	ANIC 48 000 195

Appendix 4.1 continued

Not stated	WA	Derby		ANIC 48 000 195
Not stated	WA	Ejah Breakaway		ANIC 48 000 184
	WA	Kathleen Valley		WAM 58125
	WA	Kennedy Range National Park		WAM 58126
	WA	Madura		WAM 58127
Kangaroo wallow	WA	Mileura Station		Doube (1975b)
	WA	North West Cape		WAM 58136 – 58139, 58141 - 58143
Reptiles	WA	south-eastern Australia	Western	Sharrad and King (1981)

Chapter 5

The roadside plant communities along two transects spanning an ecological gradient, surveyed during a period of drought at a locality near Mt. Mary, South Australia

Note to examiners

Vegetation descriptions are often incorporated with zoological studies as the plants are a food source for some animals, and at the same time make up the physical structure of the habitat and create the microclimate the animals inhabit. In Chapter 5, I report on the results of vegetation surveys I did to determine the plant communities along two transects at the Bunday Bore study site. This chapter allows a comparison of parasites and vegetation categories for Chapter 6 in which I examine if correlations exist between the presence and absence of the various parasites of the sleepy lizards. This chapter is a manuscript that I am intending to submit to the journal *Transactions and Proceedings of the Royal Society of South Australia* and it is therefore formatted according to the specifications of the journal. The referencing style was however changed to make it consistent with that of the other parts of my thesis. The numbering of the figures and tables were changed for clarification and so that they appear chronologically in the chapter. I took the photographs and therefore own the copyright. The raw data of the vegetation surveys are provided at the end of the thesis (Appendices 7.2 to 7.5). The font size in the appendices is reduced to keep the pagination to a minimum.

Abstract

Plant species composition and structure of a specific plant community are indicative of the abiotic and/or biotic factors that prevail. Vegetation descriptions are therefore important aspects of ecological studies since the different plant species assemblages are a source of energy for animals while their structural arrangement create microclimates suitable for a variety of animal species in addition to being indicators of prevailing environmental conditions. Vegetation surveys were conducted along two transects as part of a long-term study of host/parasite/environment interactions in the Mid North of South Australia. The vegetation surveys used the modified Braun-Blanquet cover abundance scale and the floristic data was analysed with the Weighted Two-Way Indicator Species Analysis algorithm. Two major plant communities were identified consisting of nine sub-communities with a mosaic distribution pattern and the roadside vegetation was found to usually be an extension of that of the neighbouring habitat. The results also indicated that the largest part of the study area has been exposed to different degrees of disturbance. Since this study was conducted during a prolonged drought period it is imperative that long-term monitoring of these survey plots be continued, especially after good rainfall, to be able to add new species to the list and more accurately reflect the site diversity across varying environmental conditions.

Introduction

As in other organisms, plant species have evolved to thrive within the limits of specific abiotic factors (i.e. carbon dioxide, light, oxygen, soil composition and minerals, temperature and water), and the world's biomes are to a large extent a result of these factors (Tivy 1993, Mucina 2019). In addition to the abiotic drivers, plant communities are also influenced by biotic factors (i.e. animals, anthropogenic activities and competition with other plants), which are usually more apparent regionally. For instance, the eradication of rinderpest in the Serengeti ecosystem of East Africa resulted in an increase in the populations of grazing herbivores, which led to increased grazing pressure and reduced fire extent, and as a result the grasslands experienced bush encroachment by woody species (Holdo et al. 2009). Plant species composition and structure of a specific plant community are therefore indicative of the abiotic and/or biotic factors that prevail (van As et al. 2012).

There is usually an association between specific animal species and particular plant communities. Habitat preferences have been noted for various animal species such as ungulates (Hirst 1975), primates (Henzi et al. 2011, Pasternak et al. 2013), as well as birds (Wiens and Rotenberry 1981, Wood et al. 2010), reptiles (Jellinek et al. 2004, Souter et al. 2007) and invertebrates (Bradley 1986, Aguirre-Gutiérrez et al. 2017). Vegetation descriptions are therefore important aspects of ecological studies since the different plant species assemblages are a source of energy for animals while their structural arrangement create microclimates suitable for a variety of animal species (Tews et al. 2004).

Sleepy lizards, *Tiliqua rugosa* (Gray 1825), have a natural distribution that extends over most parts of southern Australia (Cogger 2014). They are abundant in the area north of Mt. Mary in the Mid North region of South Australia. During the 1970's this area was used for studies pertaining to the biology and parapatric boundary of the two reptile ticks, *Amblyomma limbatum* Neumann, 1899 and *Bothriocroton hydrosauri* (Denny 1843) that often infest sleepy lizards (Smyth 1973, Sharrad 1979). Since 1982 the area in and surrounding Bunday Bore, ca. 25 km northwest of Mt. Mary, has been used for an ongoing long-term study, initiated by the late Prof. Michael Bull, on sleepy lizards and their ticks (Godfrey and Gardner 2017). Apart from a general description of the study site (e.g. Sharrad 1979, Bull et al. 1981), and studies pertaining to what the lizards eat (Dubas and Bull 1991), their thermoregulatory and sheltering behaviour (Kerr et al. 2003, Kerr and Bull 2004), no descriptions of the vegetation or its

structure in this locality have been made. For most of the research conducted in this area, the lizards were primarily collected as they cross the roads or where they were feeding, basking or sheltering among the vegetation along the roadsides (Bull et al. 1989). The purpose of this study was therefore to describe the roadside plant communities along two transects of the Bunday Bore study site.

Material and methods

The study was conducted at the Bunday Bore study site, ca. 160 km north of Adelaide, in the Mid North region South Australia (Fig. 5.1). This region has a semi-arid climate, with an average annual rainfall of less than 250 mm, most of which usually falls during the austral winter (Smyth 1973). The habitats in this region consist of a mosaic of areas that have been cleared for agriculture and grazing, with interspersed fragments of the grassy arid mallee scrub and the chenopod shrubland that used to dominate the south-western and north-eastern side of the study site respectively (Jessup 1948, Smyth 1973). This region has calcareous solonized brown soils, with deposits of kunkar on ridges where the soil is shallow, and alluvial clay and silt deposits in depressions where water drains from the area (Jessup 1948, Sharrad 1979).

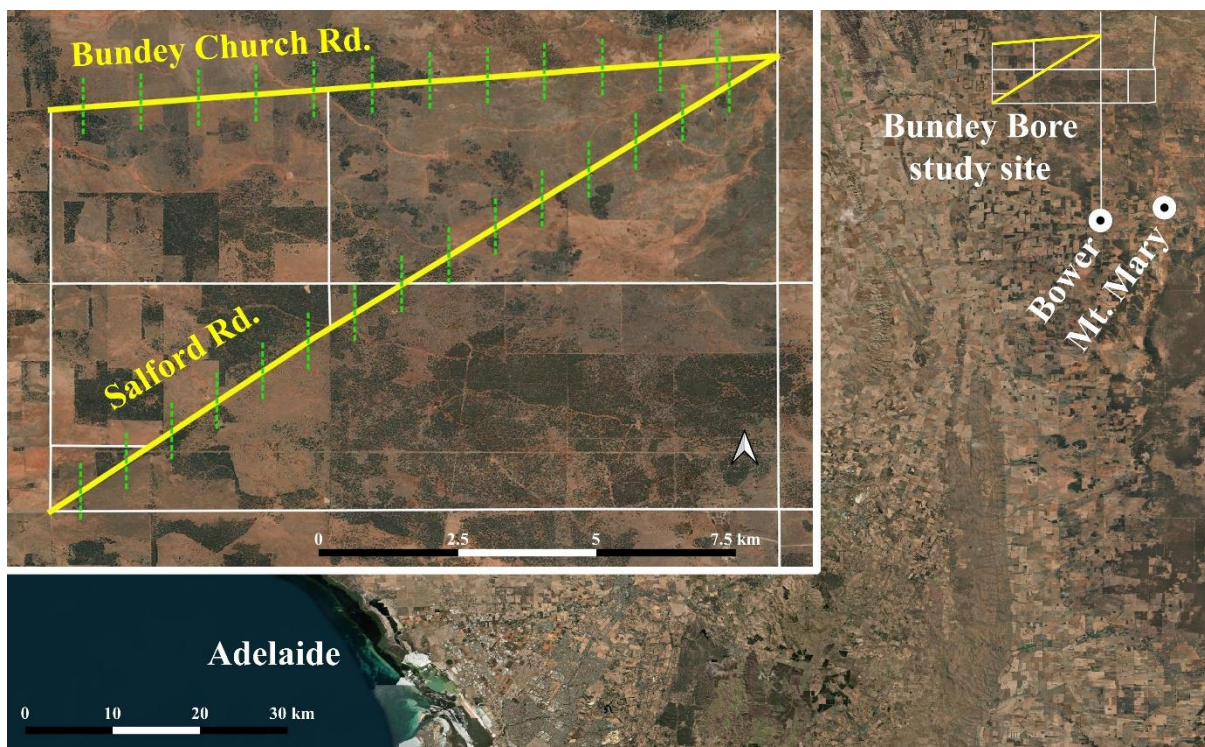


Figure 5.1. The position of the Bunday Bore study site is situated ca. 160 km north of Adelaide, in the Mid North region South Australia, and a layout of the intervals (dotted green lines) at which survey plots were positioned along the two transects (yellow lines) used in this study (insert).

The dirt roads of the Bunday Bore study site are used as the transects of the ongoing long-term study. Bunday Church Road (ca. 12 km long) and Salford Road (ca. 16 km long) were selected as the two transects of this study (Fig. 5.1), because they span across the ecological gradient between the more mesic mallee scrubland dominated areas and the more xeric chenopod shrubland areas, which has been seen to be important in limiting the distributions of some of the parasite species (Bull and Burzacott 2001). The vegetation along the roads are also representative of the vegetation of the entire study site. The minimal area method (Barbour et al. 1987, Kent 2012) was used to determine the minimum plot size for each area sampled. The roadside vegetation experiences less disturbances due to a near absence of grazing by sheep and can therefore be indicative of more natural vegetation. However, the lizards also utilise the more disturbed pastures, thus, vegetation sample plots were also placed within these areas. The roadside plots were placed at ca. 1 km intervals, at both sides of the road along the transects. Roadsides can be influenced by the road edge effect (i.e. the enhanced nutrient and/or moisture associated with the road promote plant growth), however, since the lizards make use of this area for foraging we specifically included these areas in the placing of the sample plots. When a roadside plot bordered a property that we had access to, an additional plot was placed, ca. 25 m from the road edge, within the neighbouring habitat. The plots were surveyed seasonally (i.e. four times during the year).

Within each sample plot, all the plant species present, except bryophytes and lichen, were identified and recorded according to the nomenclature as listed in Barker et al. (2005). The percentage of the plot covered by each species was visually estimated and scored using the modified Braun-Blanquet cover abundance scale (Mueller-Dombois and Ellenberg 1974):

r – one or a few individuals with very low cover

+ – less than 5 % of total area of stand and irregular appearance

1 – abundant, but with low cover; or less abundant, but with a greater cover, but less than 5 % of the total surface of the stand

2 – abundant, but less than 5 % cover, or 5 - 25 % cover of total surface of the stand

3 – 25 - 50 % cover of total stand surface, irrespective of the number of individuals

4 – 50 - 75 % cover of total stand surface, irrespective of the number of individuals

5 – 75 - 100 % cover of total stand surface, irrespective of the number of individuals

It is recommended that plant community descriptions be based on data gathered during the period of optimal growth (Brown et al. 2013). For vegetation classification purposes we

therefore only analysed the data collected during the period of optimal growth in late spring and early summer. Extra species observed during other periods were added to the list to be as comprehensive as possible. The floristic data was entered into an Microsoft Excel spreadsheet and imported into the computer programme JUICE (Tichý, 2002). The modified Weighted Two-Way Indicator Species Analysis (TWINSPAN) algorithm (Roleček et al. 2009), was used to derive a first approximation of the floristic data. Pseudospecies cut levels were set at 0-5-15-25-50-75 (Brown et al. 2013), with a minimum group size of 2 and minimum dissimilarity for division at 0.3. Fidelity of each community was measured using the phi coefficient of association (Chytrý et al. 2002) Further refinement of the classification was done by applying Braun-Blanquet procedures (Kent and Coker 1992, Brown et al. 2013). Within the phytosociological table the generated clusters were not altered, but the species were manually rearranged into their species groups (Brown et al. 2013).

We used the phytosociological table to describe the plant species composition of the different identified plant communities. Since this is a semi-arid area and the vegetation sparse, the lower threshold values for fidelity, frequency and cover values were set in JUICE at 65, 70, and 12 respectively, and that of the upper thresholds at 90, 90 and 50 (Brown and Bezuidenhout 2018). The diagnostic, constant and dominant species were statistically determined from the synoptic table, and were derived from the fidelity, frequency and cover values.

To measure species diversity and evenness, the cover-abundance data collected during the surveys were transformed to a numerical scale ($r = 1$; $+$ = 2; $1 = 3$; $2 = 5$; $3 = 7$; $4 = 8$; $5 = 9$) (van der Maarel 1979, 2007). The results of the Shannon-Wiener and Gini-Simpson indices were converted to effective number of species (Jost 2006) to allow for appropriate comparisons.

Results

Based on the results of the minimal area method, the largest roadside habitat plot size was 4×8 m (i.e. 32 m^2), while the largest in the pastures and mallee scrubland was 8×8 m (i.e. 64 m^2), so for practical reasons and ease of use in the field a plot size of 50 m^2 was used for all the roadside and 100 m^2 was used for the neighbouring habitats. A total of 40 sample plots ($50 \text{ m}^2 - n = 24$; $100 \text{ m}^2 - n = 16$) were placed along Bunday Church Road and 46 ($50 \text{ m}^2 - n = 29$; $100 \text{ m}^2 - n = 17$) sample plots were placed along Salford Road.

We recorded 76 vascular plant species belonging to 27 families during the study period (Table 5.1). The three largest families were the Chenopodiaceae with 16 species, reflecting 21.6% of the total flora, the Compositae with 12 species, representing 16.2% of the flora, and the Leguminosae with 6 species, representing 8.1% of the flora of the study site (Fig. 5.2). The low-growing shrub *Atriplex vesicaria* and the small shrub *Maireana sedifolia* are indicator species for low shrubland communities in arid parts of South Australia (Costermans, 2009; Specht, 1972). Optimal growth for these species were noted during the December survey so no analyses were done on the March, June and September survey data.

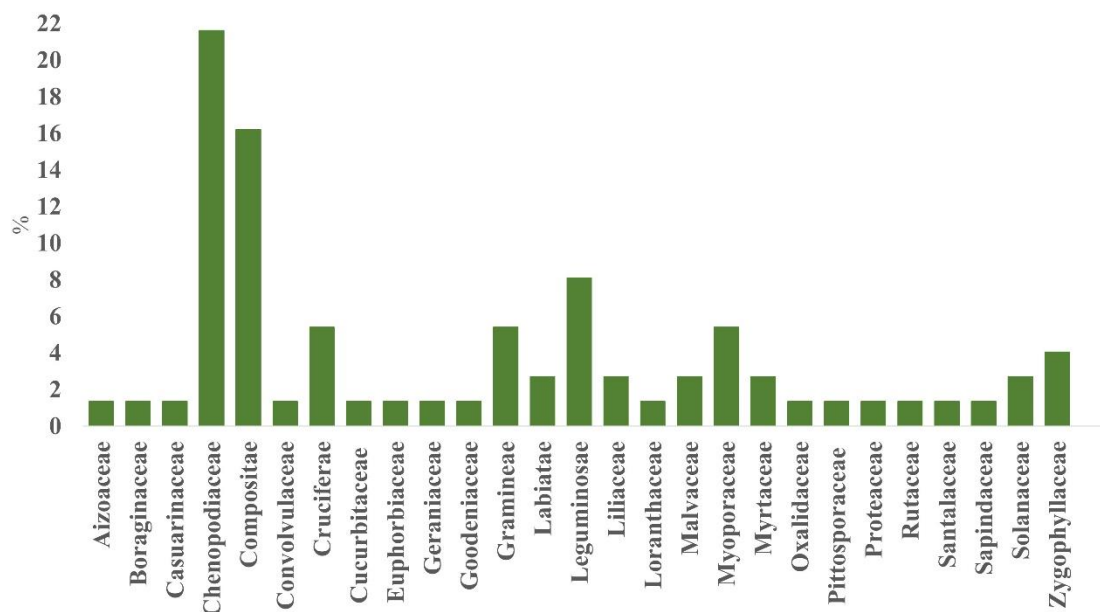


Figure 5.2. The number of species from each plant family recorded in this study as a proportion of the total number of recorded species.

Classification and description: A modified TWINSPLAN classification for the December survey data resulted in the identification of nine plant communities (Table 5.2) that could be grouped into a habitat mosaic of two major communities along the two transects:

1. *Rhagodia ulicina*–*Atriplex vesicaria* scrubland
 - 1.1 *Rhagodia ulicina*–*Atriplex vesicaria*–*Casuarina pauper* sub-community
 - 1.2 *Rhagodia ulicina*–*Atriplex vesicaria*– *Senna artemisioides filifolia* sub-community
 - 1.3 *Rhagodia ulicina*–*Atriplex vesicaria*–*Maireana pyramidata* sub-community
 - 1.4 *Rhagodia ulicina*–*Atriplex vesicaria*–*Eucalyptus oleosa* sub-community

2. *Medicago minima*–*Maireana sedifolia* shrubland
 - 2.1 *Medicago minima*–*Maireana sedifolia*–*Nicotiana velutina* sub-community
 - 2.2 *Medicago minima*–*Maireana sedifolia*–*Maireana pyramidata* sub-community
 - 2.3 *Medicago minima*–*Maireana sedifolia*–*Senna artemisioides* sub-community
 - 2.4 *Medicago minima*–*Maireana sedifolia*–*Salvia verbenaca* sub-community
 - 2.4.1 *Euphorbia drummondii* variant
 - 2.4.2 *Rytidosperma caespitosum* variant
 - 2.5 *Medicago minima*–*Maireana sedifolia*–*Carrichtera annua* sub-community

Description:

1. *Rhagodia ulicina*–*Atriplex vesicaria* scrubland

This community occurs on flat and undulating plain areas, as well as in the proximity of shallow gullies, primarily in the western side of the study site (Fig. 5.3). The vegetation comprises dry mallee scrubland species in areas with no or little disturbance, as well as secondary successional species that establish after agricultural activities have ceased (Beadle and Costin, 1952; Specht, 1972). The vegetation ranges in height from short (<0.5 m) in the areas that have in the past been cleared for grazing and the mallee scrubland understories, to tall (>5 m) in fragments of the less-disturbed mallee scrub. In all areas, bare ground patches are visible, but tends to be more apparent in the previously disturbed areas. A total of 47 species were identified within this community, with a mean of 9.86 (\pm 3.45 SD) species per sample plot, and 0.17 (\pm 0.08 SD) species per m².

The *Rhagodia ulicina*–*Atriplex vesicaria* scrubland community is characterised by species from species group A (Table 5.2). This dry scrubland community is dominated by low-growing shrubs, *A. vesicaria* and *Rhagodia ulicina* (species group A).

1.1 *Rhagodia ulicina*–*Atriplex vesicaria*–*Casuarina pauper* sub-community

This sub-community occurs on roadsides of both transects in the eastern side of the study site (Fig.5.3), and includes both roadsides and farm areas. A total of 16 plant species were identified in this sub-community, with a mean of 6.6 (\pm 2.7 SD) species per sample plot, and 0.09 (\pm 0.04 SD) species per m².

Table 5.1. Species recorded during the surveys conducted in 2018 along the two transects used in this study.

Family	Species	Growth Form	Status
Aizoaceae	<i>Mesembryanthemum crystallinum</i> L.	Prostrate succulent annual forb	naturalised
Boraginaceae	<i>Echium plantagineum</i> L.	Erect annual or rarely biennial forb	naturalised
	<i>Heliotropium europaeum</i> L.	Erect or ascending annual forb	naturalised
Casuarinaceae	<i>Casuarina pauper</i> F. Muell. ex L.A.S. Johnson	Tree	native
Chenopodiaceae	<i>Atriplex vesicaria</i> Heward ex Benth.	Erect or decumbent perennial sub-shrub	native
	<i>Chenopodium curvispicatum</i> Paul G. Wilson	Weak spreading perennial shrub	native
	<i>Dissocarpus paradoxus</i> Ulbr.	Erect annual or short-lived perennial forb or shrub	native
	<i>Enchylaena tomentosa</i> R.Br. var. <i>tomentosa</i>	Prostrate to hemispherical semi-woody perennial sub-shrub	native
	<i>Eriochiton sclerolaenoides</i> (F. Muell.) F. Muell. ex A.J. Scott	Erect or spreading perennial forb or sub-shrub	native
	<i>Maireana brevifolia</i> (R.Br.) Paul G. Wilson	Erect, short-lived perennial shrub	native
	<i>Maireana pentatropis</i> (Tate) Paul G. Wilson	Ascending perennial forb or sub-shrub	native
	<i>Maireana pyramidata</i> (Benth.) Paul G. Wilson	Hemispherical perennial shrub	native
	<i>Maireana sedifolia</i> (F. Muell.) Paul G. Wilson	Hemispherical perennial shrub	native
	<i>Maireana trichoptera</i> (J.M. Black) Paul G. Wilson	Erect perennial forb	native
	<i>Maireana turbinata</i> Paul G. Wilson	Spreading perennial shrub	native
	<i>Rhagodia crassifolia</i> R.Br.	Divaricately branched shrub	native
	<i>Rhagodia spinescens</i> R.Br.	Dense intricately branched spinescent perennial shrub	native
<i>Rhagodia ulicina</i> (Gand.) Paul G. Wilson	Dense intricately branched spinescent perennial shrub	native	

Table 5.1. continued

	<i>Salsola kali</i> L.	Erect or hemispherical annual or biennial forb	naturalised
	<i>Sclerolaena obliquicuspis</i> (R.H. Anderson) Ulbr.	Erect or spreading woody perennial to ca. 40 cm high with hairy branches	native
Compositae	<i>Brachyscome ciliaris</i> (Labill.) Less.	Erect or ascending perennial forb	native
	<i>Carthamus lanatus</i> L.	Erect rigid annual forb	naturalised
	<i>Centaurea melitensis</i> L.	Erect rigid annual forb	naturalised
	<i>Cratystylis conocephala</i> (F. Muell.) S. Moore	Densely branched, spreading perennial shrub	native
	<i>Dittrichia graveolens</i> (L.) Greuter	Erect annual forb	naturalised
	<i>Hyalosperma semisterile</i> (F. Muell.) Paul G. Wilson	Erect or ascending annual forb	native
	<i>Minuria leptophylla</i> DC.	Decumbent to ascending perennial forb	native
	<i>Olearia calcarea</i> F. Muell. ex Benth.	Spreading perennial shrub	native
	<i>Olearia magniflora</i> F. Muell. ex Benth.	Open, sparsely branched perennial shrub	native
	<i>Onopordum acaulon</i> L.	Rosette-forming biennial forb	naturalised
	<i>Sonchus oleraceus</i> L.	Erect annual or rarely biennial forb	naturalised
	<i>Vittadinia gracilis</i> N. Burb.	Rounded much-branched perennial forb or sub-shrub	native
Convolvulaceae	<i>Convolvulus remotus</i> R.Br.	Trailing and twining perennial forb	native
Cruciferae	<i>Carrichtera annua</i> (L.) DC.	Erect annual forb	naturalised
	<i>Lepidium leptopetalum</i> (F. Muell.) F. Muell.	Erect perennial shrub	native
	<i>Lepidium phlebopetalum</i> (F. Muell.) F. Muell.	Decumbent to erect annual to perennial forb	native

Table 5.1. continued

	<i>Sisymbrium irio</i> L.	Erect annual or biennial forb	naturalised
Cucurbitaceae	<i>Citrullus lanatus</i> (Thunb.) Matsum. & Nakai	Trailing annual forb	naturalised
Euphorbiaceae	<i>Chamaesyce drummondii</i> (Boiss.) D.C. Hassall	Prostrate to ascending annual to short perennial forb	native
Geraniaceae	<i>Erodium crinitum</i> Carolin	Decumbent to ascending annual forb	native
Goodeniaceae	<i>Scaevola spinescens</i> R.Br.	Rigid divaricate shrub	native
Gramineae	<i>Austrodanthonia caespitosa</i> (Gaudich.) H.P. Linder	Erect tufted perennial grass	native
	<i>Austrostipa nodosa</i> (S.T. Blake) S.W.L. Jacobs & J. Everett	Erect tufted perennial grass	native
	<i>Avena fatua</i> L.	Erect, tufted or ascending stout annual grass	naturalised
	<i>Hordeum glaucum</i> Steud.	Tufted annual grass	naturalised
Labiatae	<i>Marrubium vulgare</i> L.	Multi-branched perennial shrub	naturalised
	<i>Salvia verbenaca</i> L.	Erect perennial forb	naturalised
Leguminosae	<i>Acacia nyssophylla</i> F. Muell.	Erect or spreading shrub	native
	<i>Acacia oswaldii</i> F. Muell.	Erect or spreading tree or shrub	native
	<i>Medicago minima</i> (L.) Bartal. var. <i>minima</i>	Prostrate to ascending annual forb	naturalised
	<i>Senna artemisioides</i> (Gaudich. ex DC.) Randell ssp. × <i>coriacea</i> (Benth.) Randell	Erect shrub or small tree	native
	<i>Senna artemisioides</i> (Gaudich. ex DC.) Randell ssp. <i>filifolia</i> Randell	Erect shrub or small tree	native

Table 5.1. continued

	<i>Templetonia egena</i> (F. Muell.) Benth.	Erect, multi-stemmed leafless perennial shrub	native
Liliaceae	<i>Asphodelus fistulosus</i> L.	Erect, tufted annual, biennial or perennial forb	naturalised
	<i>Thysanotus baueri</i> R.Br.	Erect, perennial forb	native
Loranthaceae	<i>Lysiana exocarpi</i> (Behr) Tiegh. ssp. <i>exocarpi</i>	Spreading to pendent aerial hemiparasitic perennial shrub	native
Malvaceae	<i>Sida fibulifera</i> Lindl.	Prostrate to procumbent, tomentose forb or woody sub-shrub	native
	<i>Sida intricata</i> F. Muell.	Multi-branched, sprawling or upright, perennial wiry forb or sub-shrub	native
Myoporaceae	<i>Eremophila deserti</i> (A. Cunn. ex Benth.) Chinnock	Erect glabrous shrub	native
	<i>Eremophila glabra</i> (R.Br.) Ostenf.	Prostrate or erect shrub	native
	<i>Eremophila scoparia</i> (R.Br.) F. Muell.	Erect shrub	native
	<i>Myoporum platycarpum</i> R.Br.	Glabrous shrub or small tree	native
Myrtaceae	<i>Eucalyptus oleosa</i> F. Muell. ex Miq. ssp.	Mallee	native
	<i>Eucalyptus socialis</i> F. Muell. ex Miq. ssp.	Mallee or tree	native
Oxalidaceae	<i>Oxalis corniculata</i> L. ssp. <i>corniculata</i>	Erect or ascending, sometimes creeping, perennial forb	naturalised
Pittosporaceae	<i>Pittosporum angustifolium</i> Lodd.	Almost glabrous shrub or tree	native
Proteaceae	<i>Hakea leucoptera</i> R.Br. ssp. <i>leucoptera</i>	Small tree with an open branched crown or bushy shrub	native
Rutaceae	<i>Geijera linearifolia</i> (DC.) J.M. Black	Erect much-branched shrub or tree	native
Santalaceae	<i>Exocarpos aphyllus</i> R.Br.	Much-branched shrub or small tree	native

Table 5.1. continued

Sapindaceae	<i>Alectryon oleifolius</i> (Des f.) S.T. Reynolds ssp. <i>canescens</i> S.T. Reynolds	Small tree	native
Solanaceae	<i>Lycium australe</i> F. Muell.	Intricately branched woody shrub	native
	<i>Nicotiana velutina</i> H.-M. Wheeler	Erect annual forb	native
Zygophyllaceae	<i>Zygophyllum apiculatum</i> F. Muell.	Erect sub-shrub	native
	<i>Zygophyllum aurantiacum</i> (Lindl.) F. Muell.	Low, spreading woody perennial sub-shrub	native
	<i>Zygophyllum crenatum</i> F. Muell.	Stout procumbent or ascending annual forb	native

Table 5.2. A synoptic table of the species groups and the identified sub-communities and the fidelity (Fid.), constancy (Con. - %) and average non-zero cover (Cov. - %) values of the species in each species group.

Community	1.1			1.2			1.3			1.4			2.1			2.2			2.3			2.4			2.5		
No. Relevés	5			8			9			15			4			8			3			10			24		
	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.	Fid.	Con.	Cov.
SCIENTIFIC NAME																											
Species Group A - Characteristic species for the <i>Rhagodia ulicina</i>–<i>Atriplex vesicaria</i> scrubland (1)																											
<i>Atriplex vesicaria</i>	7.8	60	6.0	27.2	88	3.7	20.3	78	3.4	36.1	100	7.2		25	1.0				12.5	67	1.5		20	2.0		4	1.0
<i>Rhagodia spinescens</i>	43.1	60	2.0		13	1.0		11	13.0	4.2	20	2.7				21.2	38	2.0									
<i>Rhagodia ulicina</i>	22.9	60	1.7	15.2	50	4.8	2.4	33	1.3	43.4	87	1.9										30	1.0		13	1.0	
Species Group B - Characteristic species for the <i>Rhagodia ulicina</i>–<i>Atriplex vesicaria</i>–<i>Casuarina pauper</i> sub-community (1.1)																											
<i>Casuarina pauper</i>	100.0	100	36.8																								
Species Group C - Characteristic species for the <i>Rhagodia ulicina</i>–<i>Atriplex vesicaria</i>–<i>Senna artemisioides filifolia</i> sub-community (1.2)																											
<i>Senna artemisioides filifolia</i>				92.8	88	20.7																					
<i>Myoporum platycarpum</i>				59.0	38	6.3																					
<i>Templetonia egena</i>				47.8	25	25.5																					
<i>Scaevola spinescens</i>				61.4	50	1.8																4.7	10	2.0			
Species Group D - Characteristic species for the <i>Rhagodia ulicina</i>–<i>Atriplex vesicaria</i>–<i>Eucalyptus oleosa</i> sub-community (1.4)																											
<i>Eucalyptus oleosa</i>										92.3	87	19.2															
<i>Cratystylis conocephala</i>										34.7	13	38.0															
<i>Lepidium leptopetalum</i>				24.3	25	1.5				44.8	40	3.8															
Species Group E - Common species for subcommunities 1.2, 1.3, and 1.4																											
<i>Zygophyllum aurantiacum</i>				26.9	63	1.2	12.7	44	1.5	56.3	100	1.6							4.0	33	1.0		10	1.0		4	1.0
<i>Geijera linearifolia</i>				13.3	13	13.0	29.7	22	2.0	3.5	7	1.0															
<i>Maireana pentatropis</i>				27.2	50	2.0	2.4	22	2.0	65.9	93	2.0											10	1.0			
Species Group F - Characteristic species for the <i>Medicago minima</i>–<i>Maireana sedifolia</i> shrubland (2)																											
<i>Medicago minima</i>	40	1.0		38	1.0		33	1.3		7	2.0		50	1.0	29.8	100	1.0	5.9	67	1.5	29.8	100	1.0	23.8	92	1.1	
Species Group G - Characteristic species for the <i>Medicago minima</i>–<i>Maireana sedifolia</i>–<i>Nicotiana velutina</i> sub-community (2.1)																											
<i>Nicotiana velutina</i>				13	2.0								91.6	100	1.0											4	1.0
<i>Citrullus lanatus</i>													85.3	75	1.3												
<i>Carthamus lanatus</i>													47.8	25	1.0												
<i>Centaurea melitensis</i>													47.8	25	1.0												
<i>Dittrichia graveolens</i>													47.8	25	2.0												
<i>Echium plantagineum</i>													47.8	25	1.0												
Species Group H - Characteristic species for the <i>Medicago minima</i>–<i>Maireana sedifolia</i>–<i>Maireana pyramidata</i> sub-community (2.2)																											
<i>Maireana pyramidata</i>	35.3	80	10.8	25	2.0	33.7	78	10.4		27	1.0		41.0	88	12.0												

Table 5.2 continued

Species Group I - Characteristic species for the *Medicago minima*–*Maireana sedifolia*–*Senna artemisioides coriacea* sub-community (2.3)

Table 5.2. continued

<i>Senna artemisioides coriacea</i>	20	1.0	25.9	50	13.0										69.7	100	38.0		10	3.0		4	2.0					
Species Group J - Characteristic species for <i>Medicago minima</i>–<i>Maireana sedifolia</i>–<i>Salvia verbenaca</i> sub-community (2.4)																												
<i>Salvia verbenaca</i>	20	1.0										47.6	75	2.3					60.8	90	1.4							
<i>Vittadinia gracilis</i>												53.1	75	1.0					57.7	80	1.1	4	1.0					
<i>Convolvulus remotus</i>												51.1	50	1.5					26.2	30	1.0							
Species Group K - Common species for subcommunities 1.2, 1.3, 1.4, 2.1, 2.2., 2.3, 2.4, and 2.5																												
<i>Sclerolaena obliquicuspis</i>			6.4	75	1.7	0.2	67	1.5		47	2.0		50	7.0	25.2	100	3.1	25.2	100	2.0	10.2	80	1.8	9.6	79	1.5		
<i>Maireana trichoptera</i>			15.2	63	1.4	2.2	44	2.0	8.6	53	1.6					13	3.0	18.2	67	2.5	20.6	70	1.9	15.2	63	1.5		
<i>Austrodanthonia caespitosa</i>	20	1.0	19.8	75	1.7					33	1.4	2.1	50	1.5		38	2.0	13.9	67	1.0	16.3	70	1.6	16.9	71	1.6		
<i>Sida intricata</i>			33.4	50	1.0							9.1	25	1.0	21.3	38	1.0				4.2	20	1.5		8	2.0		
Species Group L - Common species for all subcommunities along the two transects																												
<i>Maireana sedifolia</i>	1.0	80	7.0		63	20.0		44	10.0			60	6.3	18.3	100	4.8		63	4.6	18.3	100	13.0	18.3	100	13.3	18.3	100	10.8
<i>Carrichtera annua</i>		40	1.0		63	2.0		44	1.5			60	1.2	1.2	75	1.7	21.3	100	1.9	21.3	100	2.0	21.3	100	1.7	4.6	79	2.2
<i>Enchylaena tomentosa</i>	23.8	100	1.8	4.8	75	1.3	15.4	87	1.4	13.7	87	1.5		25	1.0		50	1.0	23.8	100	2.0		60	1.3		33	1.0	
<i>Chenopodium curvispicatum</i>		20	1.0	18.3	63	1.4		22	1.5	21.3	67	1.2					13	1.0	45.7	100	1.3		20	1.0		33	1.1	
<i>Austrostipa nodosa</i>		20	1.0	23.3	63	1.4		22	1.0		20	1.3		25	1.0				1.1	33	1.0	29.0	70	1.4	1.1	33	1.1	
Species Group M - Species with low constancy occurring along the two transects																												
<i>Acacia nyssophylla</i>										3.8	7	1.0							49.9	33	1.0							
<i>Acacia oswaldii</i>			33.6	13	1.0																							
<i>Alectryon oleifolius</i>						31.6	11	38.0																				
<i>Asphodelus fistulosus</i>						4.2	11	2.0						55.2	50	3.0					2.8	10	2.0					
<i>Avena fatua</i>																					30.0	10	1.0					
<i>Chamaesyce drummondii</i>														81.5	100	1.5		13	2.0		13.7	30	1.3					
<i>Dissocarpus paradoxus</i>	22.3	20	1.0							12.0	13	1.0					10.7	13	1.0							4	1.0	
<i>Eremophila deserti</i>																			55.5	33	3.0							
<i>Eremophila glabra</i>			33.6	13	3.0																							
<i>Eremophila scoparia</i>										34.7	13	7.5																
<i>Eriochiton sclerolaenoides</i>	27.9	20	1.0			12.2	11	1.0		4.4	7	1.0																
<i>Erodium crinitum</i>															15.1	13	1.0				28.5	20	1.5	0.2	4	2.0		
<i>Eucalyptus socialis</i>						31.6	11	13.0																				
<i>Exocarpos aphyllus</i>										24.4	7	13.0																
<i>Hakea leucoptera</i>																			55.5	33	1.0							
<i>Heliotropium europaeum</i>													60.9	75	2.0	10.6	25	2.0				15.6	30	1.3				

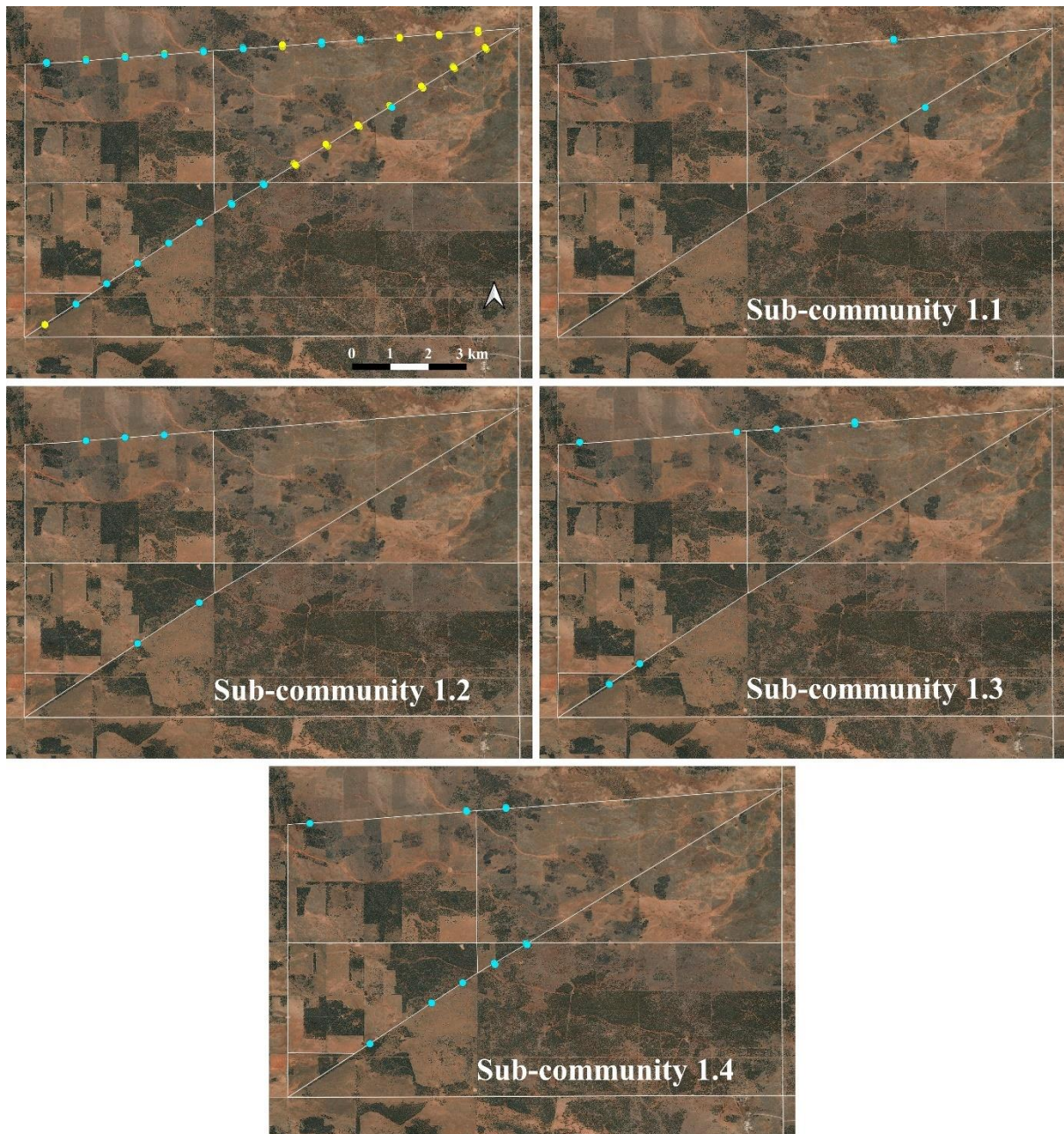


Figure 5.3. The survey plots identified as belonging to the *Rhagodia ulicina–Atriplex vesicaria* scrubland community (blue dots) in relation to those belonging to the *Medicago minima–Maireana sedifolia* shrubland community (yellow dots) along the two transects used in this study, and the positions along the transects where subcommunities 1.1 to 1.4 were recorded.

The *Rhagodia ulicina–Atriplex vesicaria–Casuarina pauper* sub-community is characterised by the dominance of the tree *Casuarina pauper* (species group B), with a constancy of 100%. The small shrubs *Rhagodia spinescens* (species group A), *Maireana pyramidata* (species group H) and *M. sedifolia*, and the forb, *Enchylaena tomentosa* (species group L) are prominent in this sub-community (Table 5.2).

1.2 *Rhagodia ulicina*–*Atriplex vesicaria*–*Senna artemisioides filifolia* sub-community

This sub-community form small stands associated with roadsides and the immediate neighbouring areas where water accumulate after rains along both transects in the western side of the study site (Fig. 5.3). A total of 29 plant species were identified in this sub-community, with a mean of 12.9 (\pm 3.23 SD) species per sample plot, and 0.24 (\pm 0.06 SD) species per m².

The *Rhagodia ulicina*–*Atriplex vesicaria*–*Senna artemisioides filifolia* sub-community is characterised by species from species group C (Table 5.2). The shrub *Senna artemisioides filifolia* (species group C) is characteristic of this sub-community and co-dominates with the shrubs *Templetonia egena* (species group C) and *M. sedifolia* (species group L), with constancies of 25%, 63% and 88% respectively (Table 5.2). The grass *Austrodanthonia caespitosa* and forbs *E. tomentosa* (species group L) and *Sclerolaena obliquicuspis* (species group K) are prominent in this sub-community.

1.3 *Rhagodia ulicina*–*Atriplex vesicaria*–*Maireana pyramidata* sub-community

This sub-community occurs on roadsides and neighbouring flat and undulating plain areas that are degraded as a result of grazing by sheep and road construction at various localities along Bunday Church Road, as well as the at a few localities along Salford Road in the western side of the study site (Fig. 5.3). A total of 20 plant species were identified in this sub-community, with a mean of 6.22 (\pm 2.39 SD) species per sample plot, and 0.1 (\pm 0.04 SD) species per m².

The *Rhagodia ulicina*–*Atriplex vesicaria*–*Maireana pyramidata* sub-community has no characteristic species. This sub-community is characterised by the absence of species from species group B, C and D. The shrub *M. pyramidata* (species group H) and the forbs *E. tomentosa* (species group L) and *S. obliquicuspis* (species group K) are prominent in this sub-community (Table 5.2).

1.4 *Rhagodia ulicina*–*Atriplex vesicaria*–*Eucalyptus oleosa* sub-community

This sub-community occurs on roadsides along both transects on flat and undulating plain areas that have not been degraded by anthropogenic activities in the western and central parts of the study site (Fig. 5.3) and includes both roadsides and farm areas. A total of 30 plant species were identified in this sub-community, with a mean of 10.1 (\pm 2.07 SD) species per sample plot, and 0.16 (\pm 0.05 SD) species per m².

Species from species group D are characteristic for this sub-community. The tree *Eucalyptus oleosa* (species group D) (constancy 87%) and the forb *Maireana pentatropis* (species group E) (constancy 13%) co-dominate the vegetation, while the shrub *Cratystylis conocephala* (species group D), and the forbs *Zygophyllum aurantiacum* (species group E), *Chenopodium curvispicatum* and *E. tomentosa* (species group L) are prominent in this sub-community (Table 5.2).

2. *Medicago minima*–*Maireana sedifolia* shrubland

This community occurs on flat and undulating plain areas that are used for grazing, primarily in the eastern side of the study site, but also at some localities in the western side of the study site (Fig. 5.4). The vegetation comprises dry chenopod shrubland (also known as shrub-steppe) species (Beadle and Costin, 1952; Specht, 1972), with localised stands of species associated with moist conditions in places where water sporadically accumulate. A total of 49 species were identified within this community, with a mean of 7.53 (\pm 3.23 SD) species per sample plot, and 0.12 (\pm 0.08 SD) species per m².

The *Medicago minima*–*Maireana sedifolia* shrubland community is dominated by the forb *Medicago minima* (species group F) and the shrub *M. sedifolia* (species group L) (Table 5.2). The forbs *Carrichtera annua* (species group L) and *S. obliquicuspis*, and the grass *A. caespitosa* (species group K) are prominent in this community (Table 2).

2.1 *Medicago minima*–*Maireana sedifolia*–*Nicotiana velutina* sub-community

This sub-community is localised at roadside localities along Salford Road, as well as in shallow in depressions within farmed areas in the eastern side of the study site (Fig. 5.4). A total of 25 plant species were identified in this sub-community, with a mean of 11.3 (\pm 2.87 SD) species per sample plot, and 0.18 (\pm 0.1 SD) species per m².

The *Medicago minima*–*Maireana sedifolia*–*Nicotiana velutina* sub-community is characterised by species from species group G (Table 5.2). The forb *Nicotiana velutina* is characteristic of this sub-community, with a constancy of 100%. The forbs *Citrullus lanatus* (species group G), *Salvia verbenaca*, *Vittadinia gracilis* (species group J), and *C. annua* (species group L), are prominent in this community (Table 5.2).

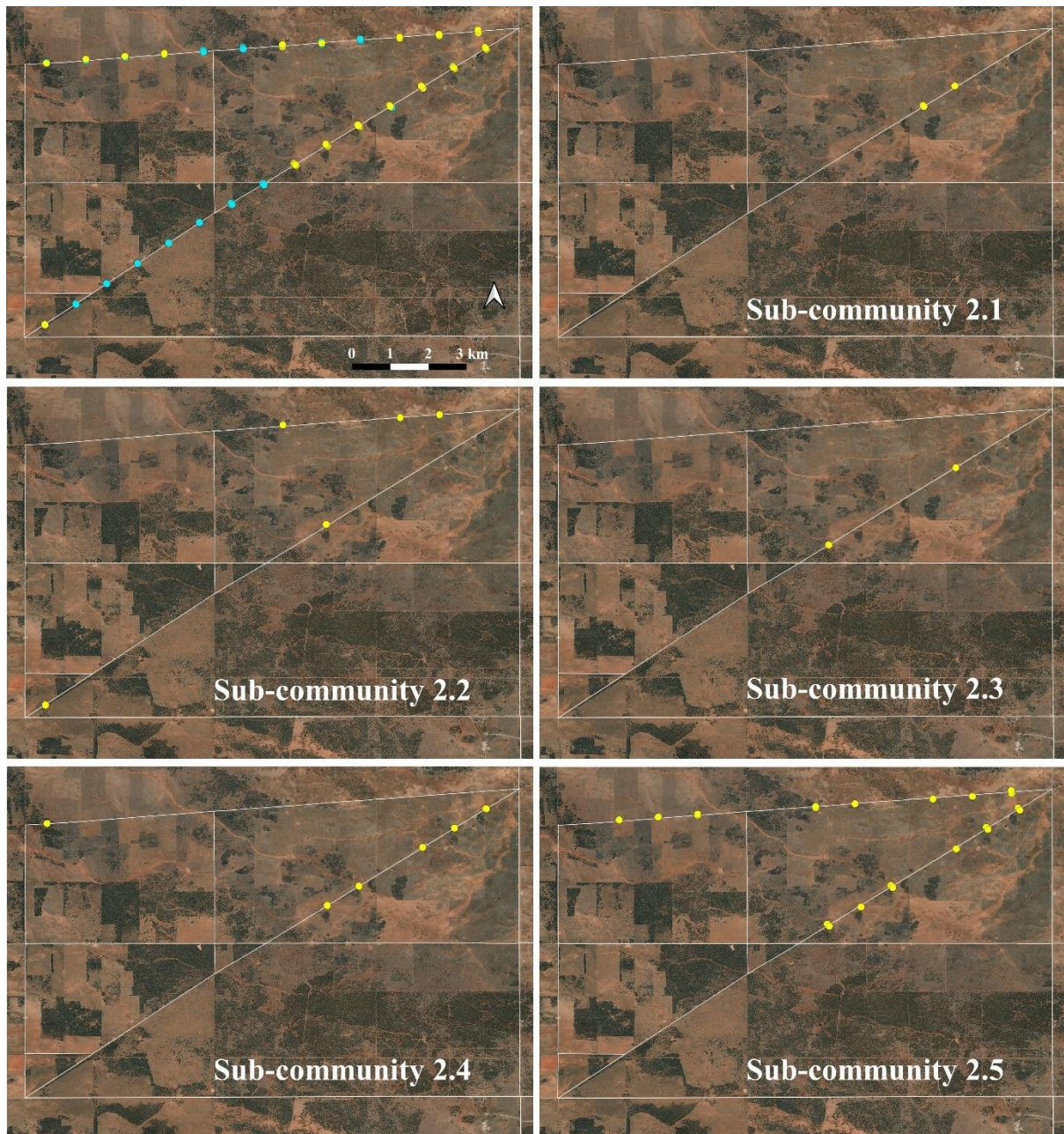


Figure 5.4. The survey plots identified as belonging to the *Medicago minima*–*Maireana sedifolia* shrubland community (yellow dots) in relation to those belonging to the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland community (blue dots) along the two transects used in this study, and the positions along the transects where subcommunities 2.1 to 2.5 were recorded.

2.2 *Medicago minima*–*Maireana sedifolia*–*Maireana pyramidata* sub-community

This sub-community is localised at raised degraded roadsides, as well as in some localities within farmed areas in the central and eastern side of the study site (Fig. 5.4). A total of 16 plant species were identified in this sub-community, with a mean of 6.5 (± 1.77 SD) species per sample plot, and 0.11 (± 0.5 SD) species per m².

The *Medicago minima*–*Maireana sedifolia*–*Maireana pyramidata* sub-community is characterised by the dominance of the shrub *M. pyramidata* (species group H), with a constancy of 88% (Table 5.2). Other prominent species in this community are the forbs *S. obliquicuspis* (species group K) and *C. annua* (species group L).

2.3 *Medicago minima*–*Maireana sedifolia*–*Senna artemisioides* sub-community

This sub-community occurs on roadsides where run-off water accumulates at the side of the road along Salford Road in the eastern side of the study site (Fig. 5.4). A total of 17 plant species were identified in this sub-community, with a mean of 10 (± 2 SD) species per sample plot, and 0.2 (± 0.04 SD) species per m².

The shrub *Senna artemisioides coriacea* is characteristic of the *Medicago minima*–*Maireana sedifolia*–*Senna artemisioides coriacea* sub-community, and co-dominates with the shrub *M. sedifolia* with a constancy of 100%. The grass *A. caespitosa* and the forbs *Maireana trichoptera* (species group K), *C. annua*, *C. curvispicatum*, *E. tomentosa*, and *S. obliquicuspis* (species group L) are prominent in this community (Table 5.2).

2.4 *Medicago minima*–*Maireana sedifolia*–*Salvia verbenaca* sub-community

This sub-community is restricted to roadsides where water accumulates at the edge of the road after rains, primarily along Salford Road in the eastern side of the study site (Fig. 5.4). This sub-community can be sub-divided further into two variants based on the presence or absence of the forbs *Asphodelus fistulosus*, *Chamaesyce drummondii* and *Heliotropium europaeum* (species group G). However, since this study was done during a period of drought, we are taking a conservative approach and therefore group the two variants in the same sub-community. A total of 32 plant species were identified in this sub-community, with a mean of 10.8 (± 3.22 SD) species per sample plot, and 0.22 (± 0.06 SD) species per m².

The *Medicago minima*–*Maireana sedifolia*–*Salvia verbenaca* sub-community is characterised by the absence of species from species group G, and the dominance of the shrub *M. sedifolia* (species group L) with a constancy of 100%, and the prominence of the grasses *R. caespitosum* (species group K) and *Austrostipa nodosa* (species group L), and the forbs *S. verbenaca*, *V. gracilis* (species group J), *M. trichoptera*, *S. obliquicuspis* (species group K), *C. annua* and *E. tomentosa* (species group L) (Table 5.2).

2.5 *Medicago minima*–*Maireana sedifolia*–*Carrichtera annua* sub-community

This sub-community occurs on roadsides and farmed areas on flat plain areas at various localities along Bunday Church Road, but is more restricted along Salford Road in the eastern side of the study site (Fig. 5.4). A total of 23 plant species were identified in this sub-community, with a mean of 5.58 (± 1.79 SD) species per sample plot, and 0.07 (± 0.04 SD) species per m².

The *Medicago minima*–*Maireana sedifolia*–*Carrichtera annua* sub-community has no characteristic species but is characterised by the absence of species from species groups G and H, and the prominence of the grass *R. caespitosum*, the forbs *M. trichoptera*, *S. obliquicuspis* (species group K) and *C. annua*, and the shrub *M. sedifolia* (species group L) (Table 5.2).

Species richness and diversity: An overall average of 0.15 (\pm SD = 0.08) species per 1 m² was recorded in this study, and the species richness of the sub-communities of the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland (community 1) and *Medicago minima*–*Maireana sedifolia* shrubland (community 2) ranged from 16 to 30 (mean \pm SD = 23.8 \pm 5.9) and 16 to 32 (mean \pm SD = 22.6 \pm 5.8) species respectively (Fig. 5.5), but did not differ significantly ($t = 0.258$, $df = 7$, $P = 0.804$), with a 95% confidence interval. Within each sub-community the effective number of species for the Shannon entropy and Gini-Simpson index indicate that there is a degree of dominance (Fig. 5.5). The Shannon entropy and Gini-Simpson index of sub-communities 1.1., 2.2, 2.3. and 2.5 were similar and were lower than those of sub-communities 1.2, 1.3, 1.4, 2.1 and 2.4 (Fig. 5.6). The evenness index of all the sub-communities indicates that there is a degree of dominance, although that of sub-community 2.5 indicates more variability in this sub-community (Fig. 5.6).

Discussion

This study primarily focussed on the roadside plant communities surrounding the Bunday Bore long term study site of sleepy lizards and their ectoparasites. In the majority of the instances where we had access to habitats beyond the roadsides we found that the roadside vegetation is an extension of that of the neighbouring habitat, or at least that they have some affinity with this nearby habitat. The roadside plant community descriptions are therefore largely representative of the plant communities along the two transects, as well as the factors that

determine their composition.

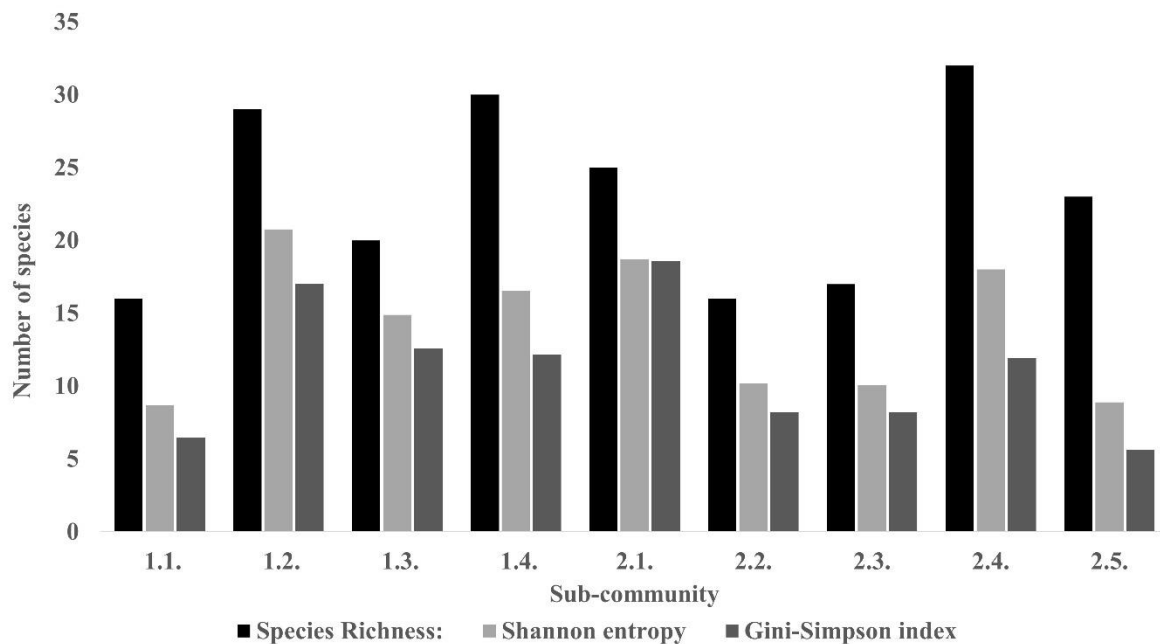


Figure 5.5. The species richness and effective number of species for the Shannon entropy and Gini-Simpson index of the different sub-communities recorded in this study.

Jessup (1948) and Stephens (1956) characterised the soil of this area as calcareous solonized brown soils. The richness of Chenopodiaceae species in the study site is indicative of the semi-arid climate and solonized soils in the area (Foulkes et al. 2013). And the presence of the species, *C. pauper* (sub-community 1.1), *M. pyramidata* (sub-community 2.3) and *M. sedifolia* (all communities), as well as prominent species such as *M. trichoptera* and *S. obliquicuspis* (sub-communities 1.2 – 2.5) throughout the study site is indicative of calcareous sandy-loam soils (Beadle 1981, Cunningham et al. 1981). Even though this indicate that the soils of the area are generally the same, variations in the composition and other abiotic (e.g. moisture) and biotic (e.g. grazing) factors likely determine the composition of the various sub-communities.

The extent of the distribution of the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland (community 1) and *Medicago minima*–*Maireana sedifolia* shrubland (community 2) are determined to a large extent by topography, soil depth and precipitation. *Atriplex vesicaria* and *M. sedifolia* are widespread in xeric habitats of South Australia, and although they may co-occur in the same communities, they usually sole dominate, often in adjacent communities (Jessup 1948, Specht 1972). Both species are adapted to arid habitats and can absorb moisture through their leaves, but *A. vesicaria* is usually associated with shallow soils with restricted

water penetration whereas *M. sedifolia* tend to favour soils that are deeply permeable to water penetration (Wood 1925, Carrodus and Specht 1965, Specht 1972). *Atriplex vesicaria* and *M. sedifolia* are also greatly influenced by land use/grazing (Heshmatti et al. 2002, Facelli and Springbett 2009). *Maireana pyramidata*, which is a prominent species in all the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland sub-communities (1.1 – 1.4) but limited to only the *Medicago minima*–*Maireana sedifolia*–*Maireana pyramidata* sub-community (2.2), requires little moisture for germination and tends to establish after periods of relatively good rain (Heshmatti et al. 2002, Duncan et al. 2019). This species is therefore an indicator that the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland (community 1) experiences higher precipitation and/or run-on (i.e. water flow on to it from neighbouring areas) than the *Medicago minima*–*Maireana sedifolia* shrubland (community 2) does.

The dominant and prominent species from sub-communities 1.1 and 1.4 from the *Rhagodia ulicina*–*Atriplex vesicaria* scrubland, as well as those from sub-communities 2.4 and 2.5 from the *Medicago minima*–*Maireana sedifolia* shrubland were also noted in plant associations described by Jessup (1948) and Specht (1972), indicating that these sub-communities are remnants of the original plant communities of this locality. These sub-communities were recorded in areas used for grazing sheep (*Ovis aries*), and feral goats (*Capra hircus*) are present in even the localities that are not used for agricultural purposes (Norval, personal observation). The forb *S. obliquicuspis* tends to become dominant as a result of disturbances, such as overgrazing in halophytic shrublands (Beadle 1981). As the dominance of this species in the above stated sub-communities is lower than in the associated sub-communities 1.2, 1.3, 2.2 and 2.3, it can be concluded that sub-communities 1.1, 1.4, 2.4 and 2.5 experience a degree of disturbance, but less than the other sub-communities.

Sub-communities 1.2 and 2.3 are localised and restricted primarily to the roadsides (Fig. 5.7 and 5.8) and are characterised by the dominance of two sub-species of the shrub *Senna artemisioides*, both of which prefer loam or sandy loam soils and tend to establish in disturbed habitats following large rainfall events (Tiver and Andrew 1997, Sinclair 2005, Gavel et al. 2010). These sub-communities are therefore indicative of areas with well-drained soils that tend to be moist as a result of road edge effect during periods of good rainfall. As these sub-communities are largely restricted to roadsides, it can be deduced that the disturbance that drive them is likely road construction and not grazing because the neighbouring paddocks are fenced

to prevent stock from venturing into the road.

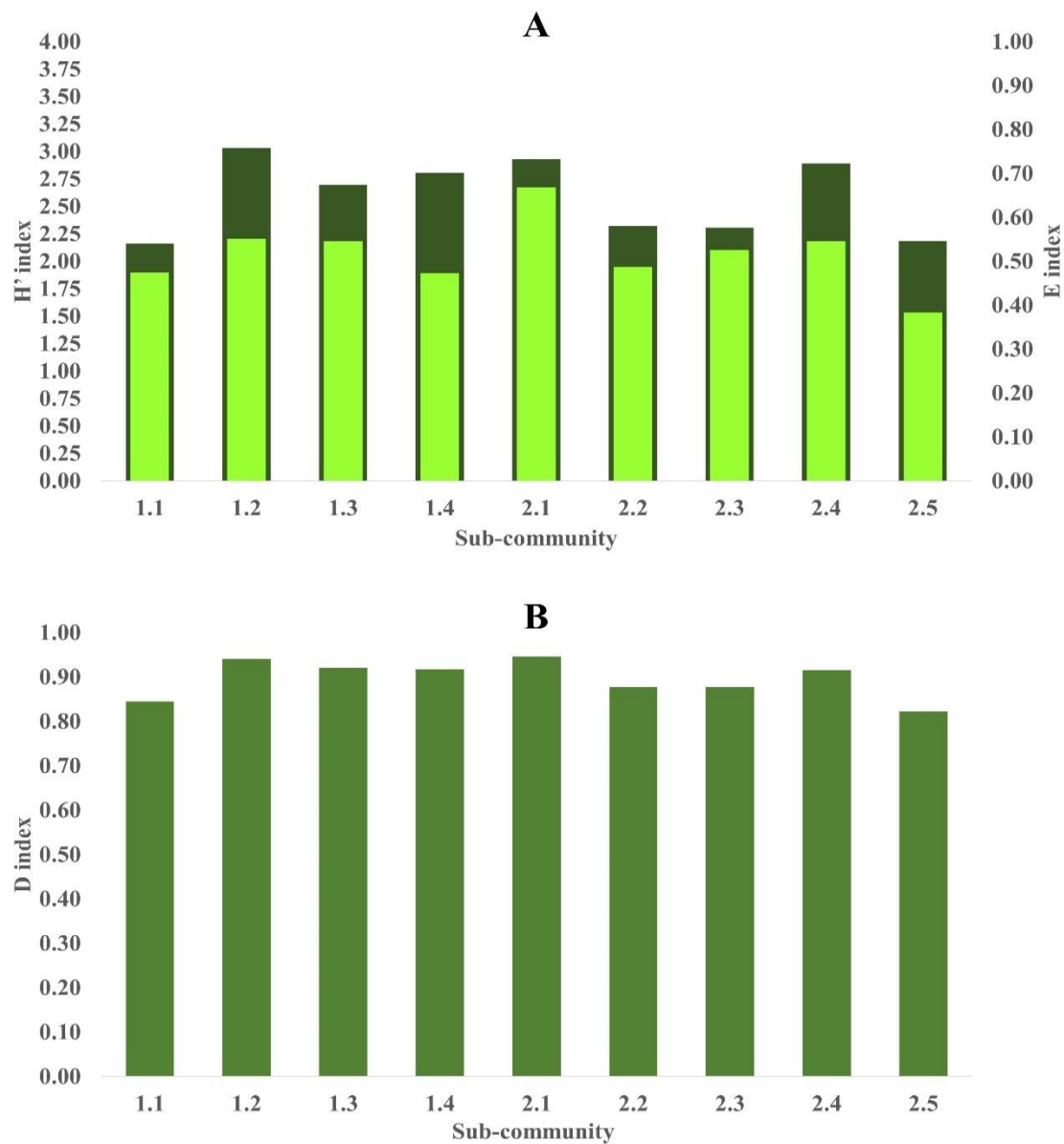


Figure 5.6. The Shannon entropy (H' Index), evenness index (E index) (A) and Gini-Simpson index (D' index) (B) of diversity of the different sub-communities recorded in this study.

Sub-communities 1.3 and 2.2 occur at roadsides as well as within neighbouring habitats. The shrub *M. pyramidata* is the characteristics species of sub-community 2.2. and is also prominent in sub-community 1.3. The disturbance to the soil surface in areas with adequate moisture promote the establishment of *M. pyramidata* (Heshmatti et al. 2002, Facelli and Springbett 2009, Duncan et al. 2019). The prominence of *M. pyramidata* in sub-communities 1.3 and 2.2 indicates that these plant communities are the result of disturbance such as over grazing,

trampling and/or road construction, and that these areas are moist during periods of good rainfall.

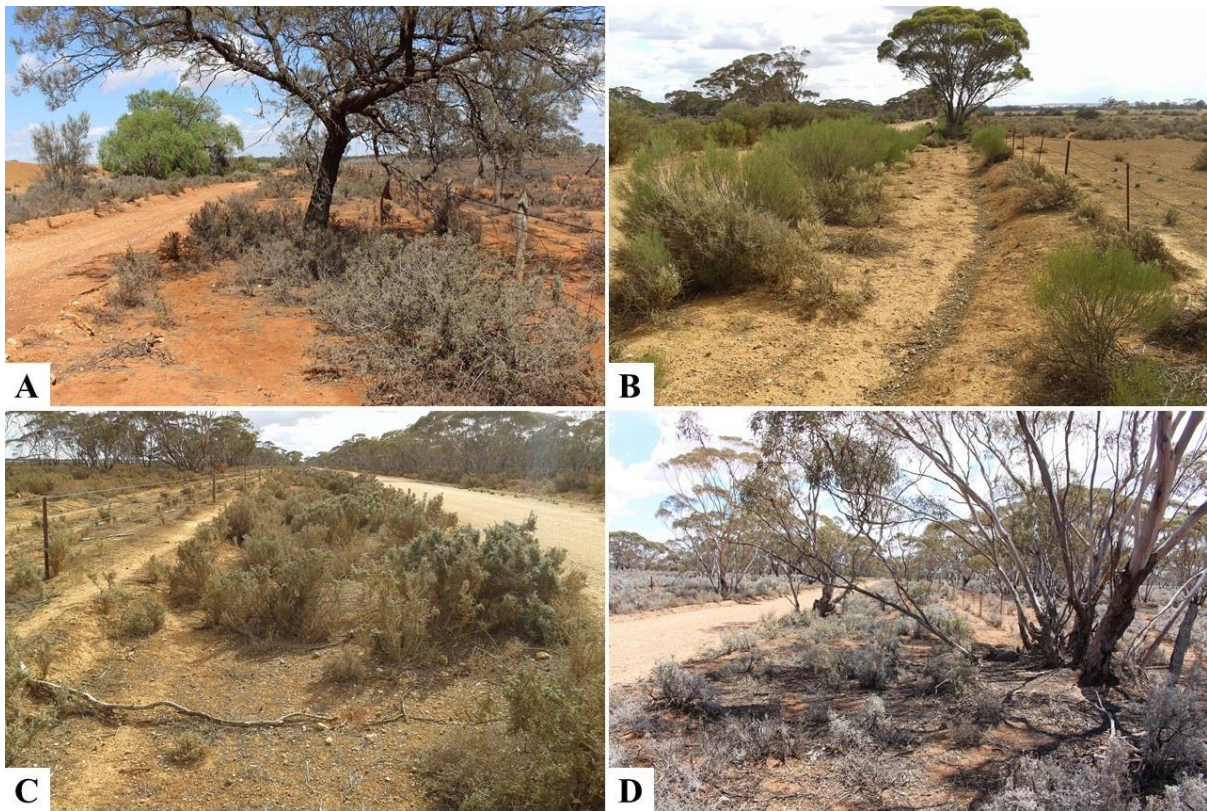


Figure 5.7. Examples of sub-communities 1.1 (A), 1.2 (B), 1.3 (C) and 1.4 (D). Note how sub-communities 1.2 and 1.3 are restricted to the roadsides.

Sub-community 2.1 is localised and restricted to the roadsides and areas within neighbouring habitats where water tends to accumulate during rains. The characteristic species of this sub-community, *N. velutina*, as well as some of the prominent species, such *Citrullus lanatus* and *Salvia verbenaca*, are usually associated with disturbed moist sites with sandy soils (Cunningham et al. 1981). This community is therefore indicative of areas that remain moist for some time after good rainfall, and that are disturbed by road construction or grazing.

Sub-community 2.4 is restricted to roadsides and has a strong affinity to sub-community 2.5, from which it is differentiated by the presence of species from species group J. These species tend to grow at the roadside edge (Fig. 5.8) where water accumulates after rains. The distinction between these two sub-communities is therefore the roadside effect in sub-community 2.4, which is absent in sub-community 2.5. The prominence of the forbs *S. obliquispis* (species group K) and *C. annua* are indicative of disturbance by road construction and overgrazing

(Beadle 1981, Cooke et al. 2011), and the dominance of the shrub *M. sedifolia* (species group L) indicates xeric conditions in areas where these sub-communities are found.

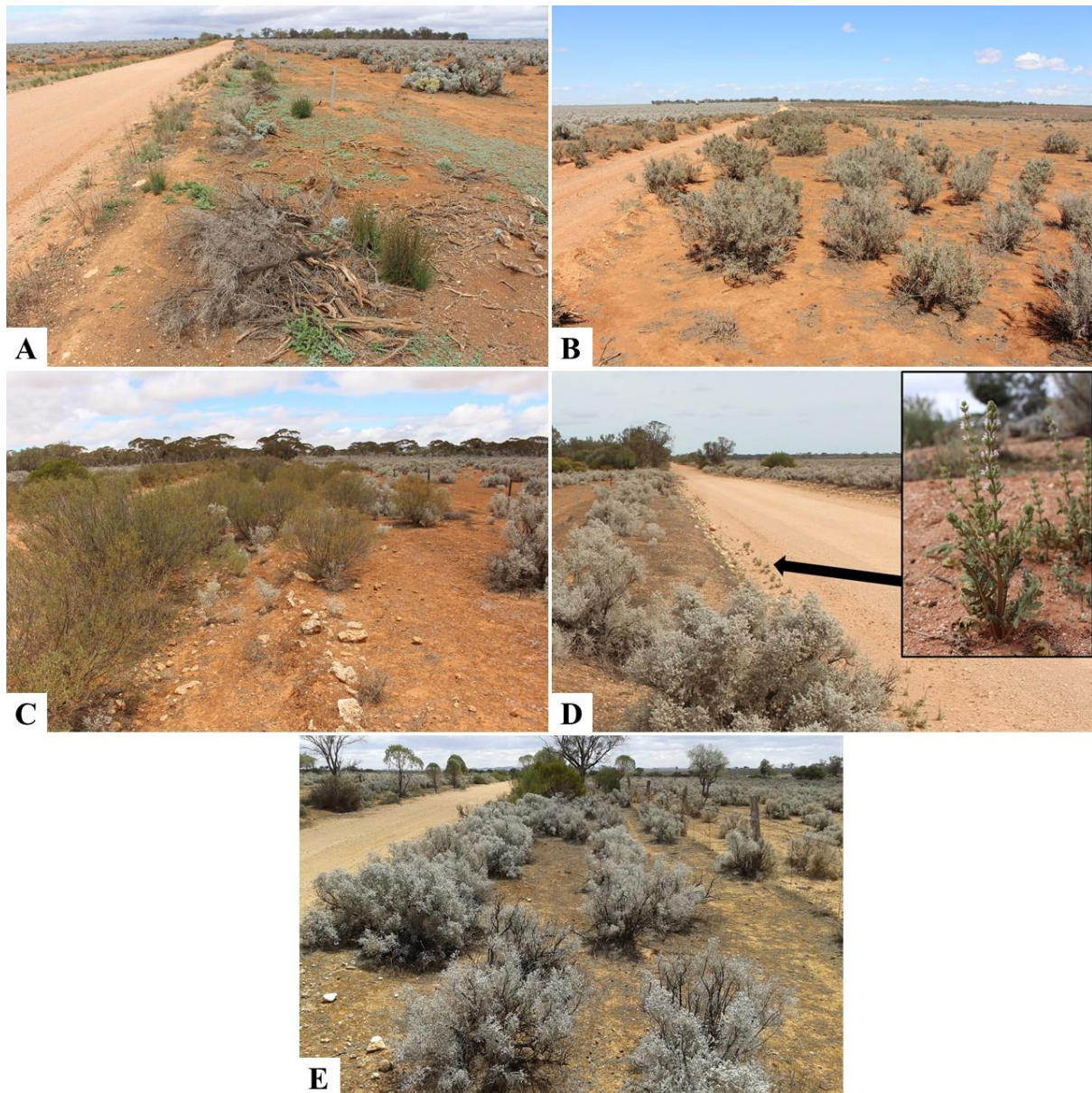


Figure 5.8. Examples of sub-communities 2.1 (A), 2.2 (B), 2.3 (C) 2.4.1 (D) and 2.4.2 (E) and 2.5 (F). Note how the diagnostic species of this sub-community, *Salvia verbenaca* (inset), is primarily distributed along the road edge.

Conclusion

This study provided a description of the plant communities along two transects at a study site in the Mid North of South Australia utilised for long-term studies on the ecology of sleepy lizards and their parasites. Two major plant communities were identified consisting of nine sub-communities with a mosaic distribution pattern. The roadside vegetation is usually an

extension of that of the neighbouring habitat. The largest part of the study area has been exposed to different degrees of disturbance. This analysis of the plant communities will assist in future studies of the parasite populations. Since this study was conducted during a prolonged drought period it is imperative that long-term monitoring of these survey plots be continued, especially after good rainfall, to be able to add new species to the list.

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Chapter 6

The distribution of ecto- and endoparasites of sleepy lizards (*Tiliqua rugosa*) sampled across an ecological gradient in the Mid North region of South Australia and their habitat association

Note to examiners

The distribution of parasites is not uniform throughout their geographic range. Variations in limiting factors, such as precipitation, in an area can create habitats that are favourable to some species, not for others. In this chapter I consolidate all the data I gathered from the sleepy lizards I caught in the years 2017 to 2019 to map the distribution of the ecto- and endoparasites I recorded. I then use a sub-sample of this data to examine associations between the various parasite species and habitat categories (derived from the results of Chapter 5), which could explain the distribution patterns of particular parasite species. This chapter is a manuscript that I am intending to submit to the journal *Transactions and Proceedings of the Royal Society of South Australia* and it is therefore formatted according to the specifications of the journal. The referencing style was however changed to make it consistent with that of the other parts of my thesis. The numbering of the figures and tables were changed for clarification and so that they appear chronologically in the chapter. I took the photographs and therefore own the copyright. The raw data that were collected from the lizards that were used in this study are provided at the end of the thesis (Appendix 7.6). The font size in the appendices is reduced to keep the pagination to a minimum.

Abstract

Parasites play vital roles in ecosystems but are often overlooked in ecological studies. As a result even fundamental information, such as their distribution is often lacking or incomplete. In this study we studied sleepy lizards (*Tiliqua rugosa*) over a three-year period at a long-term study site in the Mid North region of South Australia and examined them for ectoparasites, gastrointestinal helminths parasites and haemoparasites. The distribution of the recorded parasites were mapped, and then tested for an associations between their distribution and habitat categories of the study site. The study revealed that the parasite community of the lizards at this locality is more complex than has previously been reported. The study reconfirmed an association between more arid habitats and the tick *Amblyomma limbatum*, and more mesic habitats and the tick *Bothriocroton hydrosauri*. However, no association among any of the other recorded parasites and the different habitats was found, indicating the drivers of the other parasites' distribution are different to that of these two tick species. This study indicates that habitat modifications can alter the distribution of the ticks *A. limbatum*, and *B. hydrosauri*.

Introduction

Parasites are important components of ecosystems, as parasites play vital roles in host population dynamics, energy flow and/or interspecies competition (Marcogliese 2004, Hudson et al. 2006, Wood and Johnson 2015). They can also influence the evolution of the host species and speciation through their influence on mate selection (Clayton 1991, Karvonen and Seehausen 2012, Brunner et al. 2017). However, parasites are often overlooked in ecological studies, and even fundamental information, such as their distribution and which host species they infect or infest is often lacking or incomplete (Gómez and Nichols 2013).

The geographic distribution of a particular parasite species often does not correspond with that of its hosts and in some instances may exceed it. This is because some parasite species are not host specific and therefore may parasitise different host species throughout their range (Cumming 1999, Poulin et al. 2011, Clark et al. 2018). Alternatively, in many instances the distribution of the parasite species is more restricted than that of the host (Poulin et al. 2011, Clark et al. 2018). This is because part of the life cycle of most parasite species is off or outside of the host in the environment where environmental conditions will influence its survival and as a result play a role in its distribution (Cumming 2002, Poulin et al. 2011, Clark et al. 2018).

Subsequently, vegetation descriptions are often incorporated with zoological studies as the plants are a food source for some host animals, and at the same time make up the physical structure of the habitat and create the microclimate the animals inhabit (e.g. Hirst 1975, Jellinek et al. 2004, Wood et al. 2010, Suggitt et al. 2012). Vegetation often determines the distribution of potential host species and creates the microclimate favoured by the parasite. In many habitat studies, vegetation cover and types have been used to examine the distribution of parasites such as ticks (Londt and Whitehead 1972, Schwarz et al. 2009). To a large extent, climate is a determining factor of vegetation types (Box and Fujiwara 2013), explaining why, at a regional scale, vegetation types could be used as suitable predictors for the distribution of certain tick species (Lindström and Jaenson 2003, Ledger et al. 2019, Talbot et al. 2019, Kwak et al. 2021) although Cumming (2002) suggests climate is a better predictor of the distribution of ticks. Vegetation types have also been used in studies pertaining to the distribution of other parasitic arthropods, such as chigger mites (Matthee et al. 2020) and mosquitoes (Young et al. 2021).

The sleepy lizard (*Tiliqua rugosa*) is an endemic lizard species that inhabits a variety of habits

throughout most of the southern parts of Australia (Cogger 2014). It is a relatively well-studied species, to a large extent due to the long-term ecological studies on a population at a study site north of Bower and Mt. Mary in the Mid North region of South Australia (Fig. 1) and on the distribution of the ticks that infest them, initiated by the late Prof. Michael Bull (Godfrey and Gardner 2017, Norval and Gardner 2020). A variety of parasite species have been reported to parasitise *T. rugosa* (Norval et al. 2019), but the distributions of most of these parasites are poorly known, and only the ticks *Amblyomma albolimbatum* Neumann, 1907, *Amblyomma limbatum* Neumann, 1899 and *Bothriocroton hydrosauri* (Denny 1843) have been studied to some extent (Smyth 1973, Bull et al. 1981, Petney 1981, Sharrad and King 1981, Andrews and Petney 2008). At the study site north of Bower and Mt. Mary, in addition to the distribution of the ticks *A. limbatum* and *B. hydrosauri*, that of the cestode *Oochoristica trachysauri* (MacCallum 1921), nematode *Thelandros trachysauri* Johnston & Mawson, 1947, and haemogregarine *Hemolivia mariae* Smallridge & Paperna, 1997 have been assessed in parts of the study site (Bull and Smyth 1973, Bull et al. 1981, Smallridge 1998, Gyawali 2011). However, non targeted distribution studies for all the parasites have been done throughout the study site, and even though Sharrad (1979) and Bull et al. (1981) made reference to an association between habitat types and the distribution of the ticks, the association has not been empirically examined for the ticks or any of the other parasites. The aims of this study therefore were to (1) examine sleepy lizards throughout the study site for ectoparasites, gastrointestinal helminths parasites and haemoparasites, and map the distribution of the recorded parasites and; (2) test for an association between the distribution of these parasites and habitat types of the study site.

Material and methods

Study site: The study was conducted at the Bunday Bore Station study site, ca. 160 km north of Adelaide, in the Mid North region of South Australia (Fig. 6.1), and the surrounding areas as part of the ongoing long-term study on sleepy lizards and their ticks. This region has a semi-arid climate, with an average annual rainfall of less than 250 mm, of which most usually falls during the austral winter (Smyth 1973). Large areas have been cleared for agriculture and grazing, and the grassy arid mallee scrub and the chenopod shrubland, which used to dominate the south-western and north-eastern side of the study site respectively, remains only in fragments (Jessup 1948, Smyth 1973).



Figure 6.1. The Bundey Bore study site in relation to Adelaide (star), South Australia, and the layout of the dirt roads (white lines in inset) that are used as the transects for the long-term monitoring of sleepy lizards (*Tiliqua rugosa*), and position of the two transects that were used for investigating the association between habitat categories and the parasite community assemblage (yellow lines in inset).

Lizards sampling and measuring: Sleepy lizards were found using the random encounter survey method (Bull 1988) by driving slowly along the dirt roads passing through the study site (i.e. established transects), and looking for lizards crossing the road or at the road sides, during the period late August 2017 to mid-December 2019, primarily in the months August to December of each year when lizards are expected to be most active (Bull 1987b, Bull et al. 1991, Firth and Belan 1998). The GPS location of each capture locality was recorded with a Garmin Etrex 10 GPS receiver. Each capture was regarded as an independent event because

the lizards were not recaptured on the same day and the main aim of this study was to determine the distribution of the parasites, which would not be negatively influenced by recapturing the same lizard multiple times. The lizards were captured by hand and the snout-vent length (SVL) was measured (to the nearest mm) with a transparent plastic ruler. The sexes can not easily be distinguished in sleepy lizards in the field. Generally the head sizes of the sexes are sexually dimorphic, but there is a degree of overlap (Bull and Pamula 1996) and as a result, secondary features, such as the extension of the hemipenes have been applied for confirmation (Bull 1987b). However, juvenile females can have hemipenis-like structures that are retracted into the base of the tail (Ziegler and Böhme 1997). For this study no attempts were therefore made to distinguish between the sexes. Individual lizards were identified by their unique toe-clips, and newly captured lizards were individually marked by toe-clipping as part of the ongoing long-term study on this species at this study site, initiated in 1982 (Bull 1987b, Godfrey and Gardner 2017).

Ectoparasites: The area around the cloaca, in the ear and nasal openings, and under the scales of the body of the animal were visually examined for attached ticks before releasing the lizard back into the wild at the point of capture. If observed, ticks were counted and their attachment sites were noted, after which voucher specimens were removed with a pair of fine forceps. With the exception of a few *O. gurneyi* specimens that were to be used in moulting experiments, all the collected tick specimens were preserved in 95% molecular grade ethanol. The keys in Roberts (1970) were used to verify the species of the sampled ticks.

In 2019 a subsample of lizards was also examined for the presence of snake mite, *Ophionyssus natricis* (Gervais 1844). This parasite is easily overlooked during visual inspections, so the infestation by *O. natricis* was determined by brushing the lizards with a fine paintbrush (Norval et al. 2020). Mite specimens were preserved in 70% ethanol and identified according to the descriptions in Camin (1953), Domrow (1988) and Moraza et al. (2009).

Gastrointestinal parasites: As a response to being handled sleepy lizards will sometimes spontaneously defecate (Gyawali et al. 2013). In 2017 and 2018 such faecal samples were collected and placed into a 50 ml Falcon centrifuge tube. At the end of the day the faecal sample, or part of it, was placed inside a 25 ml single-use syringe so that the syringe was up to ca. half full. Normal saline solution (0.9%) was drawn up into the syringe to liquify the faecal sample, after which the plunger was pressed down to force ca. 3 ml of the liquified

faecal sample out into a 10 ml Falcon centrifuge tube. Six millilitres of DESS, a non-hazardous solution containing dimethyl sulphoxide, disodium EDTA, and saturated NaCl (Yoder et al. 2006), was added as a preservative for helminth and DNA samples. In 2018, in addition to the sample preserved in DESS, a second sample was prepared in the same manner, but 6 ml of SAF, a 10% formalin solution for gastrointestinal parasite examinations (Marti and Escher 1990), was used as the preservative. The remaining faecal material in the syringe was then poured out into a petri dish and searched under a dissection microscope for helminths. When helminths were present, adult specimens were collected and preserved in 70% ethanol for morphological examinations. Some cestode proglottids were sent to Prof. Ian Beveridge (Faculty of Veterinary and Agricultural Sciences, University of Melbourne, Melbourne, Australia) to verify the species, and the nematodes were identified based upon the reference keys of Anderson et al. (1974) and the descriptions in Johnston and Mawson (1947). The liquified faecal samples preserved in DESS and SAF were used for preparing direct smears and faecal flotation, using magnesium sulphate (MgSO₄) solution with a specific gravity of ca. 1.2 (Klingenberg 1993), which were examined under an Olympus® CH30 compound microscope at magnifications of ×200 and ×400. The different species were recorded as present or absent, and no attempts were made to quantify the eggs or adults as the aim of this study was to determine the presence of the various helminths for mapping their distribution in the study site.

Haemoparasites: Throughout the study period, attempts were made to obtain blood samples for the preparation of smears (coverslip-slide method) from the captured lizards, unless it was a recaptured lizard from which a blood sample had been taken less than two weeks prior. Blood samples were taken from the caudal vein by venipuncture with a disposable Livingstone® 23G needle per 1-mL syringe for adult and subadult lizards (SVL ≤ 20 cm) or Livingstone® 21G needle per 1-mL syringe for juveniles (SVL > 20 cm) (Bull 1995, Lancaster et al. 2012, Sykes and Klaphake 2015). The blood film smears were prepared immediately in duplicate and air-dried in the field, fixed for ca. 5 minutes in absolute methanol (Smallridge and Bull 1999). Initially a slide from each sample pair was stained with a Giemsa stain, but subsequently was switched to using a Wrights-Giesma stain as this stain also makes it possible to determine the presence or absence of haemoparasites, but also for possible future cytological studies. All staining was done according to the manufacturer's specifications and cover-slipped with Eukitt® Quick-hardening mounting medium (Sigma-

Aldrich). Each slide was scanned under 100× oil immersion objective (i.e. ×1,000 magnification) on an Olympus® CH30 compound microscope for 6 minutes, examining 6,000–10,000 erythrocytes, to determine the presence or absence of haemoparasites. When no haemoparasites were detected after 6 minutes of screening, the blood smear was considered to be uninfected. No attempts were made to determine the degree of parasitemia.

Comparison of the parasite community across the ecological gradient: To compare the parasite community across the ecological gradient, only the sleepy lizards that were caught in 2018 along Bunday Church Rd. and Salford Rd. were used in the analyses (Fig. 1). Only one capture record of each lizard was used in the analyses, so if a lizard was caught more than once in 2018, only the capture event in which the most complete parasite data (i.e. ectoparasites, endoparasites and/or haemoparasites) were recorded was included.

For every lizard that was included, the habitat surrounding the locality where the lizard was captured was assigned to one of the following categories based on the plant communities as described Chapter 5 on both sides of the road (Table 6.1):

1. The surrounding habitat consists of sub-community 1.4 on both sides of the road.
2. The surrounding habitat consists of a combination of sub-communities 1.3 and 1.4.
3. The surrounding habitat consists of sub-community 1.1 on both sides of the road, or combinations of sub-communities 1.1 and 1.2 or 2.1; or 1.2 and 2.5.
4. The surrounding habitat consists of sub-community 2.2 on both sides of the road, or combinations of sub-communities 1.1, 2.4 and 2.5.; 2.2. and 2.5; 2.1, 2.3, 2.4 and 2.5; or 2.3 and 2.5.
5. The surrounding habitat consists of a combination of sub-communities 2.4 and 2.5.

Analyses and mapping: The SVL measurements from the lizards were not normally distributed, so a Mann-Whitney *U* test, with $\alpha = 0.05$, was used for comparing the SVLs of lizards sampled in different years, using the Prism 8 package (Graphpad Software, San Diego, California, USA). The prevalence (the number of infected or infested hosts in the population) of the various parasite species was described as in Bush et al. (1997).

For analyses, we first created a parasite dataset, containing presence (score of 1) or absence (score of 0) of the parasite species in and/or on each sleepy lizard at each capture along Bunday Church Rd. and Salford Rd. in 2018. When faecal and/or blood samples were not collected, no

Table 6.1. The habitat features of the five habitat categories the localities at which lizards were caught along Bunday Church Road and Salford Road in 2018 were assigned to.

Category	Habitat features
1	The surrounding habitat is a mallee scrubland that has not been extensively disturbed
2	The surrounding habitat is an area that has been extensively disturbed, and is often dominated by black bluebush (<i>Maireana pyramidata</i>), bordering a less disturbed mallee scrubland
3	The surrounding habitat is a stand of black oak (<i>Casuarina pauper</i>); a stand of black oak bordering a roadside dominated by fine-leaf desert cassia (<i>Senna artemisioides filifolia</i>) or velvet tobacco (<i>Nicotiana velutina</i>) and other forbs; or a roadside dominated by desert cassia bordering a chenopod shrubland dominated by blue bush (<i>Maireana sedifolia</i>), that is periodically used for grazing by sheep
4	The surrounding habitat is a localised stand of black oak, or a localised roadside area dominated by black bluebush, broad-leaf desert cassia (<i>Senna artemisioides coriacea</i>) and/or wild sage (<i>Salvia verbenaca</i>) and other forbs, surrounded by a chenopod shrubland, dominated by blue bush, that is periodically used for grazing by sheep
5	The habitat on both sides of the road is a chenopod shrubland that is periodically used for grazing by sheep

scores were entered into the dataset. We used a two-factor experimental design of transects (2 levels, Bunday Church Rd. and Salford Rd.; fixed factor), and habitat category (5 levels; habitat categories 1 to 5; fixed factor) for the parasite community datasets. PERMutational ANalysis Of VAriance (PERMANOVA) was used on the full design for either univariate (Euclidean distances) or multivariate (Bray-Curits similarities; +1 dummy variable) analyses, with 9999 permutations to detect differences at the $p = <0.05$ level, using PRIMER v7 software (Clarke et al. 2014), with the PERMANOVA+ add-on (Anderson 2001). If fixed factors or interactions were significant, we conducted multiple pair-wise tests to identify which groupings contributed to the differences from PERMANOVA, and similarity percentage routine (SIMPER) was used to identify the taxa responsible for contributing most to the similarities within, and dissimilarities between factors (Clarke et al. 2014).

The open software QGIS (Version 3.16.3; available at <http://qgis.osgeo.org>) was used for creating the study site and parasite species distribution maps.

Results

Lizards: We caught 557 lizards during the study period, primarily in the months September to December of each year. Sixty-one of the lizards were recaptured multiple times (range = 1 – 4; mean \pm SD = 1.3 ± 0.6), and the period between recaptures ranged from 5 to 741 days (mean \pm SD = 230.5 ± 194.1). The SVL of the lizards ranged from 177 mm to 341 mm, with a mean and standard deviation of 300.7 mm and 24mm respectively. The SVLs of the lizards caught in each year (Table 6.2) did not differ significantly ($H = .414$, $df = 2$, $P = .414$).

Table 6.2. The numbers (n) and the range, mean and standard deviation (mean \pm SD) of their snout-vent lengths (SVL; in mm) of sleepy lizards (*Tiliqua rugosa*) caught in each year for this study.

Year	n	Range	Mean \pm SD
2017	149	186 – 338	297.1 \pm 29.0
2018	214	177 – 341	303.0 \pm 20.5
2019	194	202 – 334	300.0 \pm 23.3

Ectoparasite infestation: All the lizards that were caught during the entire study period were examined for the presence of ticks and 75.1% were infested by at least one species. The majority of the lizards were infested by *Amblyomma limbatum* (43.3%) and 19.9% of them were infested by *Bothriocroton hydrosauri* (Fig. 6.2). A small number of lizards (9.3%) were co-infested by *A. limbatum* and *B. hydrosauri* (Fig. 6.2). The soft tick *O. gurneyi* was rarely found infesting the lizards and usually co-infested the lizard with one or both of the other tick species (Fig. 6.2). On the 17th of February 2019, one lizard was found infested by a chigger (Trombiculidae) and on the 17th of August 2019 another lizard was found infested by *O. natricis*. *Amblyomma limbatum* had a more restricted distribution than that of *B. hydrosauri* and *O. gurneyi* (Fig. 6.3).

Gastrointestinal parasite infection: We collected faecal samples from 84 of the lizards that were caught in 2017 and 2018, and of these in only three (3.6%) did we not find any indications of helminth infections. In all the other scats (96.4%) we observed the eggs and/or adults of the nematode *T. trachysauri*. In nine (10.7%) of the scats we observed the eggs and/or the

proglottids of the cestode *O. trachysauri*, which seemed to have a more restricted distribution than that of *T. trachysauri* (Fig. 6.4).

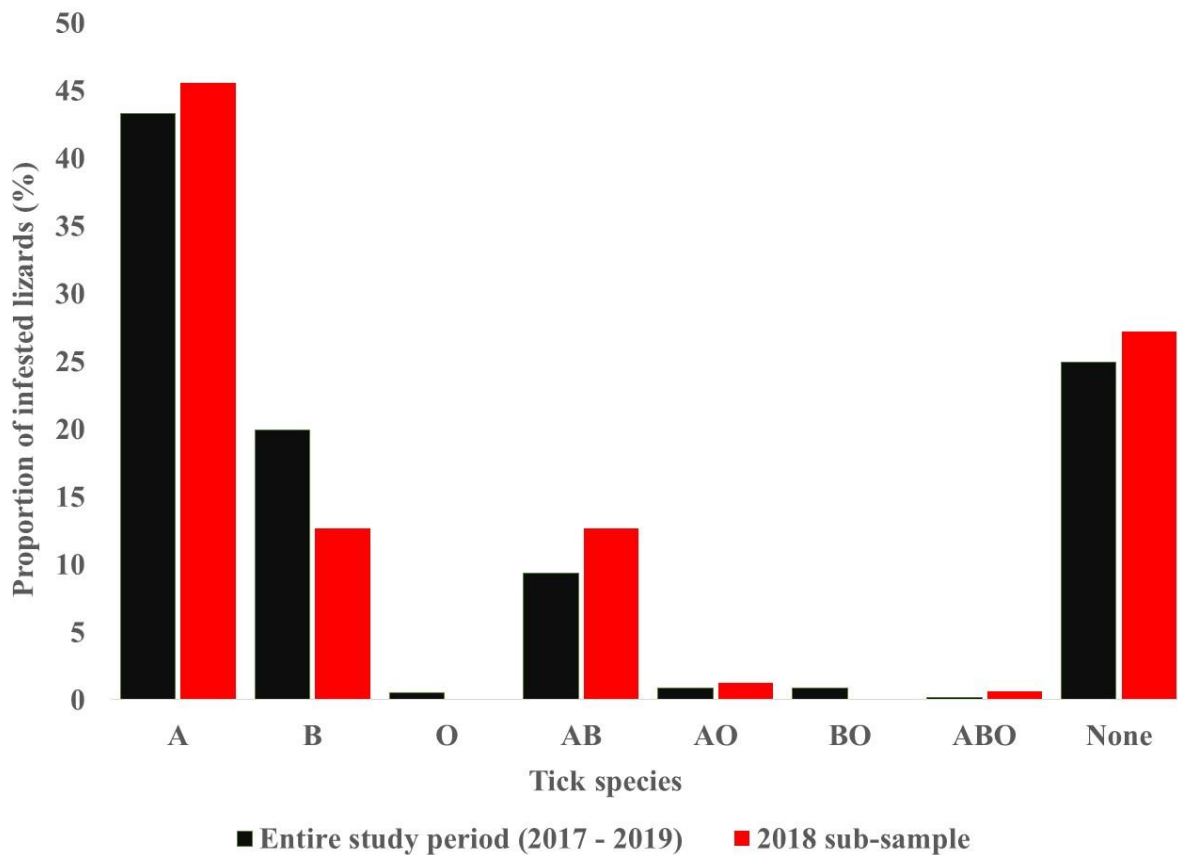


Figure 6.2. The proportion of lizards from the entire study period and the sub-sample of lizards collected in 2018 that were infested by *Amblyomma limbatum* (A), *Bothriocroton hydrosauri* (B) and/or *Ornithodoros gurneyi* (O).

Haemoparasite infection:

Throughout the study period we collected blood samples from 386 lizards. The slides that were prepared from 21 of these samples were not suitable for examination and were therefore excluded from the analyses (Appendix 7.6). Sixteen of the lizards (5.8%) were infected by the haemogregarine *H. mariae*. Other intraerythrocytic inclusions were also observed, but since they have not been identified to particular species yet, they are herein referred to as intraerythrocytic inclusion type 1 (IIT-1), intraerythrocytic inclusion type 2 (IIT-2) and intraerythrocytic inclusion type 3 (IIT-3). The prevalence of IIT-1, IIT-2 and IIT-3 were 60.2%, 38.6%, and 63.6% respectively, and the lizards were usually infected by more than one type of intraerythrocytic inclusion (Fig. 6.5). *Hemolivia mariae* had a more restricted distribution than IIT-1, IIT-2 and IIT-3, which were recorded throughout the study site (Fig. 6.6).

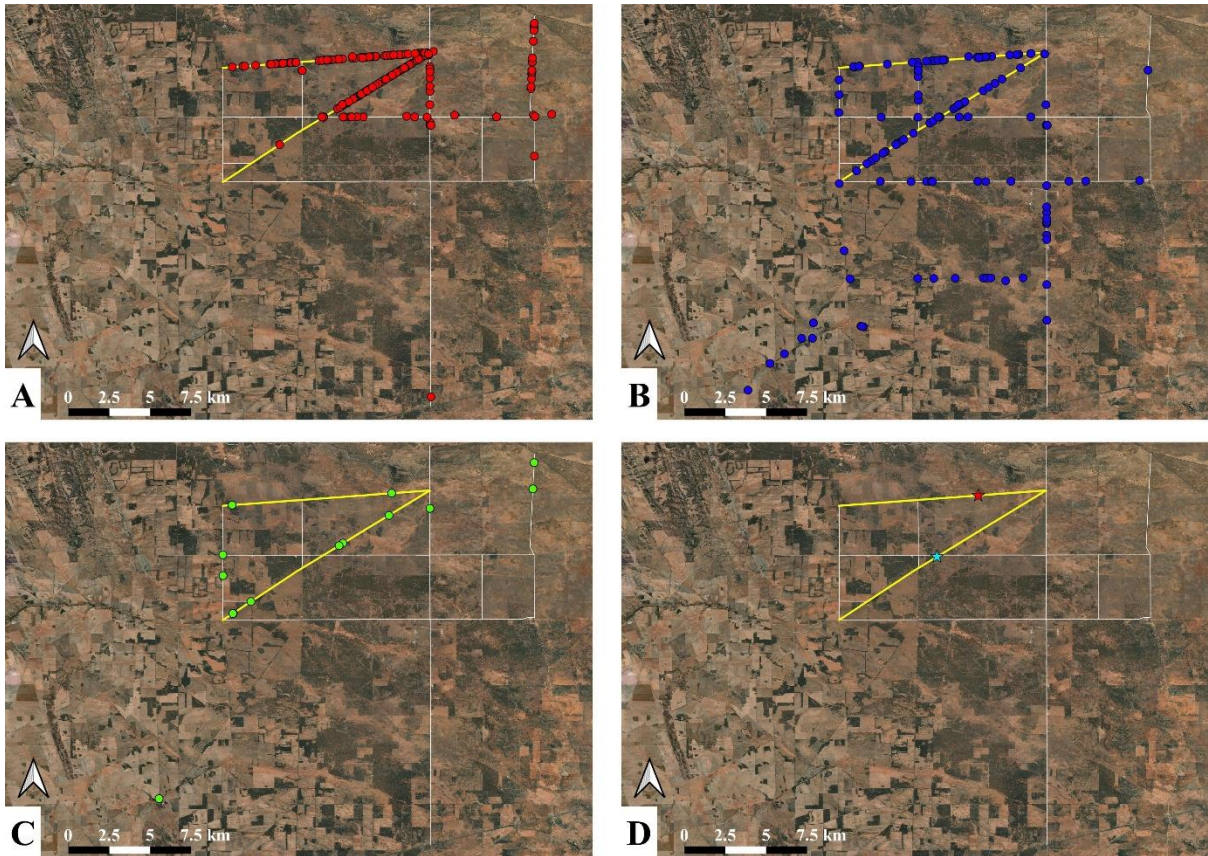


Figure 6.3. The distribution of where sleepy lizards (*Tiliqua rugosa*) that were infested by *Amblyomma limbatum* (A; red dots), *Bothriocroton hydrosauri* (B; dark blue dots), *Ornithodoros gurneyi* (C; light green dots), *Ophionyssus natricis* (D; red star) and an unidentified trombiculid (light blue star) were caught at the Bunday Bore study site during the period 2017 to 2019.

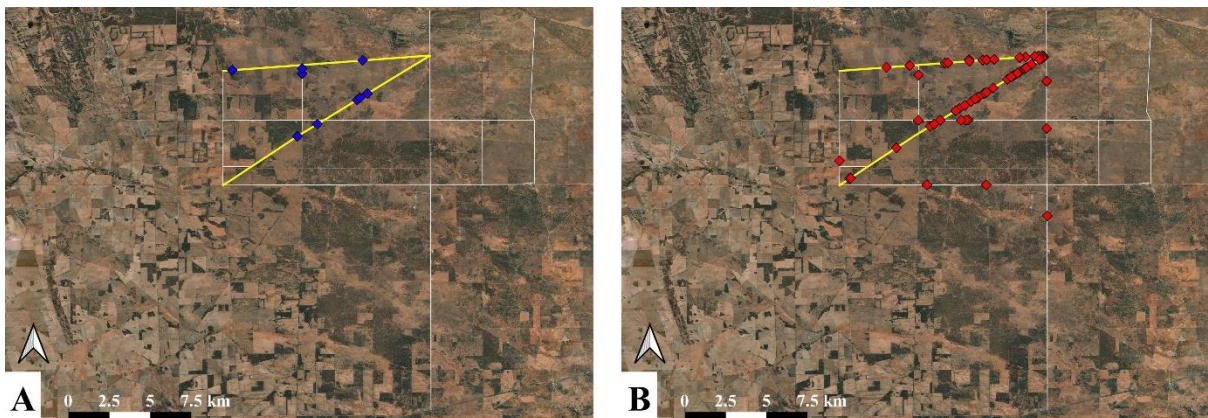


Figure 6.4. The distribution of where sleepy lizards (*Tiliqua rugosa*) that were infected by *Oochoristica trachysauri* (A; dark blue diamonds) and *Thelandros trachysauri* (B; red diamonds) were caught at the Bunday Bore study site during the period 2017 to 2018.

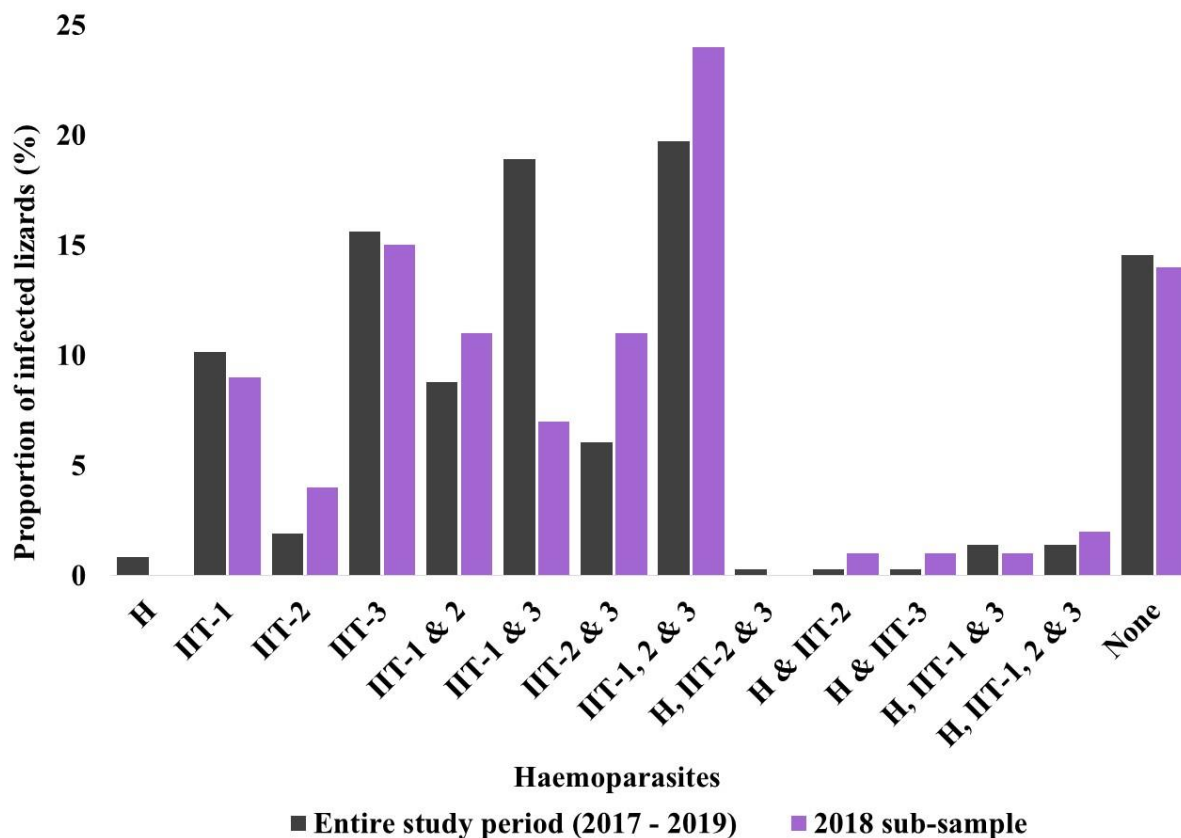


Figure 6.5. The proportion of lizards from the entire study period and the sub-sample of lizards collected in 2018 that were infected by *Hemolivia mariae* (H) intraerythrocytic inclusion type 1 (IIT-1), intraerythrocytic inclusion type 2 (IIT-2) and intraerythrocytic inclusion type 3 (IIT-3).

Parasite community comparisons: Only 145 of the 214 lizards that were caught in 2018 were used for the parasite community across the ecological gradient comparisons and the SVL's of this sub-sample of lizards ranged from 177 mm to 335 mm, with a mean of 302.5 ± 20.5 mm (\pm SD), which did not differ significantly ($U = 4159.5$, $P = .757$) from those of the other lizards caught in 2018. With the exception of trombiculid and snake mite that were not recorded in 2018, all the other recorded parasite species were also recorded from the sub-sample of lizards from 2018 (Table 6.3).

Overall, the tick infestation patterns of the sub-sample of the lizards that were caught in 2018 showed similar trends to that noted during the entire study period (Fig. 6.2). The majority (48.3%) of the lizards that were used for the parasite community comparisons were infested by *A. limbatum*, and 13.8% were infested by *B. hydrosauri*, while 13.8% of the lizards were co-infested by *A. limbatum* and *B. hydrosauri* (Fig. 6.2). We rarely recorded parasitism by *O. gurneyi* (Fig. 6.2). No other ectoparasites were observed in the sub-sample of lizards. The distribution of *A. limbatum* along the two transects was more continuous, in the north eastern

ends of the transects, than that of *B. hydrosauri*, which tended to be clustered at certain localities along the transects (Fig. 6.7). An inverse association was apparent between the prevalence of *A. limbatum* and *B. hydrosauri* across the different habitat categories (Fig. 6.8). *Ornithodoros gurneyi* was recorded too infrequently to provide a clear distribution pattern (Fig. 6.7).

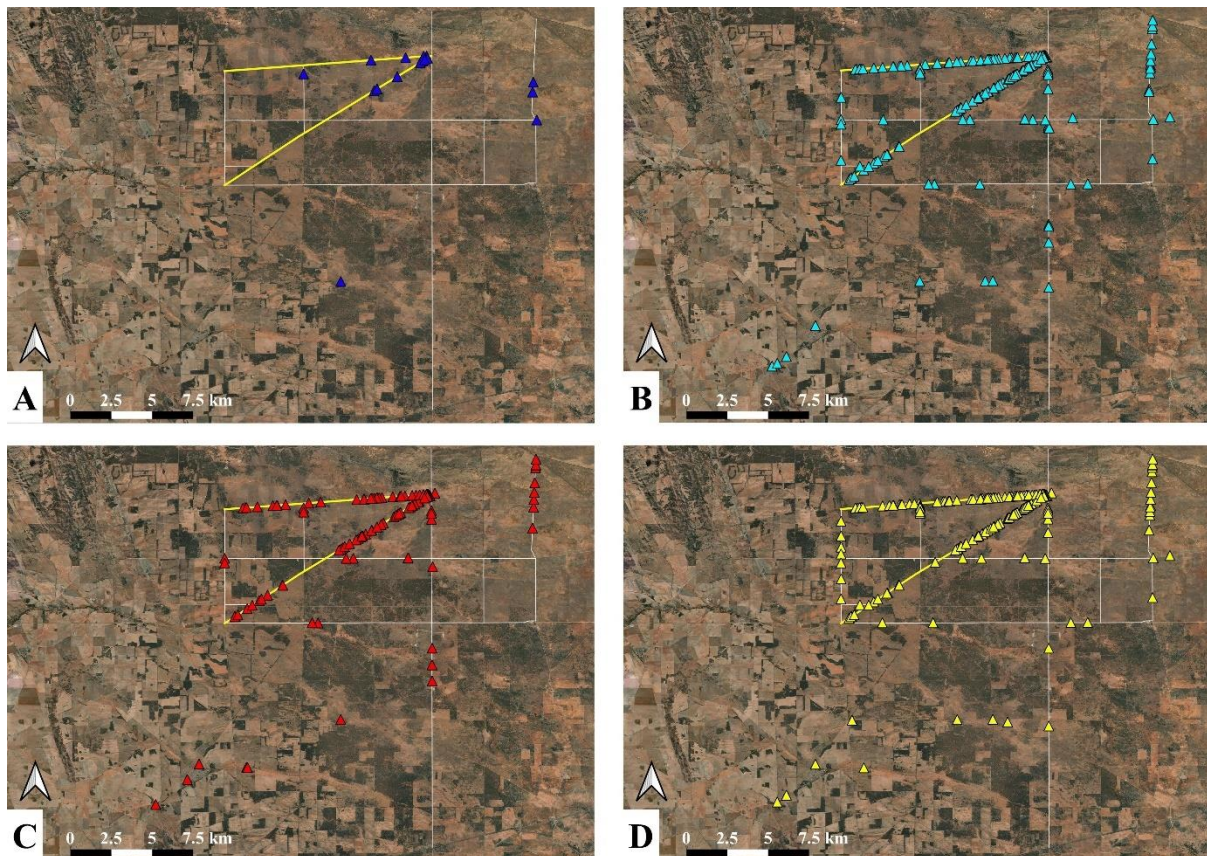











Figure 6.6. The distribution of where sleepy lizards (*Tiliqua rugosa*) that were infected by *Hemolivia mariae* (A; dark blue triangles), intraerythrocytic inclusion type 1 (B; light blue triangles), intraerythrocytic inclusion type 2 (C; red triangles) and intraerythrocytic inclusion type 3 (D; yellow triangles) were caught at the Bunday Bore study site during the period 2017 to 2019.

We collected faecal samples from 40 of the sub-sample of lizards that were caught in 2018, and the infection patterns of these 40 lizards were similar to that of all those caught during 2017 and 2018. Only one of the lizards (2.5%) of the sub-sample had no indications of a helminth infection. All the other lizards (97.5%) were infected by *T. trachysauri*, while 10% of the lizards were also infected by *O. trachysauri*. As was noted for the faecal samples collected from all the lizards caught in 2017 and 2018, *O. trachysauri* seemed to have a more restricted distribution than *T. trachysauri* (Fig. 6.9).

Table 6.3. A list of the parasites that were collected during the 2018 surveys along the two transects and the habitat types (1 to 5) the associated lizards were found in.

Parasite	1	2	3	4	5	Pictogram
ECTOPARASITE						
<i>Amblyomma limbatum</i>	X		X	X	X	
<i>Bothriocroton hydrosauri</i>	X	X	X	X	X	
<i>Ornithodoros gurneyi</i>	X			X	X	
GASTROINTESTINAL PARASITE						
<i>Oochoristica trachysauri</i>	X			X	X	
<i>Thelandros trachysauri</i>	X	X	X	X	X	
HAEMOPARASITE						
<i>Hemolivia mariae</i>			X	X	X	
Intraerythrocytic inclusion type 1 – distinct basophilic inclusions	X	X	X	X	X	
Intraerythrocytic inclusion type 2 – haemosporidian-like inclusions	X	X	X	X	X	
Intraerythrocytic inclusion type 3 – piroplasm-like inclusions	X	X	X	X	X	

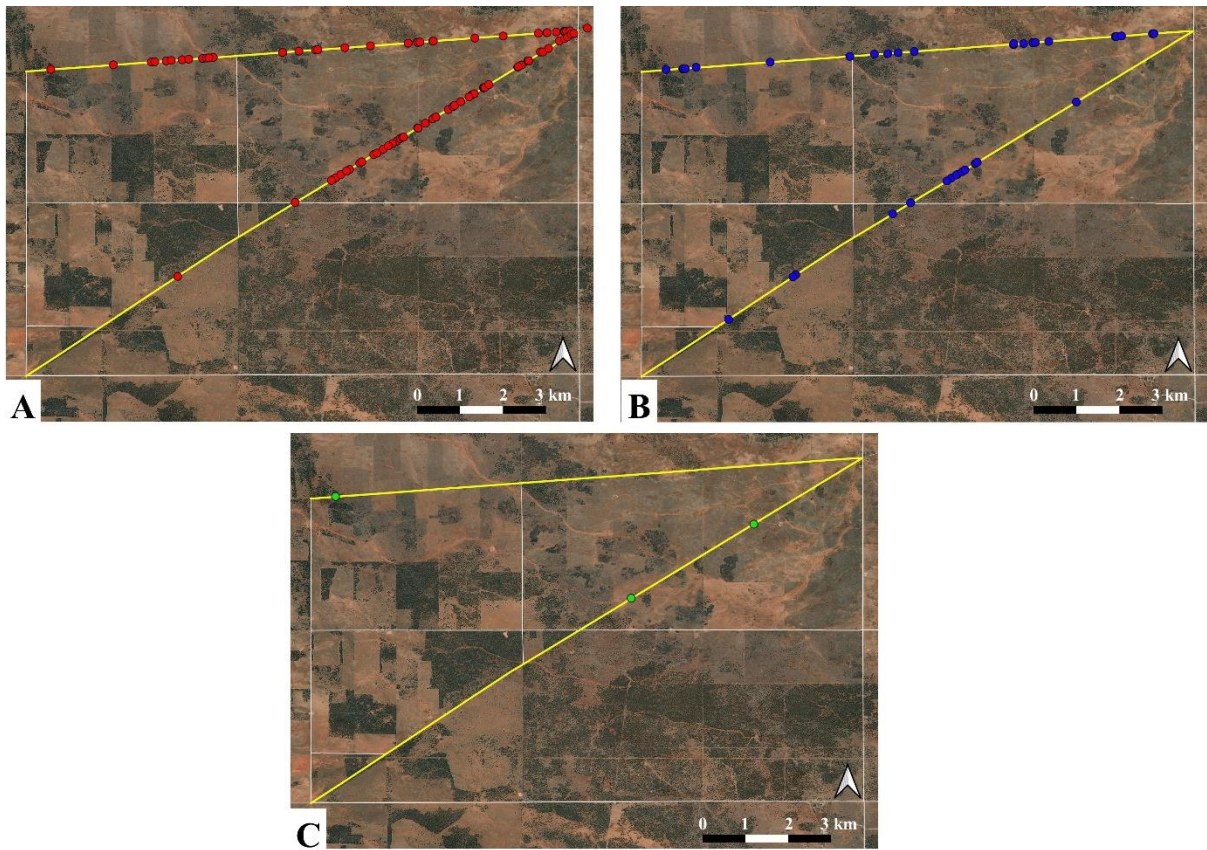


Figure 6.7. The distribution of where the 2018 sub-sample of sleepy lizards (*Tiliqua rugosa*) that were infested by *Amblyomma limbatum* (A; red dots), *Bothriocroton hydrosauri* (B; dark blue dots) and *Ornithodoros gurneyi* (C; light green dots) were caught along two transects at the Bunday Bore study site.

We obtained blood film slides suitable for examination from 100 of the sub-sample of lizards that were caught in 2018. In general, the infection patterns of these 100 lizards showed similar trends to those noted during the entire study period (Fig. 6.5). In 14% of the 100 lizards we did not note any haemoparasite infections (Fig. 6.5). *Hemolivia mariae* was observed in five of the examined lizards and all five were also infected by other types of intraerythrocytic inclusions. We recorded IIT-1, IIT-2 and IIT-3 in 54%, 53% and 53% of the examined lizards respectively, and in most instances more than one type of intraerythrocytic inclusion were observed in each blood film (Fig. 6.5). The distribution patterns of *H. mariae* and the three types of intraerythrocytic inclusions along the two transects were also similar to that of the entire study period, and *H. mariae* had a more restricted distribution than IIT-1, IIT-2 and IIT-3, which were recorded more widely distributed along the transects (Fig. 6.10).

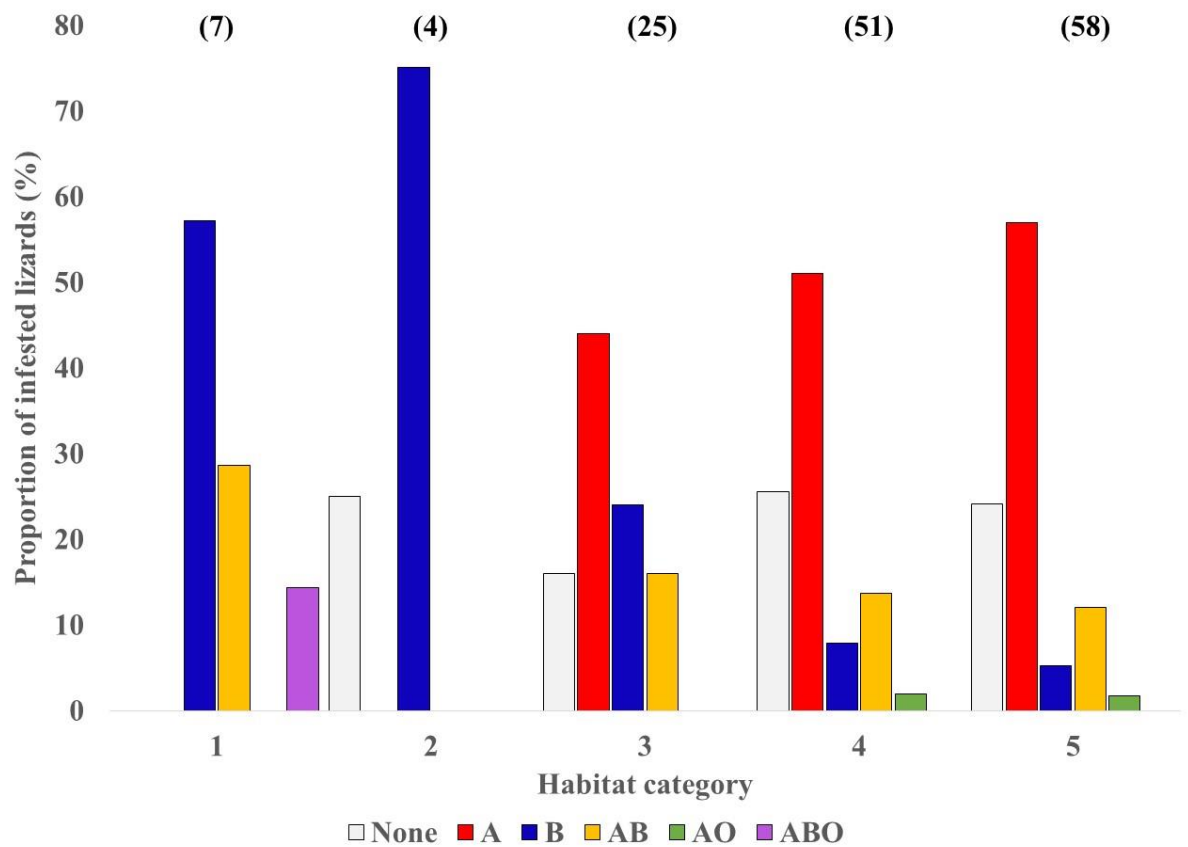


Figure 6.8. The number of (indicated in brackets) of the 2018 sub-sample of lizards that were caught in each habitat category, and the proportion of them that were infested by *Amblyomma limbatum* (A), *Bothriocroton hydrosauri* (B) and/or *Ornithodoros gurneyi* (O).

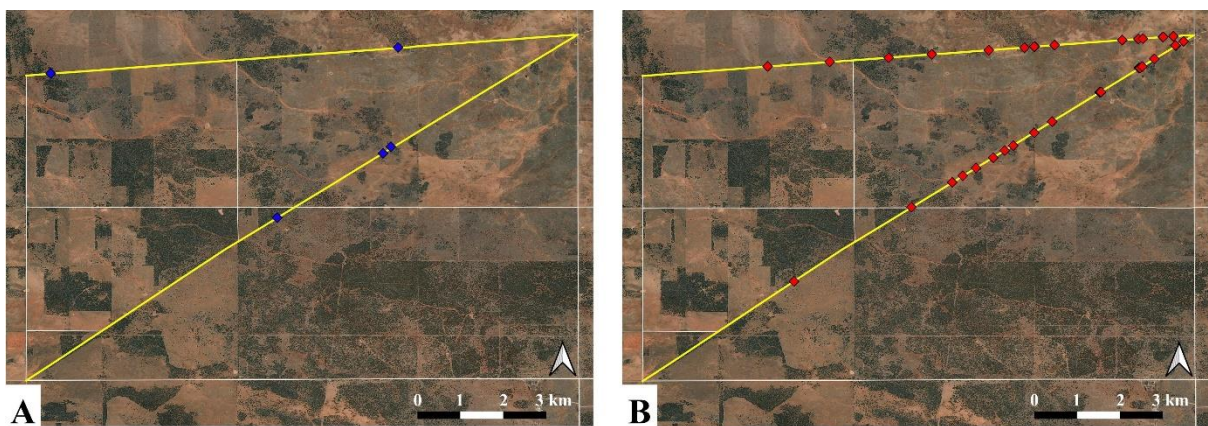


Figure 6.9. The distribution of where the 2018 sub-sample of sleepy lizards (*Tiliqua rugosa*) that were infected by *Ochoristica trachysauri* (A; dark blue diamonds) and *Thelandros trachysauri* (B; red diamonds) were caught along two transects at the Bunday Bore study site.

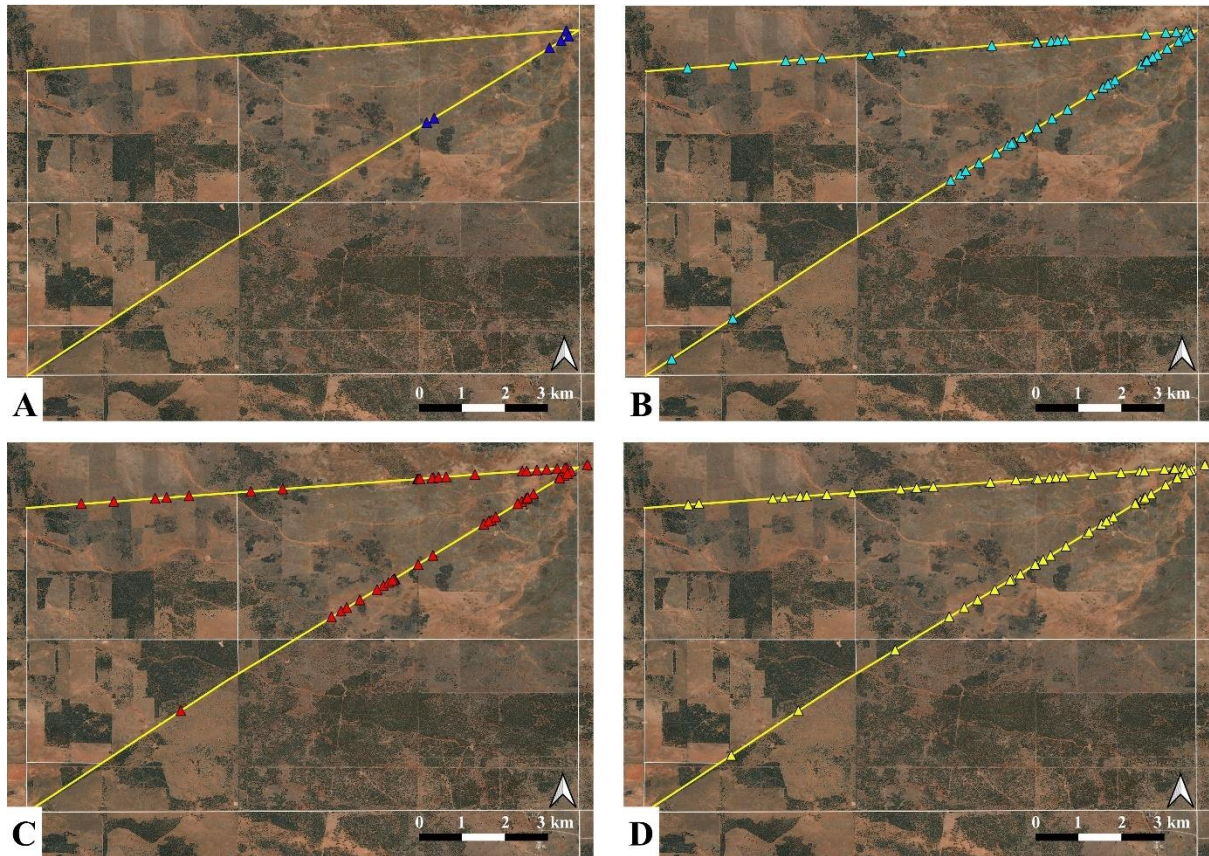


Figure 6.10. The distribution of where the 2018 sub-sample of sleepy lizards (*Tiliqua rugosa*) that were infected by *Hemolivia mariae* (A; dark blue triangles), intraerythrocytic inclusion type 1 (B; light blue triangles), intraerythrocytic inclusion type 2 (C; red triangles) and intraerythrocytic inclusion type 3 (D; yellow triangles) were caught along two transects at the Bunday Bore study site.

Only 30 of the sub-sample of lizards from 2018 had complete data on their ectoparasites, gastrointestinal helminths and haemoparasites, and were therefore could be used for the recorded parasite community assemblage comparisons. None of these lizards were caught in habitat categories 2 or 3. In the separate analyses of each transect, there were no significant differences in the recorded parasite community assemblage by transect or habitat types (PERMANOVA; $p > 0.05$). However, when the data from both transects were combined, a significant difference in the parasite community assemblage by habitat was detected (PERMANOVA, $df = 2$, res 136, Pseudo-F = 7.48, $p = 0.0001$). Based on SIMPER outputs the distinction between the parasite community assemblage of habitat types 1 and 4 was driven by the presence or absence of *B. hydrosauri*, *O. trachysauri*, IIT-1 and IIT-3. Also, habitat types 1 and 5 were driven by the presence or absence of *B. hydrosauri*, *O. trachysauri*, IIT-1, IIT-2 and IIT-3 (Fig. 6.11; Table 6.4). When all the ectoparasites, gastrointestinal helminths and haemoparasites that were recorded from all the sub-sample of lizards caught in 2018 were

analysed separately, there was no significant difference in the gastrointestinal helminths or haemoparasites assemblage based on transect or habitat types (PERMANOVA; $p > 0.05$). However, there was a significant difference in the recorded ectoparasite parasite community assemblage by habitat types within transects (PERMANOVA, $df = 4$, res 134, Pseudo-F = 2.71, $p = 0.0001$) and between habitats across transects (PERMANOVA, $df = 4$, res 134, Pseudo-F = 6.18, $p = 0.0001$). For the ectoparasites, there was not a significant difference in the ectoparasite assemblage by transects alone (PERMANOVA; $p > 0.05$). Based on SIMPER outputs, the distinction between the ectoparasite community assemblages of the different habitat types were driven by the presence or absence of *A. limbatum* and *B. hydrosauri* (Fig. 6.12; Table 6.5).

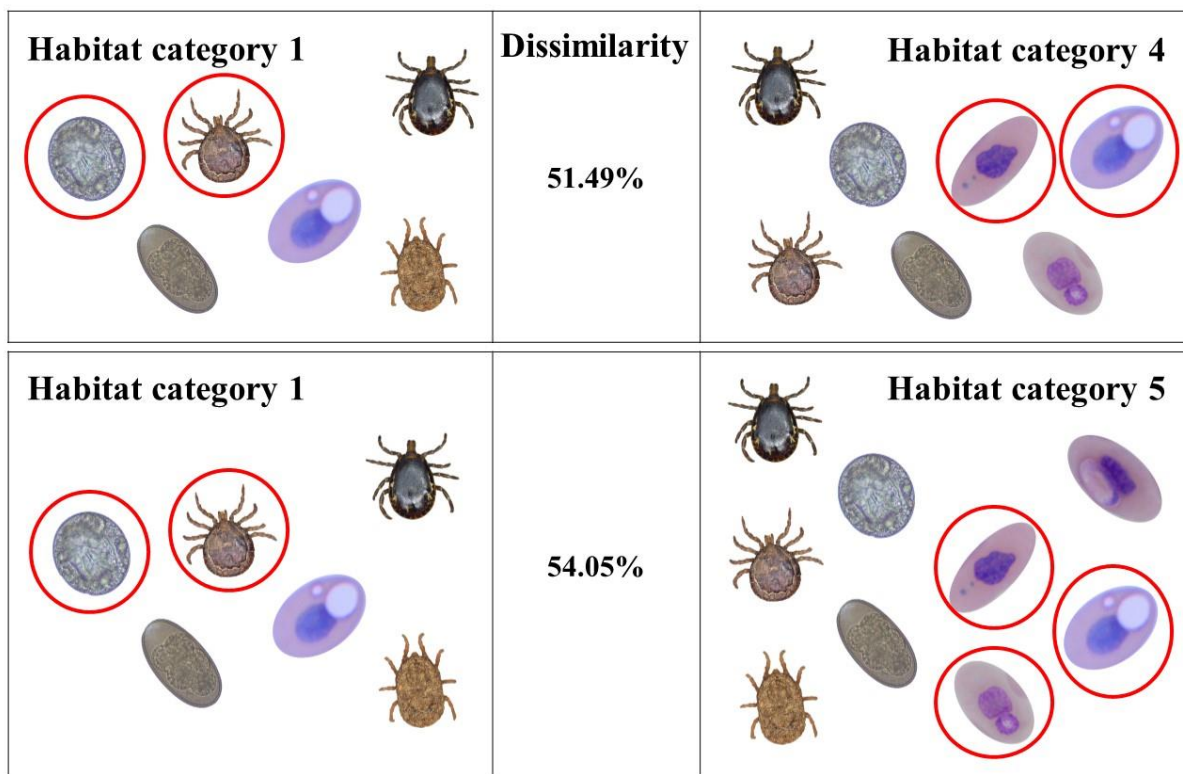


Figure 6.11. A Venn diagram of the parasite community assemblage based on the SIMPER analysis for the factor habitat category, indicating the percentage of dissimilarity between the habitat categories, and the parasites (see table 6.3 for pictogram key) recorded from each habitat category, with taxa contributing the most to the dissimilarity circled in red.

Discussion

This study reconfirmed the ticks *A. limbatum*, *B. hydrosauri* and *O. gurneyi*, the cestode *O. trachysauri*, the nematode *T. trachysauri* and the haemogregarine *H. mariae* as parasites of sleepy lizards at the study site (Bull and Smyth 1973, Sharrad 1979, Bull et al. 1981, Smallridge

1998, Gyawali 2011), but we also recorded described and undescribed parasites that were not reported in former studies. Thus, illustrating that the parasite community of sleepy lizards at the Bunday Bore study site is more complex than had previously been reported. Some parasite species were more prevalent than others and variations were noted in their distribution patterns.

Table 6.4. The SIMPER analysis for habitat category as the factor, indicating the similarity values (Avg. sim) of the recorded parasite community assemblage and the average abundance (Avg. abund.) and the similarity divided by the standard deviation (Sim./SD) of the indicator taxa (A cumulative cut off of 75% was applied).

	Avg. sim.	Species	Avg. abund.	Sim./SD
Category 1	66.27%	<i>Bothriocroton hydrosauri</i>	1.00	7.94
		<i>Thelandros trachysauri</i>	1.00	7.94
Category 4	59.11%	<i>Thelandros trachysauri</i>	0.92	1.95
		Intraerythrocytic inclusion type 3	0.75	1.06
		Intraerythrocytic inclusion type 1	0.58	0.66
Category 5	73.62%	<i>Thelandros trachysauri</i>	1.00	4.80
		Intraerythrocytic inclusion type 3	0.80	1.22
		Intraerythrocytic inclusion type 2	0.80	1.26

The two ticks, *A. limbatum* and *B. hydrosauri* were the only parasites recorded in this study that showed a clear association with the habitat types. This is not surprising since most tick species spend most of their life off their host in the environment where they are at risk of dehydration (Anderson and Magnarelli 2008). *Amblyomma limbatum* and *B. hydrosauri* are three-host ticks, which means that the larvae, nymphs and females detach after feeding for immature stages to moult to the next life stage or for females to lay their eggs (Roberts 1970, Bull et al. 1984). Detachment usually takes place under a shrub that the host uses as a refuge (Satrawaha and Bull 1981, Petney and Bull 1984). The eggs, engorged larvae and engorged females of *A. limbatum* are more resistant to desiccation than those of *B. hydrosauri* and as a result can inhabit dryer and warmer habitats than *B. hydrosauri* (Bull and Smyth 1973, Bull et al. 1981, Chilton and Bull 1993a, 1994). The larvae and nymphs of both tick species also utilise the leaf-litter under the shrubs differently, where *B. hydrosauri* requires plant species that create a thicker leaf-litter layer, and in contrast, those of *A. limbatum* can survive under shrubs that create a thinner leaf-litter layer (Petney and Bull 1984, Chilton and Bull 1993b). Therefore, the distribution of *B. hydrosauri* throughout the study site is determined by humidity and the

associated vegetation. Habitat categories 1 to 3 included plant communities made up of at least some species that are associated with moist conditions (Chapter 5). The overlap in distribution and co-infestation of lizards by *A. limbatum* and *B. hydrosauri* are in part due to biotic factors enabling the continued survival of both species in the same area. The home range of a sleepy lizard is ca. 2 to 5 ha and a tick can be dispersed up to 800 m in the tick's life by the movement of the lizard (Bull 1978, Bull and Baghurst 1998). This would explain the occurrence of *B. hydrosauri* and *A. limbatum* in the north-eastern and eastern parts of the study site which are generally dominated by chenopod shrublands. Plant communities associated with more moist conditions (e.g. earth dams and depressions) provide suitable habitat for *B. hydrosauri*. In the western and south-western parts of the study site in localities that have been degraded due to grazing and/or other agricultural activities induce the establishment of plant communities that have an affinity with habitat categories 3 to 5. Within these localities *B. hydrosauri* become less prevalent, while *A. limbatum* becomes more prevalent and can even be found in localities generally associated with mallee scrublands and the occurrence of *B. hydrosauri*. This inverse association has been shown not to be due to habitat suitability, because in survival experiments on both tick species in the mallee scrubland and chenopod shrubland, *A. limbatum* tended to have a higher survival rate than *B. hydrosauri* (Petney 1981, Bull 1987a). Also, the association has been shown not to be due to direct or indirect interspecific competition (Andrews and Petney 1981, Andrews et al. 1982) or predation (Chilton and Bull 1996). These results suggest that there is likely another biotic factor associated with *B. hydrosauri* that is preventing *A. limbatum* from being sympatric with *B. hydrosauri*. A study by Staines et al. (2020) identified *Rickettsia* spp. associated with *B. hydrosauri* as a possible controlling factor for the distribution of *A. limbatum*, however additional empirical studies are needed to confirm this. The results from this study also indicate that the boundary is possibly wider than previously reported, which may be due to the way the boundary has been defined, as in the studies by Bull and Burzacott (2001) used proportions of each species rather than presence/absence.

Ornithodoros gurneyi was recorded as an infrequent parasite of sleepy lizards at the study site. These ticks are primarily parasites of macropods and only opportunistically parasitise sleepy lizards (Browning 1962, Doube 1975, Sharrad and King 1981). A study into the distribution of this tick in kangaroo wallows at the Bunday Bore study site found that they occur throughout the study site (Norval et al. 2022), and our study found no apparent association between the habitat categories and the distribution of this tick. Still, it should be noted that as *O. gurneyi* is

a nidicolous parasite, it is reliant on macropods and their use of the habitat (Browning 1962, Doube 1975). Studies into the association between *O. gurneyi* parasitism on sleepy lizards and particular habitat types would require incorporating the use of the vegetation types by macropods as a factor.

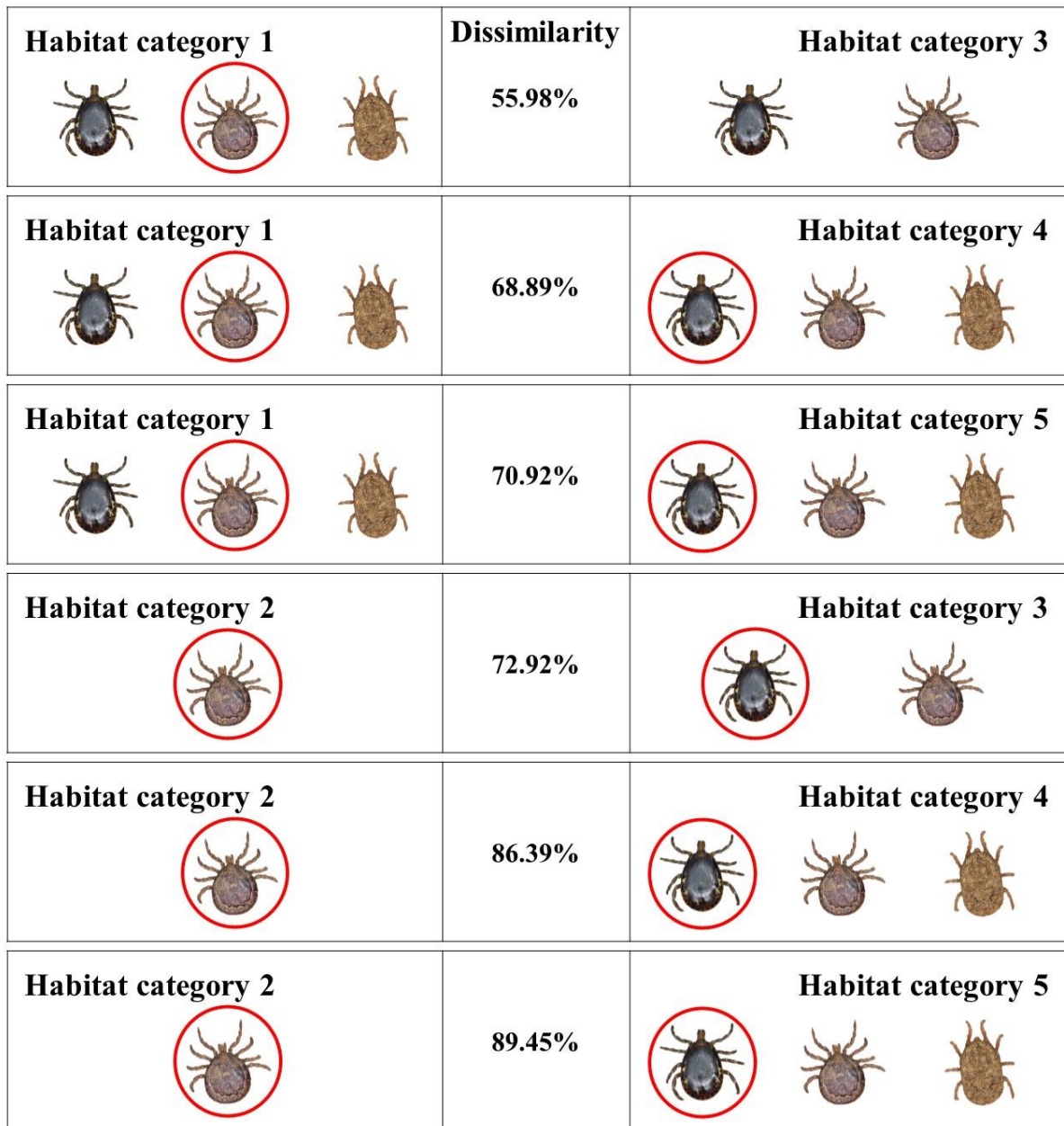


Figure 6.12. A Venn diagram of the tick community assemblage based on the SIMPER analysis for the factor habitat category, indicating the percentage of dissimilarity between the habitat categories, and the tick species (see table 6.3 for pictogram key) recorded from each habitat category, with taxa contributing the most to the dissimilarity circled in red.

Ochoristica trachysauri was infrequently recorded in this study, but similar results were

obtained in another study that involved the dissection of the lizards (Norval et al. 2021), so it is unlikely that the prevalence recorded in this study is an underestimate. Sleepy lizards are the definitive host of this parasite, and also requires an, as yet, undetermined arthropod intermediate host (Norval et al. 2019, Norval et al. 2021). This cestode has a wide distribution and has been recorded in parts of Queensland, New South Wales, Victoria, and South Australia (Johnston 1932). We found no association between *O. trachysauri* and the vegetation categories at the study site, which suggests that the intermediate host is also likely to be a habitat generalist. The low prevalence of this parasite in the sleepy lizards is likely due to the diet of the lizards. Sleepy lizards are omnivorous, but primarily feed on vegetation and only occasionally will prey on insects (Norval and Gardner 2020). The likelihood of a sleepy lizard ingesting an infected intermediate host is therefore relatively low.

Table 6.5. The SIMPER analysis for habitat category as the factor, indicating the similarity values (Avg. sim) of the recorded tick community assemblage and the average abundance (Avg. abun) and the similarity divided by the standard deviation (Sim./SD) of the indicator taxa (A cumulative cut off of 75% was applied).

	Avg. sim.	Species	Avg. abun.	Sim./SD
Category 1	75.87%	<i>Bothriocroton hydrosauri</i>	1.00	3.23
Category 2	50.00%	<i>Bothriocroton hydrosauri</i>	0.75	0.91
Category 3	41.27%	<i>Amblyomma limbatum</i>	0.60	0.72
Category 4	42.84%	<i>Amblyomma limbatum</i>	0.67	0.90
Category 5	47.73	<i>Amblyomma limbatum</i>	0.71	1.01

Almost all the lizards examined in this study were infected by the oxyurid *T. trachysauri*. Similar results were found from studies involving sleepy lizards at the Bunday Bore study site as well as elsewhere (Angel and Mawson 1968, Jones 1992, Gyawali et al. 2013, Norval et al. 2021). In this study we found no association between *T. trachysauri* and vegetation types. This is very likely because the eggs of *T. trachysauri* are, to some extent, resistant to desiccation (Hallas and Bull 2006). As these worms are haplodiploid (Adamson 1990) only a few fertilised eggs would likely be required to infect a lizard. Infection is via the faecal-oral route (Telford 1971, Frank 1981) so infection most likely takes place when lizards use tongue flicks to investigate chemical cues from their own scats or those of conspecifics.

Hemolivia mariae was infrequently recorded in this study, and the prevalence was lower than that recorded by Smallridge and Bull (2000) at the same study site, most likely due to differences in where the samples were collected. The tick *A. limbatum* is the primary definitive host of this haemogregarine, although it can also infect *B. hydrosauri*, and the lizards, which are the intermediate host, can become infected if they ingest an infected tick (Smallridge and Paperna 1997, Smallridge and Bull 1999). The samples for the study by Smallridge and Bull (2000) were collected in the part of the study site where *A. limbatum* is the dominant tick and as a result a higher prevalence can be expected. As was stated for the cestode *O. trachysauri*, a low prevalence of this parasite in the lizards is not out of the ordinary considering the main diet of the lizards. Despite *A. limbatum* being the primary definitive host of *H. mariae* we found no association between this haemogregarine and the vegetation types, most likely as a result of the small sample size. As these parasites have a higher prevalence in the ticks (Smallridge and Bull 2001), future studies into the association between this parasite and vegetation types should also incorporate the definitive hosts.

Snake mite and chiggers were not recorded as parasites of the sub-sample of lizards that were collected in 2018 and therefore were not included in the association with vegetation types analyses. It should still be noted that *O. natricis* and many chigger species are reliant on humid off-host microhabitats to avoid desiccation (Camin 1953, Domrow 1967) and thus likely have a similar vegetation type association as *B. hydrosauri*. However, this needs to be verified through empirical studies.

All three types of unidentified intraerythrocytic inclusions had no apparent association with the habitat types. These organisms and any possible associated intermediate and/or definitive hosts would have to be identified before examining their association with habitat types and distribution.

In summary, in this study we surveyed the ecto- and endoparasites of sleepy lizards that were sampled across an ecological gradient at a study site in the Mid North region of South Australia, and tested for an association between the parasites and the habitats that the lizards were caught in. Our study reconfirmed previously recorded ecto- and endoparasite species as parasites of sleepy lizards in this locality, but also indicated that the parasite community is more complex as parasite species that were not previously reported were also found to parasitise the lizards.

An association between more arid habitats and *A. limbatum*, and less arid and *B. hydrosauri* was found. No association was found between the habitat categories and any of the other parasites. This study not only reconfirms the parapatric boundary between *A. limbatum*, and *B. hydrosauri* but also indicated that habitat modifications can alter their distribution and that the boundary may be wider than previously reported.

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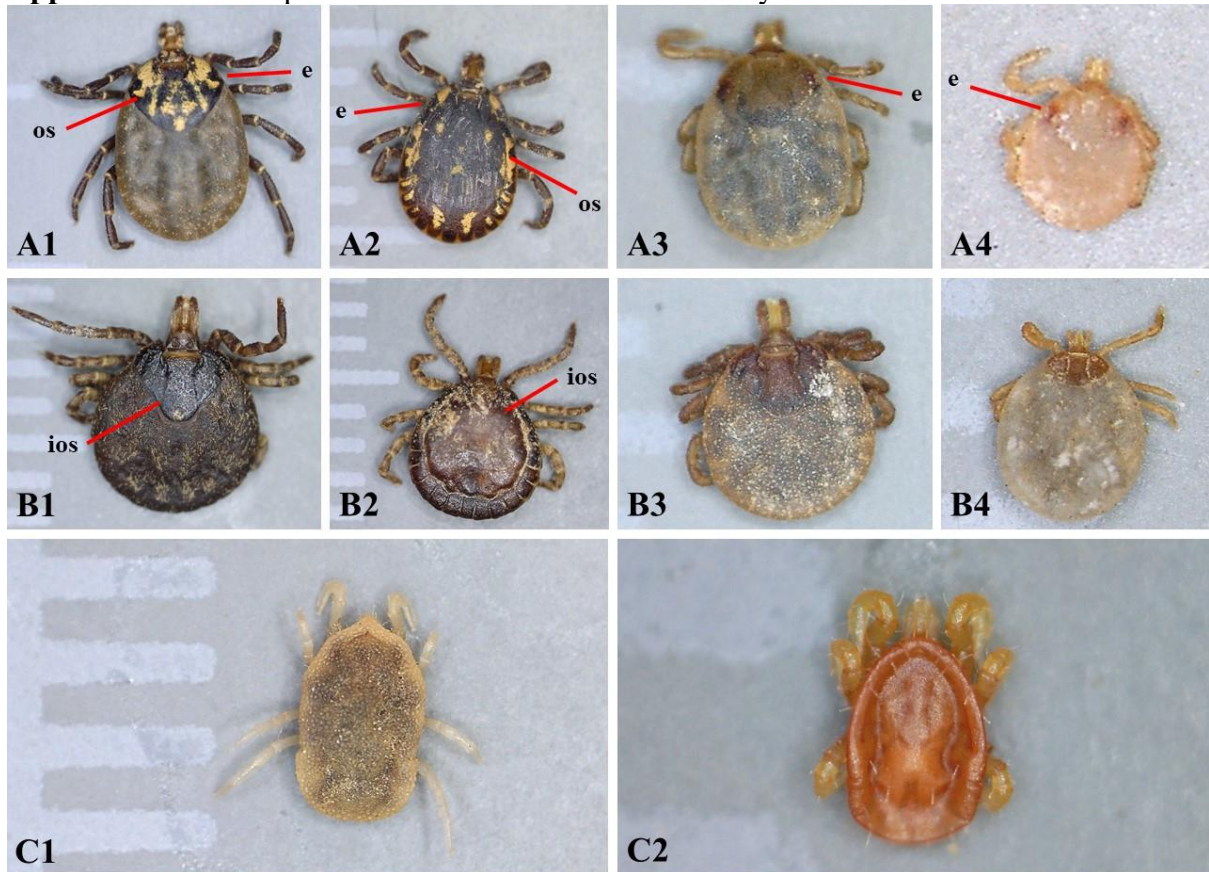
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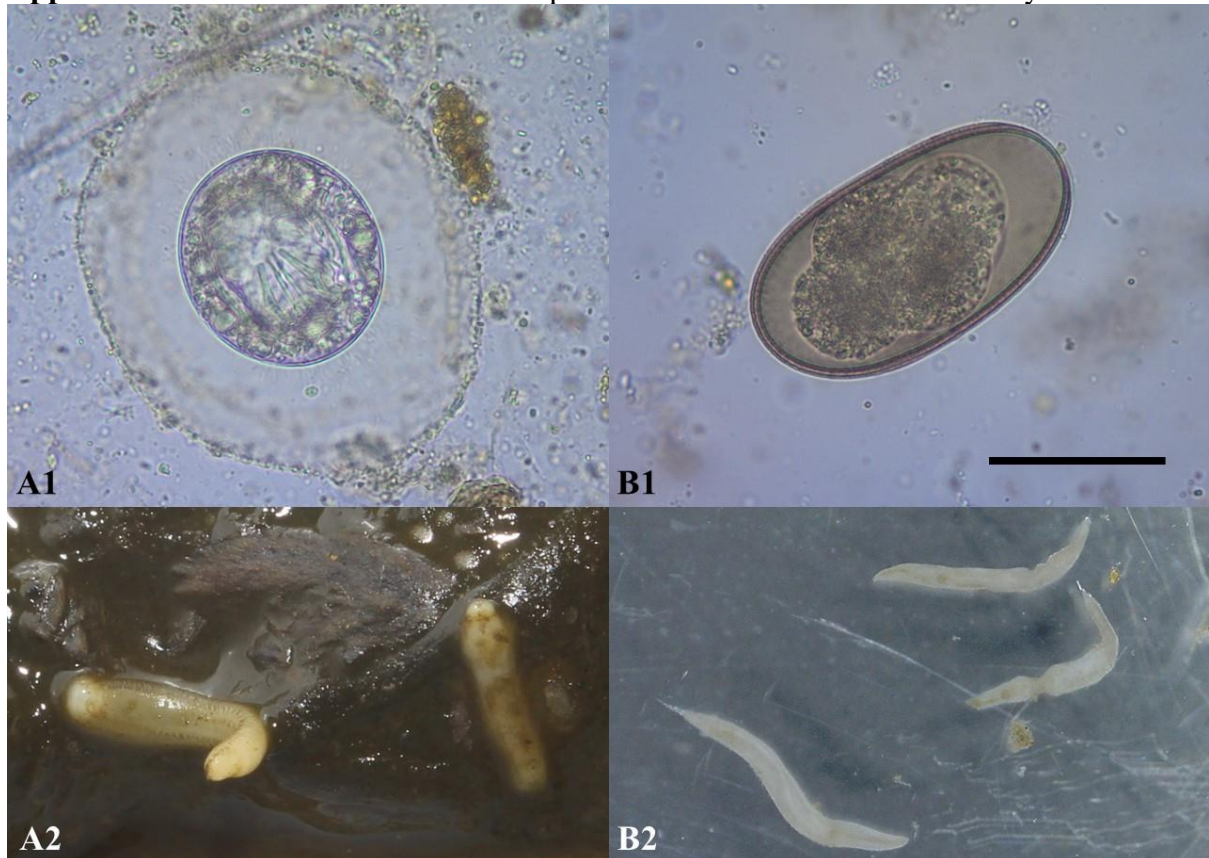
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Appendix 6.1. Tick species that were recorded in this study



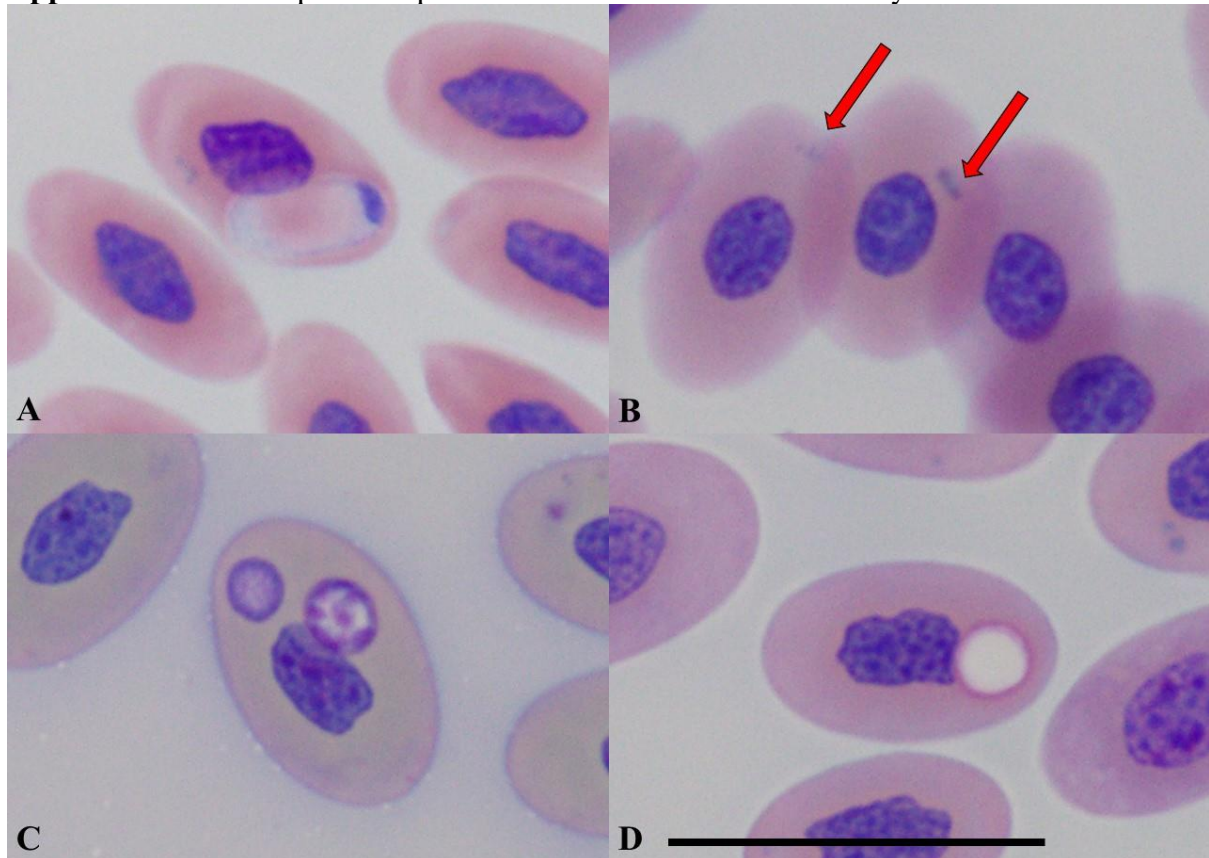
The three tick species record in this study could be distinguished from one another on the following characteristics. The female (A1) and male (A2) of *Amblyomma limbatum* has an ornate scutum (os), whereas that of the female (B1) and male (B2) of *Bothriocroton hydrosauri* is inornate (ios). Eyes (e) are present in the female (A1), male (A2), nymph (A3) and larva (A4) of *A. limbatum*, whereas eyes are not present in the female (B1), male (B2), nymph (B3) or larva (B4) of *B. hydrosauri*. The nymph (C1) and larva (C2) of the soft tick *Ornithodoros gurneyi* can be distinguished from the other ticks based on the absence of a scutum; instead the body is covered by a leathery cuticle.

Appendix 6.2. Gastrointestinal helminth species that were recorded in this study



Infection by gastrointestinal helminths were determined by the presence of eggs (A1; magnified $\times 400$) and/or the proglottids (A2) of the cestode *Oochoristica trachysauri* and/or the eggs (B1; magnified $\times 400$) and/or adults (B2) of the nematode *Thelandros trachysauri* in the faecal samples collected from the sleepy lizards (*Tiliqua rugosa*) that were captured for this study (scale bar = 50 μm).

Appendix 6.3. Haemoparasite species that were recorded in this study



Four kinds of haemoparasites and inclusions were observed in the blood smears prepared from the sleepy lizards (*Tiliqua rugosa*) that were caught for this study; *Hemolivia mariae* (A), distinct basophilic inclusions (B), haemosporidian-like inclusions (C), and piroplasm-like inclusions (D) (scale bar = 20 μm).

Chapter 7

Thesis Discussion

The work in this thesis examined the composition and distribution of the parasite community of sleepy lizards (*Tiliqua rugosa*) that inhabit an area which spans an ecological gradient as part of the ongoing long-term research into the ecology of these lizards and their parasites. Parasites make up a very large part of the world's biodiversity (Windsor 1998) and are important components of ecosystems (Valkiūnas 2001), but ecologists have only in recent decades started to appreciate the roles parasites play in ecosystems (Windsor 1995, Gómez and Nichols 2013). By having a negative impact on the fitness of the host, many parasites help to regulate host populations (Scott and Dobson 1989, Tompkins and Begon 1999). They also influencing energy flow , and mediate interspecific competition (Schall 1992, Valkiūnas 2001). Parasites can also protect the host against more harmful parasites (Abbate et al. 2018, Bhattacharjee et al. 2019, Shen et al. 2019) and can even promote speciation (Clayton 1991, Karvonen and Seehausen 2012). But, since in the past parasites have been neglected in ecological research there are gaps in our understanding of their natural history, and even fundamental information, such as the host species they infect or infest and/or their geographic distribution is often poorly understood.

In this thesis, I have addressed the knowledge gaps relating to the parasite community of a sleepy lizard population that have been used for long-term monitoring and ecological studies for the past 40 years. My studies focussed on the ectoparasites, gastrointestinal helminths and haemoparasites, with a focus on their distribution within the study site and the possible drivers that may determine their presence or absence. This work is essential because it contributes to our understanding of the distribution of the various parasites and their natural history and forms a basis for further work exploring the interactions among parasites within this in-depth study system. My study also contributes to research pertaining to the sleepy lizards at the study site as they are used as the model species for studies into sociality and monogamy in lizards, as well as research into parasite transmission in social networks. For my research I caught sleepy lizards throughout the study site to record the ectoparasites that infest them, and to collect faecal samples and blood smears to determine which gastrointestinal helminths and haemoparasites infect the lizards. I also dissected dead specimens and searched for some parasite species in the environment. To test for an association between the various recorded

parasite species and different habitats in the study site, I classified habitats along two representative transects based on vegetation surveys that I conducted. Whilst various parasites have been very well studied in this system, there has been limited focus on examining the relationships among them and between the vegetation at the site and the relationship with the parasite distributions. My work aimed to fill this gap. How this work addresses the specific research aims within this broad objective of my doctoral studies are discussed below.

Key research findings

Gastrointestinal helminth community

Gastrointestinal parasites can be problematic to identify to the species level based on their eggs, and if a host is infected by immature stages of some helminth species, such infections are easily overlooked in faecal flotation examinations (Kassa et al. 2016). One of the primary aims of the project was therefore to dissect sleepy lizards that were found dead on roads to verify which helminth species infect the lizards at the study site by. By also dissecting other roadkill lizards and snakes, it was possible to also gain a broader understanding of the gastrointestinal helminth community of reptiles in the area. It was found that all adult sleepy lizards are infected by the oxyurid *Thelandros trachysauri* and that ca. 10% of the lizards were also infected by the cestode *Oochoristica trachysauri* (Norval et al. 2021a). Both these helminth species have previously been recorded at the study site (Gyawali et al. 2013), but this project established a baseline for comparisons with the results from faecal examinations. It also provided additional support for the conclusion that *O. trachysauri* is likely host specific. None of the other helminth species that were collected from other reptile species, some of which were new host and/or distribution records, were found in the sleepy lizards (Norval et al. 2021a), indicating that although these reptile species are sympatric in this area, differences in their diets and behaviour likely drive variations in their gastrointestinal helminth communities.

An exotic invasive parasite of sleepy lizards

This project aimed to record as many parasite species as possible to provide an improved overview of the parasite components of this system. As a result a species that would not have been considered a potential parasite of sleepy lizards in this area was also recorded. The snake mite (*Ophionyssus natricis*) is an exotic invasive parasite of snakes and lizards in Australia (Walter and Shaw 2003). This species is sensitive to desiccation so it is generally assumed to be only associated with reptiles in captivity and free-living snakes and lizards in urban areas

(Camin 1953, Domrow 1988). This project recorded sleepy lizards as a new host record of this parasitic mite. The associated review of the literature and museum specimens also demonstrated that *O. natricis* occurs in the wild and that it can occur in arid areas under some conditions (Norval et al. 2020, Norval et al. 2021b). These findings highlight the need for additional studies to determine the distribution of this parasite in Australia and what other Australian indigenous species host free living populations of snake mites.

Confirming that sleepy lizards are competent hosts of the kangaroo soft tick

Ticks are in general host specific to some extent, especially as adults (Arthur 1962, Oliver 1989, McCoy et al. 2013). When ticks attach to atypical hosts the tick may not feed effectively and/or fail to moult (Hoogstraal and Aeschlimann 1982). The kangaroo soft tick (*Ornithodoros gurneyi*) is generally assumed to be mainly a parasite of macropods in arid areas (Browning 1962, Doube 1972). However, these ticks have occasionally been recorded to parasitise sleepy lizards (Sharrad 1979, Sharrad and King 1981), but it is not known if these lizards are competent hosts of this tick. This project therefore aimed to determine the prevalence of these ticks as parasites of sleepy lizards and to ascertain whether the lizards are competent hosts of this tick. The distribution of the kangaroo soft tick in the study site was determined by surveying kangaroo wallows. The competence of sleepy lizards as a host for kangaroo soft ticks was examined by keeping larvae and nymphs of this tick that had fed on sleepy lizards, in captivity to determine if they would survive and moult. I found that that the kangaroo soft tick occurs throughout the study site, and that they primarily parasitise sleepy lizards in late spring and summer. Larvae and nymphs that had fed on sleepy lizard blood had a high survival rate and usually moulted, indicating that sleepy lizards are competent hosts for this tick. Additional studies are needed to explore the likelihood of more frequent *O. gurneyi* parasitism on *T. rugosa* by adults and later-stage nymphs that may feed and detach while the lizards are resting and are therefore overlooked.

Descriptions of plant communities

Vegetation descriptions are often incorporated with zoological studies as the plants are a food source for some animals, and at the same time create the physical structure of the habitat and microclimates the animals require (van As et al. 2012). This also applies to some parasites, and as a result vegetation descriptions and classifications have been used to examine the distribution of parasites such as ticks. Some general description of the vegetation the study site

have been made (Sharrad 1979, Bull et al. 1981), but the plant communities at this locality have not been described. An important aim of this project was therefore to investigate the plant communities of the Bunday Bore study site. As sleepy lizards often feed at roadsides and will use the associated vegetation as shelters, sample plots were primarily placed at roadsides along two transects where the vegetation is representative of that of the study site. Two major plant communities were identified consisting of nine sub-communities with a mosaic distribution pattern. Species that can indicate certain environmental factors, primarily humidity and disturbance associated with human activities, were identified. Additional studies and long-term monitoring of the vegetation of the study site are needed, especially after good rainfall, to be able to determine a comprehensive understanding of the plant communities of this area.

The distribution of parasites and their association with different habitats

As stated earlier, parasites play vital roles in ecosystems but are often overlooked in ecological studies (Thomas et al. 1999, Horwitz and Wilcox 2005, Gómez and Nichols 2013), and as a result even fundamental information, such as their distribution is often lacking or incomplete. In this project sleepy lizards were caught over a three-year period throughout the study site, and were examined for ectoparasites, gastrointestinal helminth parasites and haemoparasites. The study revealed that the parasite community of the lizards at this locality is more complex than was previously reported. The recorded presence-absence data were used to map the distribution of the recorded parasites. The plant community descriptions from the study site (Chapter 5) were used to arrange the habitats in the study site into five categories. Next, associations were sought between the distribution of the recorded parasite species and the habitat categories. The study reconfirmed an association between arid habitats and the tick *Amblyomma limbatum*, and mesic habitats and the tick *Bothriocroton hydrosauri*, but the distribution of these ticks were not primarily concentrated on either side of the 250-mm rainfall isohyet as had been previously stated (Smyth 1973, Sharrad 1979, Petney 1981). Instead, it was found that habitat modifications can alter the distribution of these ticks and that they can occur in areas that generally would be considered beyond their usual distribution. No association was found between any of the other recorded parasites and the different habitats.

Research implications

The long-term ecological studies of sleepy lizards and the ticks that infest them at the study site north of Mt. Mary in the Mid North region of South Australia was initiated by the late Prof.

Mike Bull to investigate the parapatric distribution of the ticks *A. limbatum* and *B. hydrosauri* (Smyth 1973, Godfrey and Gardner 2017). It was found that the parapatric boundary between the ticks broadly coincided with the 250-mm rainfall isohyet and the associated vegetational ecotone, and that the one species replaced the other within a zone of ca. 2 km (Smyth 1973, Sharrad 1979, Bull et al. 1981, Petney 1981). Subsequent research showed that *B. hydrosauri* is sensitive to desiccation and that its distribution is dictated by humidity and the associated vegetation (Petney and Bull 1984, Chilton and Bull 1993a, b, 1994). This however does not explain the distribution of *A. limbatum*, which Petney (1981) experimentally demonstrated can survive in different habitats on either side of the boundary. Direct or indirect interspecific competition has been ruled out as the limiting factor for the distribution of *A. limbatum* (Andrews and Petney 1981, Andrews et al. 1982). Chilton and Bull (1996) also demonstrated that ant predation does not play a role, and as the haemogregarine blood parasite, *Hemolivia mariae* primarily infects *A. limbatum* (Smallridge and Bull 1999) and no other blood parasites that are exclusively associated with *B. hydrosauri* were found, Bull and Possingham (1995) proposed a model based on increased environmental heterogeneity that reduces the opportunity for the coexistence of competing species as a possible explanation for the parapatric distribution of these two reptile tick species.

The work in this thesis (Chapter 6) provides support for earlier findings that the distribution of *B. hydrosauri* is limited by its sensitivity to desiccation. However, it also found that the zone in which one species replaces the other can be wider than previously reported. This is likely because habitat alterations create habitats that favour each species of tick, and if lizards that are infested by a particular tick species disperse to or through these habitats, these ticks can establish colonies in these areas. It was also found that in areas where *B. hydrosauri* was more prevalent, *A. limbatum* tended to be less prevalent, even if the surrounding habitat was the type that normally would favour *A. limbatum*. This suggests that there is likely a biotic factor associated with *B. hydrosauri* that is preventing *A. limbatum* from being sympatric with *B. hydrosauri*. The *B. hydrosauri* associated *Rickettsia* sp. that is present in the study site has been proposed as a possible biotic factor limiting the distribution of *A. limbatum* (Staines et al. 2020), but this needs to be verified.

Invasive species is one of the main causes of biodiversity loss (Ejeta and Bekele 2017, Kearney et al. 2019). The snake mite is considered an introduced species in Australia and even though

some specimens have been collected from free-living snakes and skinks in parts of coastal eastern and southern Australia, it is generally assumed it does not occur in the wild (Domrow 1988, Walter and Shaw 2003, Wildlife Health Australia 2018). During the course of the data gathering for this thesis, snake mite were collected from a sleepy lizard at the Bunday Bore study site (Norval et al. 2020) and in a subsequent follow-up investigation additional records were found in this area (Norval et al. 2021b). These observations indicate that snake mite are present in rural parts of Australia and even though this species is sensitive to desiccation, under some conditions they can occur in xeric habitats. This highlights the need for surveys to determine the extent of the distribution of this parasite in Australia and studies to determine its impact on local wildlife.

The host specificity of different life stages of particular tick species varies, with some parasitising only specific hosts while others will feed on a variety of host species (Arthur 1962, Oliver 1989, McCoy et al. 2013). Our understanding of this aspect of tick ecology is incomplete, as most studies tend to focus on species that are of economic and public health concern. The kangaroo soft tick is endemic to xeric parts of Australia, and is therefore rarely encountered and as a result its natural history is not well-documented (Doube 1972, Barker and Walker 2014). Based on its presence in kangaroo wallows it is inferred that its primary hosts of the kangaroo soft tick are large macropods (Doube 1972, Barker and Walker 2014). In addition to reports of it to opportunistically biting humans and domestic animals such as cattle (*Bos taurus*), dogs (*Canis familiaris*), horses (*Equus caballus*) and pigs (*Sus scrofa*) (Henry 1938, Wilkinson 1958, Roberts 1970, Doube 1975, Norval et al. 2021c), immature life stages have also been collected from a greater bilby (*Macrotis lagotis*), short-beaked echidna (*Tachyglossus aculeatus*), a bearded dragon (*Pogona barbata*, reported as *Amphibolurus barbatus*) and sleepy lizards (Doube 1975, Sharrad and King 1981, Barker and Walker 2014). As feeding on atypical hosts could negatively impact survival (Hoogstraal and Aeschlimann 1982) it was not known if kangaroo soft ticks can use blood meals from these atypical hosts to survive and/or moult. My research into parasitism by kangaroo soft ticks on sleepy lizards (Chapter 4) indicated that larvae and nymphs can use bloodmeals from sleepy lizards to moult. This suggest that at least the immature stages are likely host generalists. Additional studies are needed to determine if this also applies to the adults. These atypical hosts could be a means of dispersal for these ticks and may help them persist in a xeric habitat during times when the preferred hosts do not make use of the kangaroo wallows that the ticks inhabit. The range of

natural hosts of the kangaroo soft tick and the roles that atypical hosts play in the life cycle of this tick requires additional empirical studies.

Vegetation types and/or habits have been shown to be suitable predictors for the distribution of particular tick species (Lindström and Jaenson 2003, Ledger et al. 2019, Talbot et al. 2019, Kwak et al. 2021). Similarly, herein I found that there is an association between the distribution of the tick *B. hydrosauri* and certain habitats (Chapter 6). The analyses also found an association between the distribution of *A. limbatum* and habitats that are unsuitable for *B. hydrosauri*. This suggests that an abiotic factor associated with *B. hydrosauri* is preventing these ticks from being sympatric as has been well documented (Bull 1987, Bull and Burzacott 2001). I also found that habitat modification can influence the distribution of the ticks. Climate change is predicted to alter the distribution of some tick species (Gray et al. 2009, Ogden et al. 2020, Gilbert 2021), and very likely also holds true for *A. limbatum* and *B. hydrosauri*. As habitats become more arid, the distribution range of *B. hydrosauri* can be expected to shrink, while that of *A. limbatum* would likely then expand. The distribution of these ticks should therefore be monitored and studies should be undertaken to identify possible pathogens that could be emerging wildlife diseases if these tick species spread to areas where they did not formerly occur.

Future research directions

Natural history is an integral component of ecology, and an in-depth understanding of the ecology of a species is essential for successful conservation outcomes and effective management plans (Greene and Losos 1988, Greene 1994, Bury 2006, Tewksbury et al. 2014). Unfortunately, parasites have often been overlooked in ecological studies and are still usually not incorporated into conservation and management plans (Gómez and Nichols 2013). To address this shortcoming, it is essential to develop a better understanding of the parasite community of a particular species, as well as the natural history and ecology of the various parasite species.

A better understanding of the ecology of the sleepy lizard will require additional research on the natural history of the parasites. For instance, chiggers have not previously been recorded as parasites of sleepy lizards (Norval et al. 2019) and it is possible that the record reported herein (Chapter 6) was an instance of the chigger attaching to an atypical host. Still, some

chiggers in Australia have been recorded to parasitise reptiles (Domrow and Lester 1985), and due to their small size, chiggers can easily be overlooked, which means that they may be parasites of sleepy lizards in certain areas. Therefore, in addition to identifying the chigger species, surveys should be done to determine the prevalence of these mite as parasites of these lizards.

The unidentified haemoparasites were too prevalent to be instances of a parasite infecting an atypical host (Chapter 6). Godfrey et al. (2006) demonstrated that the mobility of the vector has an influence on the prevalence and spatial distribution of their associated haemoparasites. The unidentified haemoparasites recorded herein had no association with particular habitats and were prevalent in areas associated with both tick species (Chapter 6). These unidentified haemoparasites need to be identified and additional research is needed to elucidate their natural history and the role they play in the ecology of sleepy lizards.

Even for some of the known parasites require empirical study to unravel their natural history. *Oochoristica trachysauri* has been recorded as a parasite of sleepy lizards in in parts of Queensland, New South Wales, Victoria, and South Australia (Johnston 1932). The intermediate hosts of *O. trachysauri* are not known, but likely a coleopteran and/or orthopteran (Gyawali et al. 2013, Norval et al. 2021a). Several of these arthropods have been recorded as food items of sleepy lizards (Norval and Gardner 2020), and additional empirical studies are needed to determine the life cycle of *O. trachysauri* and whether any of them are the intermediate hosts of this cestode.

It is not just the life cycles of the parasites that require additional research, but also their impact on the host. The impact of the various parasites on the fitness of sleepy lizards is not well known. The ticks *A. limbatum* and *B. hydrosauri* and the haemogregarine blood parasite *H. mariae* that infects the ticks and the lizards have been studied to some extent, and it was found that these parasites inhibit the activity of sleepy lizards (Main and Bull 2000, Bouma et al. 2007). Analysis of the long-term collected data at the Bunday Bore study site suggest that infestation by *B. hydrosauri* reduces survival in adult and juvenile lizards, and that *H. mariae* negatively impact the survival of juvenile lizards (Jones et al. 2016). However, these conclusions were often based on observations on lizards with manipulated parasitism intensities. In addition, some parasites may have beneficial impacts on the lizards that are being

overlooked. For instance, *T. trachysauri* has a very high prevalence in sleepy lizards (Angel and Mawson 1968, Jones 1992, Gyawali et al. 2013, Norval et al. 2021a). Fenner and Bull (2008) manipulated nematode (*Pharyngodon tiliquae* and *T. trachysauri*) infections in gidgee skinks (*Egernia stokesii*) with antihelminthic drugs and found that dewormed lizards spent more time basking and moved about for longer times during observation sessions. They concluded that the nematode infection alters the behaviour of the host and reduces its fitness. *Thelandros trachysauri*, like other members of the family Pharyngodonidae (order: Oxyurida), inhabit the posterior gut of the host and feed on bacterial flora (Adamson 1990), and are likely non-pathogenic. Considering that being exposed while basking and/or moving about can increase the risk of predation to a lizard, it could be that deworming the lizards removed a service, such as the promotion of digestion and absorption of nutrients, provided by the worms and as a result the lizards had to compensate by basking longer and possibly forage more. Further research should be directed at understanding the implications of the observed behavioural changes.

Another factor that should be considered is coinfection. When multiple parasite species infest and/or infect the host, their combined impact may be greater than that of each species on its own (Dunlap and Mathies 1993). But this is not always the case because coinfections may also entail interspecific competition between parasite species, which results in lower impact on the host than that of some of the parasites on their own (Thompson et al. 2018). Evidently more empirical studies are needed to determine the roles the various parasites play in the ecology of sleepy lizards and other species.

This study indicated that the sleepy lizards at the Bunday Bore study site are infected by an unidentified haemoparasites (Chapter 6), which potentially may be transmitted by the ticks *A. limbatum* and *B. hydrosauri*. There are likely other unidentified parasites in the parasitic arthropods recorded in this study. Staines et al. (2020) identified *Rickettsia* spp. associated with *B. hydrosauri* at this study site, and Stenos et al. (2003) recorded *Rickettsia honei*, the causative agent of Flinders Island spotted fever, and *Wolbachia* sp. in *B. hydrosauri* that were collected on Flinders Island. In addition, the snake mite is a possible vector of several blood-borne bacterial, filariid and viral pathogens of snakes (Wozniak and DeNardo 2000). Efforts should therefore be made to determine which micro-organisms infect sleepy lizards and their parasites and these organisms should be incorporated in future studies pertaining to the ecology of the

lizards and their parasites.

Ultimately a better understanding of the parasite communities of sleepy lizards can be instrumental in studies into aspects such as parasites as a selective force in speciation. Present evidence of parasite-driven speciation is derived from instances after reproductive isolation has already occurred (Nunn et al. 2004, Sommer et al. 2014, El Nagar and MacColl 2016) and empirical evidence for parasite-driven population divergence at the earliest stages of the speciation process is currently lacking. Recent analysis on population genetic structure within the sleepy lizard population at the Bunday Bore study site indicates a population genetic divergence in the major histocompatibility complex gene region, which has been suggested to drive speciation through mate choice (Andreou et al. 2017), appears to be associated with the two reptile tick species that parasitise the lizards (O'Reilly 2017). The ongoing long-term surveys and ecological studies of the lizard population at the Bunday Bore study site, combined with an indepth understanding of the associated parasite community, therefore provides a powerful model to ask fundamental questions about host diversification by parasites in the very early stages of the diversification process.

Conclusion

This thesis contributed to our understanding of the parasite community of a sleepy lizard population that inhabit an area that spans a precipitation-induced ecological gradient. The research presented herein indicates that the parasite community of this sleepy lizard population is more complex than has previously been reported, as in addition to the parasite species that have been recorded previously in this sleepy lizard population, some rare native ectoparasite species, an exotic invasive parasitic mite, as well as some unidentified haemoparasites were recorded. It was also found that there are associations between the different tick species and different habitats, which suggests that climate change and/or anthropogenic activities will alter the distribution of the tick species and their associated pathogens. My research also indicated that the overlap in the distribution of these ticks is complex and that future research should look at mapping the boundary in more detail to understand if it is wider than often cited as being. These findings indicate that monitoring of the distribution of the ticks and other parasites are needed, and that a better understanding of their ecological roles is required in order to make sound conservation and/or management decisions. It is hoped that the research of this thesis will contribute to a better understanding of both the host and the parasites, and that the

information will be used in management plans for the conservation of the lizards and their parasites.

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Appendix 7.1. Publications that are only partly or not associated with the PhD study

Note to examiners

In some ways my attitude towards ecology and research is similar to that of late Dr. Henry Fitch, a staunch advocate for the reporting on natural history observations, said, “When an observer is fortunate enough to see and record behaviour significant in the natural history of a species, his observations should be published. The advocates of biometrical methods need to recognize that some types of behaviour are not readily quantified because they are so rarely observed. Even a single observation may constitute a valuable contribution, and may be a break-through in understanding the species’ ecology. There are many kinds of anecdotes, and the fact that some are trivial is a poor excuse for condemning all narrative statements or accounts in scientific writing.”¹ Below is a list of publications, not directly or only partly associated with my PhD thesis, that I was involved in during my candidature. Where I am the first author, in all but two publications, I made all the observations. In all the publications where I am the first author, I drafted the manuscript and was responsible for making the changes as suggested by the reviewers and was the corresponding author for the final editing of the manuscript for publishing.

¹ Fitch, H.S. 1987. The sin of anecdotal writing. *Herpetological Review* **18**: 68.

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- Norval, G., C. S. Bursey, S. R. Goldberg, L. Morrison, and M. G. Gardner. 2021a. *Egernia striolata* (tree skink). Endoparasites. *Herpetological Review* **52**:406.
- Norval, G., J. Clayton, R. D. Sharrad, and M. G. Gardner. 2018. Notes on the stomach contents of a juvenile sleepy lizard, *Tiliqua rugosa* (Gray 1825), killed by an eastern brown snake, *Pseudonaja textilis* (Duméril, Bibron, and Duméril 1854) in South Australia. *IRCF Reptiles & Amphibians* **25**:200-203.
- Norval, G., and M. G. Gardner. 2018. Predation by an eastern brownsnake, *Pseudonaja textilis* (Duméril, Bibron, & Duméril 1854), on a mallee black-backed snake, *Parasuta nigriceps* (Günther 1863). *IRCF Reptiles & Amphibians* **25**:134-136.
- Norval, G., R. D. Sharrad, and M. G. Gardner. 2019a. Sand goanna (*Varanus gouldii*) predation on a painted dragon (*Ctenophorus pictus*) and a mulga parrot (*Psephotellus varius*) in South Australia. *IRCF Reptiles & Amphibians* **26**:132–133.
- Norval, G., R. D. Sharrad, and M. G. Gardner. 2019b. Two instances in South Australia of sleepy lizards, *Tiliqua rugosa* (Gray 1825), feeding on plant species not previously recorded as part of this lizard's diet. *IRCF Reptiles & Amphibians* **26**:54-55.
- Norval, G., R. D. Sharrad, and M. G. Gardner. 2020. Three instances of reptile ticks parasitising humans. *Acarologia* **60**:607-611.
- Norval, G., R. D. Sharrad, and M. G. Gardner. 2021b. *Demansia psammophis* (yellow-faced whip snake), *Notechis scutatus* (tiger snake), *Pogona barbata* (eastern bearded dragon). Ectoparasite. *Herpetological Review* **52**:421-422.
- Norval, G., R. D. Sharrad, K. E. Ross, and M. G. Gardner. 2021c. An instance of parasitism on a human by a nymph of the kangaroo soft tick, *Ornithodoros gurneyi* Warburton, 1926 (Acari: Argasidae) in South Australia. *Ticks and Tick-borne Diseases* **12**:101632.

Appendix 7.2. The Braun-Blanquet cover abundance scores of the species recorded during the March 2018 surveys at the Bunday Bore study site.

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	
SCIENTIFIC NAME																														
<i>Acacia nyssophylla</i>																														r
<i>Atriplex vesicaria</i>											2	+		3	r	+	r	1						1	2	+	2	2	2	2
<i>Austrodanthonia caespitosa</i>			+				r	r		r									r	r	+									r
<i>Austrostipa nodosa</i>	+						r																							r
<i>Carrichtera annua</i>	+	1	r	+	1	1		1	1	r					1	+			1	2		+	r			r	r		+	
<i>Casuarina pauper</i>											+	3	3	4																
<i>Chenopodium curvispicatum</i>		r					r			+			r	r						r		+			+			+	+	
<i>Enchylaena tomentosa</i>					r	r		r	+		r	+	+	+		+	+	r		r		r		r	1	r	+	+	+	
<i>Eremophila glabra</i>																														1
<i>Eriochiton sclerolaenoides</i>																										r				
<i>Eucalyptus oleosa</i>																								3	2		2	2		
<i>Eucalyptus socialis</i>																		2												
<i>Geijera linearifolia</i>																	r													
<i>Maireana brevifolia</i>																								r	r					
<i>Maireana pentatropis</i>																								r	+			1	r	
<i>Maireana pyramidata</i>			r		2	2		1	2		1	3	r	r				1	2			r					r	r		
<i>Maireana sedifolia</i>	2	2	1	r	r	1	2	1		2	r		2	r	2	2	2		2	2	2	2	2	2	2	2		r	r	1
<i>Maireana trichoptera</i>	+	1	+	1				1							+	+			1	r	+		1	1						
<i>Maireana turbinata</i>											+		r																	
<i>Medicago minima</i>	r	r	r	r	r	r	+	r	r	r	r				r	r		r	r			r								
<i>Minuria leptophylla</i>	r																													
<i>Myoporum platycarpum</i>																														1
Appendix 7.2 continued																														
Sample plots	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	

<i>Senna artemisioides coriacea</i>	2	2	+	2	2														3	1					
<i>Senna artemisioides filifolia</i>	1	2			3	3																			
<i>Sida intricata</i>				r																					
<i>Templetonia egea</i>						2																			
<i>Vittadinia gracilis</i>									r		+	+			r			r	r	r	+		r	r	
<i>Zygophyllum aurantiacum</i>	r	r	r						1	r	+								r						

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86
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SCIENTIFIC NAME

<i>Acacia nyssophylla</i>							r																						
<i>Asphodelus fistulosus</i>																									1				
<i>Atriplex vesicaria</i>							r	+			2	2	r	2	2	r	+	2	2	1	1	1	+	r	r				
<i>Austroanthonia caespitosa</i>	1	+	+	r	1	1		r	r	+	r	r	r					r	r									+	r
<i>Austrostipa nodosa</i>					r		r							r				+		r		+				r			
<i>Carrichtera annua</i>	+		+	+	+	r	+	+	r	r	+	+	+	+	+	+		+	r		r		r	r	r	r	r	1	+
<i>Carthamus lanatus</i>				+																									
<i>Chamaesyce drummondii</i>																													
<i>Chenopodium curvispicatum</i>								r		r			+	r	r	r	r			r	r								
<i>Convolvulus remotus</i>											r																		
<i>Cratystylis conocephala</i>																													
																					3	3							

Appendix 7.2 continued

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86
-------------	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----

<i>Dissocarpus paradoxus</i>												r	r							r								r	r
<i>Enchylaena tomentosa</i>		r			+		+	+		r	r	+	+	r	r		+		r	r		r			r	r	r		
<i>Eremophila deserti</i>							1																						
<i>Eremophila scoparia</i>															+					2									
<i>Eucalyptus oleosa</i>											2	2	2	1	3	2	3				1			3					
<i>Exocarpos aphyllus</i>																		2											

Vittadinia gracilis

r r

Zygophyllum aurantiacum

+ + + + r + r + r r r + + r

Appendix 7.3. The Braun-Blanquet cover abundance scores of the species recorded during the June 2018 surveys at the Bunday Bore study site.

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	
SCIENTIFIC NAME																														
<i>Acacia nyssophylla</i>																													r	
<i>Asphodelus fistulosus</i>		+																		r										
<i>Atriplex vesicaria</i>										2	+		3	r	+	r	1						1	2	+	2	2	2	2	
<i>Austroanthonia caespitosa</i>			+				r	r		r			r						r	r	+								r	
<i>Austrostipa nodosa</i>	+						r																	r					r	
<i>Carrichtera annua</i>	1	1	r	+	1	1		1	1	r	+	+	r	+	1	+		+	1	2	r	+	r			+	+	r	+	
<i>Casuarina pauper</i>											+	3	3	4																
<i>Chenopodium curvispicatum</i>		r					r			+			r	r						r		+			+			+	+	
<i>Enchylaena tomentosa</i>					r	r		r	+		r	+	+	+		+	r	r		r		r		r	1	r	+	+	+	
<i>Eremophila glabra</i>																													1	
<i>Eriochiton sclerolaenoides</i>																									r					
<i>Erodium crinitum</i>	+	+		+	r	r		+	+				r	+	+				+	+										
<i>Eucalyptus oleosa</i>																								3	2		2	2		
<i>Eucalyptus socialis</i>																	2													
<i>Geijera linearifolia</i>																	r													
<i>Maireana brevifolia</i>																								r	r					
<i>Maireana pentatropis</i>																								r	+			1	r	
<i>Maireana pyramidata</i>			r		2	2		1	2		1	3	r	r			1	2				r					r	r		
<i>Maireana sedifolia</i>	2	2	1	r	r	1	2	1		2	r		2	r	2	2	2		2	2	2	2	2	2	2	2	2	r	r	1
<i>Maireana trichoptera</i>	+	1	+	1				1							+	+			1	r	+		1	1						
<i>Maireana turbinata</i>											+		r																	
<i>Medicago minima</i>	r	r	r	+	r	+	+	r	r	r	r	r	r	r	+	+			+	+	+	r	r			r	r			

Appendix 7.3 continued

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	
<i>Minuria leptophylla</i>	r																													
<i>Myoporum platycarpum</i>																													1	
<i>Olearia calcarea</i>																								r						
<i>Pittosporum angustifolium</i>	1																													
<i>Rhagodia spinescens</i>											r			1									+		1			1	r	
<i>Rhagodia ulicina</i>											1		r									r		r	+	+	r	r	r	
<i>Salsola kali</i>																													r	
<i>Salvia verbenaca</i>		r		r	r	+							r																	
<i>Scaevola spinescens</i>																														
<i>Sclerolaena obliquicuspis</i>	r	1		+	1	2	r	1	+	r			r	r		r					+		+			r	r	r	+	
<i>Sida fibulifera</i>																														r
<i>Sida intricata</i>	+												r																	
<i>Templetonia egena</i>																														3
<i>Zygophyllum aurantiacum</i>																								r	r	r	+	1	1	

Sample plot	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58
SCIENTIFIC NAME																													
<i>Acacia oswaldii</i>			r																										
<i>Alectryon oleifolius</i>											3																		
<i>Asphodelus fistulosus</i>																								2	1				
<i>Atriplex vesicaria</i>			r	r		+			2	1													r						
<i>Austroanthonia caespitosa</i>		+	+	+	+	+	1	1	r	+			+	+	r		+		r	r	r	r	+	r	+			r	
<i>Austrostipa nodosa</i>	r	r	r		r	+				+	+	r	+	+	r	r	r	r								r	r	r	r

Appendix 7.3 continued

Sample plot	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58
-------------	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----	----

<i>Carrichtera annua</i>	+		+	1	+	r	+	+	r	r	1	+	+	+	+	1	r	+	r		+	+	r	+	+	r	1	1						
<i>Carthamus lanatus</i>				+																														
<i>Chamaesyce drummondii</i>																						+												
<i>Chenopodium curvispicatum</i>									r	r			+	r	r	r	r		r	r														
<i>Citrullus lanatus</i>				+																								r						
<i>Convolvulus remotus</i>										r																								
<i>Cratystylis conocephala</i>																																		
<i>Dissocarpus paradoxus</i>											r	r																r	r					
<i>Enchylaena tomentosa</i>		r			r		+	+		r	r	+	+	r	r		+		r	r		r			r	r	r							
<i>Eremophila deserti</i>							1																											
<i>Eremophila scoparia</i>															+																			
<i>Erodium crinitum</i>	+		+		r			+																										
<i>Eucalyptus oleosa</i>											2	2	2	1	3	2	3											1	3					
<i>Exocarpos aphyllus</i>																													2					
<i>Geijera linearifolia</i>																													r					
<i>Hakea leucoptera</i>								r																										
<i>Lepidium leptopetalum</i>														1	r	1	+					2		+	+	r								
<i>Lysiana exocarpi</i>																													r					
<i>Maireana pentatropis</i>												+	+	1	1	+	1	+	1	+	+	+				r	r							
<i>Maireana pyramidata</i>																													r	2	3	2	1	3
<i>Maireana sedifolia</i>	2	2	2	2	2	1	2	2	2	2	2	r	2				r	r																

Appendix 7.3 continued

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	
<i>Maireana trichoptera</i>	r	r						+			r	+	r				r	r	r	r	r			r		r			
<i>Marrubium vulgare</i>																							1		+				
<i>Medicago minima</i>	r	+	r	+	+	r	r	r	r	+		+						r				+	1	r	r			+	+

Appendix 7.4. The Braun-Blanquet cover abundance scores of the species recorded during the September 2018 surveys at the Bunday Bore study site.

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	
SCIENTIFIC NAME																														
<i>Acacia nyssophylla</i>																														r
<i>Atriplex vesicaria</i>											2	+		3	r	+	r	1						1	2	+	2	2	2	2
<i>Austrodanthonia caespitosa</i>			+	r			r	r		r			r						r	r	r									r
<i>Austrostipa nodosa</i>	+						r																							r
<i>Avena fatua</i>												r																		
<i>Carrichtera annua</i>	+	2	r	+	1	1		1	1	r	r	r	r	r	1	+		r	1	2			+					r	r	+
<i>Casuarina pauper</i>											+	3	3	4																
<i>Chenopodium curvispicatum</i>							r			+			r	r							r		r			+			+	+
<i>Enchylaena tomentosa</i>					r			r		r	r	+	1	+		+	+	r			r	r	r		r	1	r	+	+	+
<i>Eremophila glabra</i>																														1
<i>Eriochiton sclerolaenoides</i>													r										r			r				
<i>Erodium crinitum</i>		+				r														r										
<i>Eucalyptus oleosa</i>																								3	2		2	2		
<i>Eucalyptus socialis</i>																	2													
<i>Geijera linearifolia</i>																	r													
<i>Hyalosperma semisterile</i>				+									+		+			r	+	r	+	+								
<i>Maireana brevifolia</i>																										r				
<i>Maireana pentatropis</i>																								r	+			1	r	
<i>Maireana pyramidata</i>					2	2		1	2		1	3	r	r		r	1	2				r					r	r		
<i>Maireana sedifolia</i>	2	2	1	r	r	1	2	1		2	r		2	r	2	2	2		2	2	2	2	2	2	2	2		r	r	1
<i>Maireana trichoptera</i>	+	1	+	1				1							+	+			+	r	r		1	1						
Appendix 7.4 continued																														
Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	

<i>Maireana trichoptera</i>	r	r								+			r	+	r			r	r	r	r	r			r		+					
<i>Marrubium vulgare</i>																						1			+							
<i>Medicago minima</i>	r	+	r	+	r	r	r	r	r	+								r				+	r		r		+	+				
<i>Nicotiana velutina</i>																						1										
<i>Olearia calcarea</i>			r					r																								
<i>Olearia magniflora</i>																									r	r						
<i>Pittosporum angustifolium</i>			r																													
<i>Rhagodia crassifolia</i>																											1					
<i>Rhagodia spinescens</i>																											+					
<i>Rhagodia ulicina</i>													1	+	1			1	+	r	2	+	r									
<i>Salsola kali</i>																											r					
<i>Salvia verbenaca</i>				+	+																						+					
<i>Scaevola spinescens</i>																											+	1				
<i>Sclerolaena obliquicuspis</i>	r	r	r	1	r	+	+	+	r	+			+	+	+												1	+	+	+	r	+
<i>Senna artemisioides coriacea</i>																												3	3			

Appendix 7.4 continued

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86					
<i>Senna artemisioides filifolia</i>																		+				2	3										
<i>Sida intricata</i>			+	r		r																											
<i>Sisymbrium irio</i>																																	1
<i>Vittadinia gracilis</i>			r	r																												r	
<i>Zygophyllum aurantiacum</i>												+	+	+	+	r	+	r	+	r	r		r	+	+	r							

Appendix 7.5. The Braun-Blanquet cover abundance scores of the species recorded during the December 2018 surveys at the Bunday Bore study site

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	
SCIENTIFIC NAME																														
<i>Acacia nyssophylla</i>																													r	
<i>Atriplex vesicaria</i>											1	+		2	r	+	r	1					1	2	+	2	2	1	2	
<i>Austrodanthonia caespitosa</i>			r	r			r	r		r			r									r								r
<i>Austrostipa nodosa</i>	+						r																							r
<i>Carrichtera annua</i>	+	1	r	r	+	+		1	1	r	r		r		1	+			+	1		r							r	+
<i>Casuarina pauper</i>											+	3	3	4																
<i>Chamaesyce drummondii</i>					+																									
<i>Chenopodium curvispicatum</i>	r	r					r			+			r								r	r				r		+	+	
<i>Enchylaena tomentosa</i>					r		r	r		r	r	+	+	+		+	+	r			r	r	r		r	1	r	+	+	+
<i>Eremophila glabra</i>																														1
<i>Eriochiton sclerolaenoides</i>													r										r			r				
<i>Erodium crinitum</i>		+				r																								
<i>Eucalyptus oleosa</i>																								3	2		2	2		
<i>Eucalyptus socialis</i>																	2													
<i>Geijera linearifolia</i>																	r													
<i>Heliotropium europaeum</i>					+																									
<i>Hyalosperma semisterile</i>																					r	r								
<i>Maireana brevifolia</i>																										r				
<i>Maireana pentatropis</i>																									r	+		1	r	
<i>Maireana pyramidata</i>					2	2		1	2		1	3	r	r		r	1	1				r						r	r	
<i>Maireana sedifolia</i>	2	2	1	r	r	1	2	1		2	r		2	r	2	2	2		2	2	2	2	2	2	2	2		r	r	1

Appendix 7.5 continued

Sample plot	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29
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Zygophyllum aurantiacum

r r r r l r +

r

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86
SCIENTIFIC NAME																												
<i>Acacia nyssophylla</i>							r																					
<i>Asphodelus fistulosus</i>																									+			
<i>Atriplex vesicaria</i>							r	+			2	2	r	1	2	r	+	1	2	1	1	1	+	r	r			
<i>Austrodanthonia caespitosa</i>	+	+	+	r	1	1		r	r	+	r	+						r	r	+								+
<i>Austrostipa nodosa</i>					r		r							r				+		r		+				r		
<i>Carrichtera annua</i>	r		+	+	r	r	+	+	r	r	r	+	r	+	r	r		r	r					r	+	r	+	r
<i>Chenopodium curvispicatum</i>							r	r		r			+	r	r	r	r		r	r			r		r			
<i>Convolvulus remotus</i>			r																									
<i>Cratystylis conocephala</i>																				3	3							
<i>Dissocarpus paradoxus</i>	r										r	r																r
<i>Enchylaena tomentosa</i>		r					+	+			r	+	+	r	r		+		r	r		r		r	+	r	r	
<i>Eremophila deserti</i>							1																					
<i>Eremophila scoparia</i>															+					2								

Appendix 7.5 continued

Sample plot	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86
<i>Eucalyptus oleosa</i>											2	2	2	1	3	2	3			1			3					
<i>Exocarpos aphyllus</i>																	2											
<i>Geijera linearifolia</i>																	r											
<i>Hakea leucoptera</i>							r																					
<i>Heliotropium europaeum</i>																												+
<i>Lepidium leptopetalum</i>														1	r	1	+		2		+	r	r					
<i>Lysiana exocarpi</i>																	r											

Appendix 7.6. The lizards that were examined for my PhD research and the parasites that were recorded from them

ID	lizard toe-clip number
Date	capture date
SVL	snout-vent length
BM	body mass
-	sample not collected
IIT 1	Intraerythrocytic inclusion type 1
IIT 2	Intraerythrocytic inclusion type 2
IIT 3	Intraerythrocytic inclusion type 3
G	Giemsa stain
WG	Wright-Giemsa stain
*	Not suitable for determining parasitemia

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i> <i>limbatum</i>	<i>Bothriocroton</i> <i>hydrosauri</i>	<i>Ornithodoros</i> <i>gurneyi</i>	<i>Ophionyssus</i> <i>natricis</i>	Chigger
623	31/08/2017	13:13	-33.91101	139.35620	male	290	725	1	0	0	-	-
17904	31/08/2017	14:52	-33.90908	139.31631	female	293	625	1	0	0	-	-
8040	1/09/2017	12:20	-33.90418	139.32449	female	265	600	1	0	0	-	-
9389	1/09/2017	14:35	-33.92007	139.35612	male	290	625	1	0	0	-	-
9373	1/09/2017	14:35	-33.92007	139.35612	female	300	700	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
17442	4/09/2017	15:45	-33.92007	139.35612	male	284	500	0	1	0	-	-
9373	6/09/2017	12:43	-33.92038	139.35619	female	325	700	0	0	0	-	-
5463	6/09/2017	13:10	-33.99239	139.35664	female	308	750	0	1	0	-	-
12962	13/09/2017	12:30	-33.89448	139.35608	female	318	750	1	0	0	-	-
501	13/09/2017	12:35	-34.11604	139.35690	male	323	600	1	0	0	-	-
27000	13/09/2017	14:23	-33.89598	139.35616	male	294	525	1	0	0	-	-
8506	14/09/2017	13:42	-33.91063	139.31390	female	310	600	1	1	0	-	-
10392	14/09/2017	13:42	-33.91063	139.31390	male	321	675	1	0	0	-	-
27001	15/09/2017	9:50	-33.88664	139.35323	female	233	300	0	0	0	-	-
11701	15/09/2017	12:00	-33.88707	139.35251	male	336	825	1	0	0	-	-
8973	19/09/2017	14:10	-33.90754	139.35620	male	306		1	0	0	-	-
27003	19/09/2017	15:39	-33.92817	139.30365	female	300	625	1	1	0	-	-
27002	19/09/2017	15:39	-33.92817	139.30365	male	319	625	1	0	0	-	-
27004	20/09/2017	11:00	-33.92824	139.30377	female	237	300	0	0	0	-	-
14937	20/09/2017	11:54	-33.95774	139.23865	female	325	675	0	1	0	-	-
12749	25/09/2017	13:40	-33.88597	139.35535	male	320	825	1	0	0	-	-
10773	25/09/2017	13:50	-33.88596	139.35506	female	338	900	1	1	0	-	-
854B	25/09/2017	14:53	-33.88598	139.35448	male	292	525	0	0	0	-	-
11670	25/09/2017	15:18	-33.88818	139.35075	male	324	775	0	0	0	-	-
4054	25/09/2017	16:03	-33.92031	139.29790	female?	310	700	1	0	0	-	-
27005	26/09/2017	12:15	-34.04074	139.35666	male	307	550	0	1	0	-	-
27006	26/09/2017	12:45	-33.99982	139.35658	male	273	425	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27007	26/09/2017	13:05	-33.99848	139.35664	male	310	625	0	1	0	-	-
2864	26/09/2017	13:45	-33.96051	139.35632	male	319	750	0	0	0	-	-
11500	26/09/2017	14:10	-33.93203	139.35629	female	312	675	1	0	0	-	-
11829	26/09/2017	15:55	-33.89765	139.33521	male	325	700	1	0	0	-	-
8012	27/09/2017	10:40	-33.89876	139.33330	female	307	625	1	0	0	-	-
8743	27/09/2017	10:40	-33.89876	139.33330	male	334	675	1	0	0	-	-
1595	27/09/2017	10:42	-33.89867	139.33308	female	315	725	0	0	0	-	-
12225	27/09/2017	11:40	-33.90499	139.32306	male	310	550	1	0	0	-	-
27008	27/09/2017	11:45	-33.90571	139.32195	female	285	500	0	1	0	-	-
2726	27/09/2017	12:30	-33.89570	139.33847	male	293	625	1	0	0	-	-
11051	27/09/2017	12:45	-33.89383	139.34131	female	318	675	0	0	0	-	-
3787	1/10/2017	16:55	-34.06472	139.35670	male	287	425	0	1	0	-	-
17067	2/10/2017	11:00	-33.96201	139.23235	male	286	525	0	0	0	-	-
27018	2/10/2017	11:33	-33.96750	139.22385	male	287	600	0	0	0	-	-
1100	2/10/2017	11:33	-33.96750	139.22385	female	294	625	0	1	1	-	-
27009	2/10/2017	13:38	-33.92201	139.29503	female	302	575	1	1	0	-	-
11256	2/10/2017	13:38	-33.92201	139.29503	male	325	700	1	0	0	-	-
29000	2/10/2017	13:45	-33.96818	139.22295	male	303	658	0	0	0	-	-
525	2/10/2017	17:41	-33.88966	139.34819	female	310	625	1	0	0	-	-
1525	3/10/2017	9:38	-33.89817	139.33453	female	324	725	0	0	0	-	-
14160	3/10/2017	9:38	-33.89817	139.33453	male	305	625	1	0	0	-	-
1609	3/10/2017	10:40	-33.90333	139.32593	male	305	625	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
4855	3/10/2017	11:10	-33.90834	139.31758	male	287	450	1	1	0	-	-
27010	3/10/2017	11:37	-33.90546	139.32239	male	255	375	1	0	0	-	-
27011	16/10/2017	11:23	-33.90189	139.35617	male	282	550	0	0	0	-	-
17153	16/10/2017	11:25	-33.90213	139.35619	female	323	800	1	0	0	-	-
27012	16/10/2017	14:33	-33.95138	139.24835	male	294	575	0	1	0	-	-
27014	16/10/2017	16:42	-33.92823	139.31145	female	288	500	1	0	0	-	-
27013	16/10/2017	17:10	-33.92822	139.30217	female	299	600	1	0	0	-	-
27020	17/10/2017	9:03	-33.88656	139.35335	-	192	200	0	0	0	-	-
2010	17/10/2017	9:23	-33.88834	139.35046	male	317	700	0	0	0	-	-
5530	17/10/2017	10:00	-33.89622	139.33742	female	309	750	1	0	0	-	-
5561	17/10/2017	10:38	-33.89821	139.33432	female	308	775	1	0	0	-	-
2342	17/10/2017	10:38	-33.89821	139.33432	male	304	750	0	0	0	-	-
27015	17/10/2017	10:42	-33.89801	139.33463	female	308	725	1	0	0	-	-
27016a	17/10/2017	12:33	-33.90925	139.31607	female	298	575	1	0	0	-	-
27016b	24/10/2017	11:33	-33.96729	139.22433	female	271	450	0	0	0	-	-
27017	24/10/2017	11:35	-33.88680	139.33049	male	291	475	0	0	1	-	-
27019	24/10/2017	14:51	-33.88984	139.34793	female	259	475	0	0	0	-	-
5561	24/10/2017	15:38	-33.89792	139.33475	female	314	750	0	0	0	-	-
2342	24/10/2017	15:38	-33.89792	139.33475	male	310	750	0	0	0	-	-
27021	24/10/2017	16:30	-33.93909	139.26704	male	300	525	0	1	0	-	-
17164	25/10/2017	10:42	-33.88599	139.35530	female	287	650	1	1	0	-	-
27022	25/10/2017	10:42	-33.88599	139.35530	female	223	275	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27024	25/10/2017	10:42	-33.88599	139.35530	male	206	250	0	0	0	-	-
27023	25/10/2017	10:42	-33.88599	139.35530	male	188	200	0	0	0	-	-
27001	30/10/2017	13:07	-33.88699	139.35271	female	242	325	0	0	0	-	-
12331	30/10/2017	13:29	-33.88793	139.35106	male	310	675	1	0	0	-	-
11833	30/10/2017	14:11	-33.91151	139.31244	female	284	675	1	0	0	-	-
27025	30/10/2017	15:14	-33.95469	139.24330	female	287	450	0	0	0	-	-
553	30/10/2017	15:52	-33.97160	139.22134	male	284	475	0	0	0	-	-
7655	30/10/2017	16:26	-33.95656	139.24039	male	301	575	0	0	0	-	-
27026	31/10/2017	11:18	-33.96742	139.22385	male	296	550	0	0	0	-	-
27027	31/10/2017	12:18	-33.91376	139.30862	male	302	550	1	0	0	-	-
10860	5/11/2017	14:35	-33.90684	139.32005	male	306	575	1	0	0	-	-
826	5/11/2017	14:35	-33.90684	139.32005	female	319	675	1	0	0	-	-
27026	5/11/2017	16:00	-33.96700	139.22469	male	292	525	0	0	0	-	-
11592	5/11/2017	16:50	-33.89764	139.33516	female	313	750	1	0	0	-	-
27028	6/11/2017	18:28	-33.99656	139.35664	male	304	625	0	1	0	-	-
837	7/11/2017	13:23	-33.95368	139.24469	male	298	575	0	0	0	-	-
528	7/11/2017	14:15	-33.91185	139.31194	female	318	750	1	0	0	-	-
27029	7/11/2017	15:05	-33.88552	139.35510	female	205	250	0	0	0	-	-
2208	7/11/2017	16:55	-33.97150	139.27895	female	314	675	0	1	0	-	-
10692	7/11/2017	18:02	-33.92822	139.29881	female	293	625	1	0	0	-	-
11391 special	8/11/2017	10:10	-33.89453	139.34019	male	284	500	1	0	0	-	-
11256	8/11/2017	11:08	-33.92200	139.29518	male	312	675	1	0	1	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27030	8/11/2017	12:00	-33.95950	139.23596	female	294	525	0	1	1	-	-
27031	8/11/2017	12:50	-33.90360	139.32536	female	297	500	1	0	0	-	-
12853	13/11/2017	17:26	-33.93372	139.35625	male	290	625	1	1	0	-	-
10508	13/11/2017	18:14	-33.92822	139.30714	female	307	550	1	0	0	-	-
525	14/11/2017	8:31	-33.88966	139.34824	female	310	625	0	0	0	-	-
8855	14/11/2017	9:02	-33.89320	139.34254	male	311	575	0	0	0	-	-
4952	14/11/2017	9:02	-33.89320	139.34254	female	308	700	1	0	0	-	-
8707	14/11/2017	15:13	-33.90332	139.32571	female	326	725	1	0	0	-	-
14426	20/11/2017	15:01	-33.91010	139.31442	female	311	700	1	0	0	-	-
5530	20/11/2017	17:53	-33.89666	139.33664	female	311	775	1	0	0	-	-
3053	20/11/2017	18:23	-33.89216	139.34412	female	303	675	1	0	0	-	-
854	21/11/2017	8:35	-33.88604	139.35403	male	318	700	0	0	0	-	-
854B	21/11/2017	9:02	-33.88599	139.35446	male	292	575	0	0	0	-	-
17183	21/11/2017	10:09	-33.90087	139.32990	female	319	750	1	0	0	-	-
12795	21/11/2017	10:50	-33.91385	139.30867	female	320	750	1	0	0	-	-
27032	21/11/2017	12:21	-33.97142	139.33258	-	285	625	0	1	0	-	-
2728	21/11/2017	17:55	-33.88682	139.32999	female	310	700	1	0	0	-	-
27033	21/11/2017	18:33	-33.88984	139.29002	female	219	275	1	0	0	-	-
17011	22/11/2017	8:58	-33.88640	139.35353	female	289	650	1	0	0	-	-
27034	22/11/2017	9:25	-33.88678	139.35304	-	186	200	0	0	0	-	-
9910	22/11/2017	10:10	-33.90113	139.32951	female	321	725	1	0	0	-	-
27035	22/11/2017	10:55	-33.93241	139.27808	female	288	600	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
10075	27/11/2017	9:48	-33.97163	139.26550	female	311	700	0	1	0	-	-
1186 special	27/11/2017	10:40	-33.97156	139.27986	male	306	675	0	1	0	-	-
12458	27/11/2017	11:20	-33.97152	139.27592	male	317	725	0	1	0	-	-
27037	27/11/2017	12:32	-33.91307	139.30978	female	211	250	0	0	0	-	-
8878	27/11/2017	13:00	-33.91044	139.31406	female	314	825	1	0	0	-	-
12364	27/11/2017	13:40	-33.90088	139.32980	female	306	600	1	0	0	-	-
17530	27/11/2017	17:30	-33.97165	139.31567	female	301	675	0	1	0	-	-
11460	28/11/2017	9:23	-33.88509	139.35341	male	302	625	1	0	0	-	-
Stumpy	28/11/2017	9:23	-33.88507	139.35358	female	298	625	1	0	0	-	-
27038	28/11/2017	10:28	-33.88509	139.35323	female	241	350	1	0	0	-	-
27039	28/11/2017	11:15	-33.88997	139.28804	female	301	575	1	1	0	-	-
8177	3/12/2017	16:12	-34.00756	139.35664	male	302	650	0	1	0	-	-
11284	4/12/2017	9:12	-33.92826	139.29788	female	303	700	1	1	0	-	-
17137	4/12/2017	10:53	-33.97152	139.24487	male	306	600	0	1	0	-	-
2391	4/12/2017	11:55	-33.92107	139.29655	female	316	875	1	0	0	-	-
7511	4/12/2017	12:24	-33.92067	139.29726	female	325	775	1	0	0	-	-
10844	4/12/2017	13:09	-33.90141	139.32896	male	308	775	1	0	0	-	-
3053	4/12/2017	13:52	-33.89279	139.34323	female	308	725	1	0	0	-	-
10773	4/12/2017	14:20	-33.88605	139.35432	female	330	1100	1	0	0	-	-
12749	4/12/2017	14:45	-33.88609	139.35522	male	314	875	1	0	0	-	-
12860	4/12/2017	16:00	-33.88680	139.35529	female	308	825	1	0	0	-	-
27040	4/12/2017	17:10	-33.91686	139.27037	female	322	800	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
555	5/12/2017	9:55	-33.97171	139.23102	male	292	600	0	0	0	-	-
12442	5/12/2017	10:23	-33.97165	139.23404	female	316	675	0	0	0	-	-
11644	5/12/2017	10:51	-33.97160	139.23033	female	287	600	0	0	0	-	-
27041	5/12/2017	11:23	-33.96433	139.22884	female	288	600	0	1	0	-	-
27042	5/12/2017	12:17	-33.94436	139.25891	female	258	400	0	1	0	-	-
6027	5/12/2017	12:54	-33.93084	139.28072	male	311	650	0	0	0	-	-
5034	5/12/2017	13:38	-33.90923	139.31609	female	321	800	1	0	0	-	-
41804	11/12/2017	11:50	-33.88768	139.30411	male	311	725	1	1	0	-	-
14533	11/12/2017	16:54	-33.90125	139.27026	male	294	625	0	1	0	-	-
27043	11/12/2017	17:20	-33.89796	139.27016	male	308	650	0	1	0	-	-
5692	11/12/2017	18:05	-33.89706	139.27026	female	328	750	0	1	0	-	-
4870	11/12/2017	18:58	-33.90958	139.31564	female	312	700	1	0	0	-	-
27044	11/03/2018	11:22	-33.93372	139.35626	female	276	425	1	1	0	-	-
8567	11/03/2018	11:22	-33.93372	139.35626	female	295	625	1	1	0	-	-
27045	11/03/2018	17:45	-33.89696	139.35619	female	285	400	1	0	1	-	-
27046	23/03/2018	13:20	-34.01046	139.35664	female	315	725	0	1	0	-	-
1922	24/03/2018	11:40	-33.98855	139.35664	female	323	675	0	1	0	-	-
17220	15/04/2018	13:32	-33.88531	139.35068	female	322	550	1	0	0	-	-
14611	20/05/2018	13:14	-33.91306	139.30994	female	282	425	0	0	0	-	-
10510	13/08/2018	12:15	-33.88736	139.32291	male	305	525	0	0	0	-	-
9367	13/08/2018	12:56	-33.88811	139.31337	male?	315	625	1	1	0	-	-
12598	13/08/2018	13:29	-33.88889	139.30186	male?	317	650	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27047	27/08/2018	12:30	-33.89104	139.27368	female?	292	550	0	0	0	-	-
9511	27/08/2018	13:19	-33.89066	139.27916	male?	305	575	0	1	0	-	-
876	27/08/2018	13:45	-33.89009	139.28583	male?	302	500	1	1	0	-	-
14138	1/09/2018	13:12	-33.89444	139.22823	female?	286	450	0	1	0	-	-
10839	1/09/2018	13:37	-33.88756	139.31955	male?	324	750	1	0	0	-	-
27048	2/09/2018	12:00	-33.88625	139.35515	female?	287	675	1	0	0	-	-
27049	2/09/2018	12:57	-33.88654	139.33377	male?	290	450	0	0	0	-	-
11833	2/09/2018	14:27	-33.91184	139.31194	male?	294	650	1	0	0	-	-
854B	9/09/2018	12:25	-33.88590	139.35439	male?	295	575	1	0	0	-	-
1000	9/09/2018	13:07	-33.88696	139.35272	female?	279	525	1	0	0	-	-
22849 special	9/09/2018	13:45	-33.89744	139.33543	male?	304	575	0	0	0	-	-
9910	9/09/2018	14:12	-33.90109	139.32951	female?	312	650	1	0	0	-	-
11833	9/09/2018	14:59	-33.91175	139.31221	female?	296	625	1	0	0	-	-
5040	9/09/2018	15:42	-33.91272	139.31053	male?	308	675	1	0	0	-	-
3101	9/09/2018	16:39	-33.89359	139.34192	male?	323	750	1	0	0	-	-
4876	9/09/2018	18:07	-33.88655	139.35347	female	313	675	1	0	0	-	-
11701	9/09/2018	18:07	-33.88655	139.35347	male	326	775	1	0	0	-	-
12606	10/09/2018	12:05	-33.89136	139.26961	male?	311	550	0	1	0	-	-
27050	10/09/2018	12:46	-33.88923	139.29744	male?	284	500	1	0	0	-	-
27051	10/09/2018	13:39	-33.88876	139.30437	male?	286	450	0	0	0	-	-
17013	15/09/2018	12:25	-33.88565	139.35506	female?	313	750	1	0	0	-	-
27727	15/09/2018	13:07	-33.88587	139.34320	female?	286	475	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27052	15/09/2018	14:09	-33.88974	139.29060	male?	302	525	1	0	0	-	-
876	15/09/2018	14:46	-33.89009	139.28581	female?	302	550	0	0	0	-	-
11612	15/09/2018	15:21	-33.89297	139.27023	female?	308	625	0	0	0	-	-
7839	16/09/2018	10:08	-33.90083	139.33002	male?	321	600	1	0	0	-	-
6103	16/09/2018	10:54	-33.90280	139.32658	female?	306	675	0	0	0	-	-
5820	16/09/2018	11:27	-33.91213	139.31133	female?	308	750	1	0	0	-	-
4880	16/09/2018	12:15	-33.91988	139.29860	female?	308	750	1	1	0	-	-
12456	16/09/2018	12:57	-33.92239	139.29449	male?	288	550	1	0	0	-	-
27053	16/09/2018	14:05	-33.88786	139.31584	male?	265	400	0	1	0	-	-
11724	23/09/2018	15:47	-33.89076	139.27579	female?	319	725	0	1	0	-	-
32494 special	24/09/2018	10:32	-33.88792	139.31583	female?	298	675	1	1	0	-	-
27054	24/09/2018	11:24	-33.88546	139.34825	female?	283	500	1	0	0	-	-
27055	29/09/2018	13:07	-33.89168	139.34492	female?	302	625	0	0	0	-	-
8770	29/09/2018	13:46	-33.89260	139.34341	female?	324	750	1	0	0	-	-
8743	29/09/2018	14:27	-33.89852	139.33365	male	323	625	1	0	0	-	-
27009	29/09/2018	15:31	-33.92266	139.29402	female?	305	575	1	1	0	-	-
41111	30/09/2018	10:33	-34.03645	139.29517	male	289	525	0	1	0	-	-
(duplicated)												
27070	30/09/2018	11:38	-34.03690	139.22453	female?	275	425	0	1	0	-	-
27071	30/09/2018	12:37	-34.01770	139.22050	male?	319	500	0	0	0	-	-
37072	30/09/2018	13:14	-34.00706	139.21974	female?	292	400	0	0	0	-	-
(duplicated)												

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
12298	30/09/2018	14:58	-33.88625	139.33797	female?	331	850	0	0	0	-	-
27056	30/09/2018	15:32	-33.88635	139.33644	female?	292	600	0	1	0	-	-
8949	30/09/2018	16:15	-33.89801	139.35617	male?	307	650	1	0	0	-	-
846	1/10/2018	9:28	-33.88760	139.31972	female?	325	650	0	0	0	-	-
27057	1/10/2018	10:20	-33.89031	139.34726	female?	321	600	1	0	0	-	-
27058	1/10/2018	11:44	-33.92815	139.28494	female?	297	550	0	1	0	-	-
10391	6/10/2018	14:34	-33.89412	139.34094	male	293	500	1	0	0	-	-
11622	6/10/2018	15:35	-33.88678	139.35533	female?	321	700	0	0	0	-	-
27072	7/10/2018	10:00	-33.86629	139.42618	female?	320	500	1	0	1	-	-
27073	7/10/2018	10:20	-33.86725	139.42615	male	312	600	1	0	0	-	-
27074	7/10/2018	10:45	-33.86725	139.42615	female?	305	700	1	0	0	-	-
27075	7/10/2018	10:50	-33.86725	139.42615	male?	320	625	1	0	0	-	-
27076	7/10/2018	11:20	-33.86856	139.42613	male	341	675	1	0	0	-	-
27077	7/10/2018	11:35	-33.87015	139.42603	female?	314	575	1	0	0	-	-
27078	7/10/2018	12:45	-33.88506	139.42532	female?	331	700	0	0	0	-	-
27079	7/10/2018	15:24	-33.89278	139.42503	female?	312	650	0	0	0	-	-
27080	7/10/2018	16:07	-33.92692	139.42566	female?	290	550	1	0	0	-	-
27081	7/10/2018	16:45	-33.90933	139.42403	female?	271	375	1	0	0	-	-
27082	7/10/2018	17:00	-33.90784	139.42412	male?	312	550	1	0	0	-	-
461	7/10/2018	18:20	-33.88432	139.35849	female?	303	550	1	0	0	-	-
27083	8/10/2018	9:38	-33.86562	139.42630	female?	302	525	1	0	0	-	-
2451	8/10/2018	10:20	-33.88950	139.42506	male	323	575	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27085	8/10/2018	10:40	-33.89933	139.42455	male	324	625	1	0	0	-	-
27084	8/10/2018	10:45	-33.90269	139.42447	male	315	500	1	0	0	-	-
27086	8/10/2018	11:35	-33.89378	139.42480	female?	309	575	0	0	0	-	-
7027	8/10/2018	12:00	-33.86535	139.42633	male	311	525	1	0	0	-	-
4516	8/10/2018	12:40	-33.86152	139.42645	female?	315	700	0	0	0	-	-
610	13/10/2018	14:25	-33.90513	139.32294	female?	304	535	0	0	0	-	-
27009	13/10/2018	16:04	-33.92237	139.29451	female?	301	575	1	0	0	-	-
11545	13/10/2018	16:31	-33.92032	139.29791	male	304	550	1	1	0	-	-
4803	13/10/2018	17:03	-33.91394	139.30832	female?	319	600	1	0	0	-	-
8807	13/10/2018	17:35	-33.90373	139.32507	female?	326	675	1	0	0	-	-
11701	13/10/2018	18:15	-33.88770	139.35152	male	332	775	0	0	0	-	-
41827	14/10/2018	9:05	-33.88829	139.31215		177	97	0	0	0	-	-
41840	14/10/2018	10:40	-33.88547	139.31035		284	444	0	0	0	-	-
12723	14/10/2018	11:00	-33.88794	139.31572	male	306	544	1	1	0	-	-
27059	14/10/2018	11:30	-33.88783	139.31633	female?	275	325	1	1	0	-	-
4952	14/10/2018	15:40	-33.89307	139.34276	female?	305	600	0	0	0	-	-
8040	14/10/2018	16:13	-33.90407	139.32466	male	297	600	1	0	0	-	-
50935	14/10/2018	16:58	-33.91796	139.30168	male	314	650	1	1	0	-	-
11885	14/10/2018		-33.88839	139.31082	male	320	584	0	1	0	-	-
9372	14/10/2018		-33.88839	139.31082	male	312	582	0	1	0	-	-
27061	14/10/2018		-33.88715	139.31635	female?	300		0	0	0	-	-
27087	15/10/2018	10:47	-34.03814	139.32904	male	305	625	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27088	15/10/2018	11:25	-34.03639	139.31458	female?	293	500	0	1	0	-	-
27089	15/10/2018	11:45	-34.03643	139.31514	female?	334	650	0	1	0	-	-
27090	15/10/2018	12:00	-34.03644	139.31398	male	323	650	0	1	0	-	-
14750	15/10/2018	13:37	-33.92104	139.29668	female?	291	475	0	1	0	-	-
27060	20/10/2018	14:03	-33.88945	139.34859	female?	283	375	0	0	0	-	-
875	20/10/2018	14:40	-33.89401	139.34109	male	313	625	1	0	0	-	-
27062	20/10/2018	16:19	-33.97292	139.21729	female?	267	350	0	1	0	-	-
27091	21/10/2018	11:38	-34.07679	139.19922	female?	257	375	0	1	0	-	-
27092	21/10/2018	12:25	-34.09363	139.17078	female?	287	500	0	1	0	-	-
6451	22/10/2018	8:25	-33.89030	139.34666	female?	283	450	1	0	0	-	-
10391	22/10/2018	9:13	-33.89405	139.34102	male	294	500	0	0	0	-	-
1769	22/10/2018	9:33	-33.89934	139.33231	male	305	675	1	0	0	-	-
7097	22/10/2018	9:33	-33.89927	139.33241	female?	302	675	1	0	0	-	-
8012	22/10/2018	9:33	-33.89925	139.33263	female?	306	525	1	0	0	-	-
27093	28/10/2018	14:11	-34.03622	139.34077	female?	314	600	0	1	0	-	-
27094	28/10/2018	15:11	-34.03659	139.28075	female?	301	500	0	1	0	-	-
27095	28/10/2018	15:46	-34.03664	139.26997	female?	262	325	0	1	0	-	-
144000 special	28/10/2018	16:48	-34.06903	139.23355	male	288	400	0	1	0	-	-
27096	28/10/2018	17:55	-34.03630	139.31635	male	316	625	0	1	0	-	-
27064	29/10/2018	10:18	-33.89245	139.25371	female?	277	300	1	0	0	-	-
17099	29/10/2018	11:53	-33.89447	139.22757	female?	308	525	0	1	0	-	-
5034	29/10/2018	17:41	-33.90942	139.31572	female?	332	675	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
1190	30/10/2018	9:34	-33.90669	139.32033	female?	321	500	1	0	0	-	-
17169	30/10/2018	9:45	-33.90825	139.31769	female?	317	450	1	0	0	-	-
27097	2/11/2018	18:03	-34.06862	139.23183	female?	214	225	0	1	0	-	-
27098	2/11/2018	18:25	-34.06881	139.23248	male?	291	475	0	1	0	-	-
2771	3/11/2018	9:15	-33.93378	139.35695	male?	308	475	1	1	0	-	-
27099	3/11/2018	12:19	-34.03630	139.31909	male	310	550	0	1	0	-	-
27100	3/11/2018	12:19	-34.03630	139.31909	female	306	600	0	1	0	-	-
27063	9/11/2018	16:00	-33.88502	139.35411	male:?	211	250	1	0	0	-	-
854	9/11/2018	16:05	-33.88507	139.35330	female?	304	675	0	0	0	-	-
4997	9/11/2018	17:33	-33.88789	139.31567	male	294	600	1	1	0	-	-
27065	9/11/2018	18:21	-33.88681	139.33017	female?	279	500	1	0	0	-	-
27066	10/11/2018	9:30	-33.88528	139.35088	female?	261	350	0	0	0	-	-
659	10/11/2018	10:19	-33.88544	139.34834	male	312	700	1	0	0	-	-
27067	10/11/2018	11:04	-33.88758	139.31969	female?	298	575	1	1	0	-	-
41274	10/11/2018	11:40	-33.88782	139.31595	male	287	425	0	0	0	-	-
11690	10/11/2018	12:45	-33.89041	139.28171	male	304	625	1	1	0	-	-
2939	10/11/2018	13:51	-33.88594	139.34209	female?	295	600	0	0	0	-	-
534	10/11/2018	16:15	-33.91616	139.30467	male	325	625	1	0	0	-	-
27068	10/11/2018	16:20	-33.91575	139.30550	male?	251	350	1	0	0	-	-
3396	11/11/2018	9:16	-33.89819	139.33429	male	315	675	0	0	0	-	-
27069	11/11/2018	9:17	-33.89828	139.33431	female?	283	375	0	0	0	-	-
5350	11/11/2018	10:32	-33.93093	139.28040	female?	292	550	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
10452	11/11/2018	11:28	-33.92822	139.34590	female?	324	775	1	1	0	-	-
6166	11/11/2018	12:34	-33.90950	139.31583	male?	301	550	1	0	0	-	-
12298	17/11/2018	15:29	-33.88624	139.33833	male?	334	825	0	0	0	-	-
27101	17/11/2018	16:31	-33.89180	139.26318	male	303	625	1	0	0	-	-
14020	17/11/2018	17:13	-33.89214	139.25810	male	306	600	1	0	0	-	-
12296	17/11/2018	17:41	-33.89280	139.24956	female?	312	675	1	1	0	-	-
839	17/11/2018	18:26	-33.89283	139.24866	male?	306	700	1	0	0	-	-
878	18/11/2018	9:13	-33.88755	139.35141	female?	320	700	1	0	0	-	-
11701	18/11/2018	9:15	-33.88762	139.35150	male	335	775	0	0	0	-	-
8927	18/11/2018	10:20	-33.89251	139.34372	female?	308	700	1	0	0	-	-
2370	18/11/2018	10:21	-33.89262	139.34351	male	319	675	1	0	0	-	-
4952	18/11/2018	10:51	-33.89286	139.34305	female?	300	650	0	0	0	-	-
9910	18/11/2018	12:00	-33.90118	139.32928	female	321	700	1	0	0	-	-
609	18/11/2018	12:00	-33.90118	139.32928	male	315	700	1	0	0	-	-
27027	18/11/2018	13:20	-33.91394	139.30838	male	309	550	1	0	0	-	-
27102	18/11/2018	13:27	-33.91475	139.30707	female?	266	500	1	0	0	-	-
27103	18/11/2018	15:47	-33.94680	139.25531	female?	310	650	1	1	0	-	-
12043	23/11/2018	13:32	-33.96766	139.22369	female?	293	525	0	0	0	-	-
27104	23/11/2018	16:00	-33.95548	139.21710	female?	291	475	0	0	0	-	-
27105	23/11/2018	17:04	-33.89441	139.22769	male	276	575	0	1	0	-	-
12244	24/11/2018	10:12	-33.89363	139.27023	male?	294	675	0	1	0	-	-
8944	24/11/2018	11:27	-33.89428	139.27022	female?	309	675	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
14086	24/11/2018	12:01	-33.89707	139.27022	female?	308	750	1	1	0	-	-
8786	25/11/2018	9:15	-33.92029	139.29782	male	297	650	1	0	1	-	-
12153	30/11/2018	16:11	-33.88943	139.34869	male	320	700	1	0	0	-	-
12094	30/11/2018	17:08	-33.92193	139.29516	male	294	650	1	1	0	-	-
11460	1/12/2018	8:54	-33.88511	139.35323	male	304	575	1	0	0	-	-
27106	1/12/2018	9:28	-33.88514	139.35260	female?	286	575	1	0	0	-	-
763	1/12/2018	10:11	-33.88566	139.34544	female?	310	725	0	0	0	-	-
12889	1/12/2018	10:57	-33.88621	139.33800	female?	297	625	0	1	0	-	-
6451	1/12/2018	17:53	-33.89095	139.34608	male?	264	475	0	0	0	-	-
8373	1/12/2018	17:57	-33.89285	139.34302	male	323	675	0	0	0	-	-
8770	1/12/2018	18:06	-33.89323	139.34219	female?	328	825	0	0	0	-	-
8855	1/12/2018	18:07	-33.89322	139.34248	male?	313	575	0	0	0	-	-
17108	2/12/2018	10:09	-33.92834	139.24544	female?	291	625	0	1	0	-	-
27107	2/12/2018	10:46	-33.92070	139.21722	male	312	675	0	0	0	-	-
11656	2/12/2018	11:33	-33.89469	139.22336	female?	318	625	1	1	1	-	-
7349	2/12/2018	12:32	-33.89257	139.25243	female?	310	700	1	0	0	-	-
17159	2/12/2018	13:15	-33.89193	139.26158	female?	293	575	1	0	0	-	-
27108	2/12/2018	13:18	-33.89185	139.26253	male?	301	600	1	0	0	-	-
11798	2/12/2018	14:38	-33.88980	139.29002	male?	306	575	1	0	0	-	-
14111	2/12/2018	15:31	-33.88876	139.30386	male	333	625	1	0	0	-	-
27056	2/12/2018	16:23	-33.88633	139.33675	female?	294	650	0	1	0	-	-
11151	3/12/2018	9:53	-33.90408	139.32460	female?	307	750	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
1521	3/12/2018	10:26	-33.90946	139.31586	female?	321	575	1	0	0	-	-
8623	3/12/2018	11:17	-33.91834	139.30115	female?	325	725	1	1	0	-	-
11597	3/12/2018	11:52	-33.90282	139.32661	female?	311	700	1	1	0	-	-
8117	8/12/2018	8:17	-33.90480	139.32346	female	312	675	1	0	0	-	-
27109	8/12/2018	8:17	-33.90480	139.32346	male	309	525	0	0	0	-	-
27110	8/12/2018	9:20	-33.90944	139.31586	female	300	550	1	0	0	-	-
11584	8/12/2018	9:20	-33.90944	139.31586	male	316	550	1	0	0	-	-
5040	8/12/2018	10:25	-33.91311	139.30906	male?	309	650	0	0	0	-	-
27111	8/12/2018	11:33	-33.94623	139.25601	female?	288	550	0	1	0	-	-
17029	8/12/2018	16:07	-33.88749	139.32097	male?	312	600	0	0	0	-	-
10839	8/12/2018	16:43	-33.88764	139.31935	female?	324	875	0	0	0	-	-
12933	9/12/2018	10:10	-33.89407	139.23093	female?	299	650	0	1	0	-	-
12201	9/12/2018	10:52	-33.89226	139.25644	male	307	675	1	0	0	-	-
12656	9/12/2018	11:37	-33.89167	139.26430	female?	330	675	1	0	0	-	-
40745	16/12/2018	15:27	-33.88814	139.31085	female?	306	650	0	1	0	-	-
4579	16/12/2018	16:20	-33.89355	139.23914	male	308	625	1	0	0	-	-
11209 special	16/12/2018	17:10	-33.89314	139.24496	female?	306	725	0	0	0	-	-
4092	17/12/2018	10:27	-33.92835	139.27007	female?	314	725	0	1	0	-	-
1988	17/12/2018	11:00	-33.92818	139.28488	male	311	600	1	1	0	-	-
12364	17/12/2018	11:54	-33.90166	139.32869	female?	300	650	1	0	1	-	-
27112	22/12/2018	11:30	-33.95748	139.23911	male	306	575	0	1	0	-	-
27113	22/12/2018	12:35	-33.92117	139.29640	female?	296	550	1	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27114	22/12/2018	17:19	-33.90714	139.31956	male	301	700	1	0	0	-	-
3101	22/12/2018	17:58	-33.89354	139.34198	male?	328	850	0	0	0	-	-
27115	22/12/2018	18:35	-33.88565	139.34579	female?	280	500	0	1	0	-	-
12760	22/12/2018	18:37	-33.88561	139.34616	male?	309	625	1	1	0	-	-
673	23/12/2018	9:30	-33.88514	139.35283	female?	311	750	1	0	0	-	-
10562	23/12/2018	10:12	-33.88631	139.33728	male?	327	750	1	0	0	-	-
5040	23/12/2018	11:15	-33.91309	139.30972	male?	308	675	0	0	0	-	-
4827	23/12/2018	11:44	-33.91335	139.30940	female?	315	725	1	0	0	-	-
11480	5/01/2019	9:48	-33.88957	139.29337	male?	323	650	1	0	0	-	-
27116	5/01/2019	11:03	-33.88759	139.32001	male	301	550	1	0	0	-	-
12760	5/01/2019	11:42	-33.88575	139.34554	male?	309	600	0	0	0	-	-
12153	5/01/2019	17:05	-33.88956	139.34865	male	321	625	1	0	0	-	-
27117	5/01/2019	17:34	-33.89042	139.34695	female?	299	550	0	0	0	-	-
27118	6/01/2019	10:20	-33.92836	139.35429	female	317	525	1	0	0	-	-
10545	6/01/2019	10:20	-33.92836	139.35429	male	323	650	1	0	0	-	-
8117	6/01/2019	11:38	-33.90445	139.32391	female?	312	650	0	0	0	-	-
100	19/01/2019	12:45	-33.90443	139.32390	female	334	1000	1	0	0	-	-
11689	19/01/2019	13:45	-33.88583	139.35501	female	334	1000	1	0	0	-	-
12759	19/01/2019	13:45	-33.88589	139.35504	male	320	900	1	0	0	-	-
27119	9/02/2019	10:50	-34.07676	139.19202	male?	259	300	0	1	0	-	-
11955	9/02/2019	11:18	-33.92839	139.28473	male?	322	600	1	1	0	-	-
526	9/02/2019	14:20	-33.89025	139.34732	female?	303	675	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
4829	9/02/2019	14:45	-33.90872	139.31691	male	307	625	1	0	0	-	-
27120	9/02/2019	16:08	-34.06642	139.19992	female?	297	500	0	1	0	-	-
27121	10/02/2019	12:29	-33.89312	139.24504	female?	215	200	0	0	0	-	-
41401	10/02/2019	13:28	-33.88822	139.31081	male?	282	475	0	0	0	-	-
29122	16/02/2019	11:51	-34.11162	139.15598	male	285	475	0	1	0	-	-
2041	16/02/2019	13:15	-33.90864	139.31717	male	308	525	1	0	0	-	-
8743	16/02/2019	15:15	-33.89827	139.33408	male?	325	625	1	0	0	-	-
10930	17/02/2019	9:07	-33.88841	139.35031	male	314	600	1	0	0	-	-
12153	17/02/2019	9:30	-33.88945	139.34854	male	320	650	1	0	0	-	-
4908 special	17/02/2019	10:09	-33.89319	139.34242	male	298	550	1	0	0	-	-
2620	17/02/2019	10:46	-33.90598	139.32150	female?	320	650	1	0	0	-	-
11872	17/02/2019	11:29	-33.92946	139.28287	female?	284	625	0	0	0	-	1
7110	16/03/2019	17:34	-33.94334	139.26062	female?	291	400	0	1	0	-	-
27122	17/03/2019	10:10	-33.89021	139.34737	female?	296	600	1	0	0	-	-
3101	31/03/2019	15:43	-33.89365	139.34180	male?	327	625	0	0	0	-	-
12761	8/06/2019	12:45	-33.90057	139.33047	male	324	625	1	0	0	-	-
27022	8/06/2019	16:23	-33.88629	139.35521	female?	299	625	1	0	0	-	-
12501	8/06/2019	16:23	-33.88629	139.35521	male?	324	650	1	0	0	-	-
27123	8/06/2019	16:23	-33.88629	139.35521	female?	311	650	1	0	0	-	-
14611	17/08/2019	10:25	-33.91315	139.30978	female?	281	500	1	0	0	-	-
27124	17/08/2019	11:16	-33.90707	139.31955	female?	309	550	1	0	0	-	-
10658	17/08/2019	13:21	-33.88845	139.31065	male	298	525	0	1	0	1	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
10662	17/08/2019	14:15	-33.88984	139.28938	female?	317	600	1	0	0	-	-
14029	24/08/2019	9:09	-33.89507	139.33957	female?	302	460	1	0	0	-	-
47110	24/08/2019	11:28	-33.88776	139.31776	female?	265	400	1	0	0	-	-
17276	24/08/2019	12:16	-33.88691	139.32870	male	324	625	1	0	0	-	-
12889	24/08/2019	12:20	-33.88619	139.33871	female?	301	575	0	0	0	-	-
27125	30/08/2019	16:35	-33.88603	139.35507	female?	323	700	1	0	0	-	-
123	30/08/2019	17:47	-33.92809	139.34030	male	313	700	0	0	0	-	-
6137	30/08/2019	17:47	-33.92809	139.34030	female?	313	725	0	0	0	-	-
12129	31/08/2019	9:44	-33.92780	139.34055	female	314	650	0	0	0	-	-
27126	31/08/2019	9:44	-33.92780	139.34055	male	311	750	1	0	0	-	-
27127	31/08/2019	12:10	-33.88760	139.31938	juvenile	238	300	0	0	0	-	-
11485	31/08/2019	12:43	-33.88709	139.32576	female?	317	650	1	0	0	-	-
12298	31/08/2019	13:23	-33.88625	139.33813	male?	332	825	1	0	0	-	-
12138	31/08/2019	15:45	-33.88811	139.31224	female?	316	625	0	1	0	-	-
9390	31/08/2019	16:30	-33.88821	139.30977	male?	318	750	1	1	0	-	-
2686	1/09/2019	9:36	-33.91874	139.30063	female?	307	650	1	0	0	-	-
27128	1/09/2019	10:25	-33.94599	139.25600	female?	302	575	0	1	0	-	-
27129	1/09/2019	11:05	-33.94617	139.25616	male?	303	575	0	1	0	-	-
27012	1/09/2019	11:55	-33.95166	139.24800	male?	296	550	0	1	0	-	-
26000	7/09/2019	9:25	-33.95288	139.24599	female?	310	600	0	0	0	-	-
27130	7/09/2019	10:10	-33.95943	139.22981	female?	293	400	0	0	0	-	-
17183	7/09/2019	14:30	-33.90100	139.32967	female?	321		1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27131	8/09/2019	11:38	-33.88633	139.33644	male?	308	600	0	1	0	-	-
877	8/09/2019	12:25	-33.88870	139.30496	female?	294	550	0	0	0	-	-
41722	13/09/2019	14:50	-33.88781	139.31595	female?	270	325	0	0	0	-	-
27053	13/09/2019	14:55	-33.88781	139.31604	male?	271	375	1	1	0	-	-
41207	13/09/2019	16:15	-33.88836	139.31049	female?	306	625	0	1	0	-	-
31800	14/09/2019	10:35	-33.88980	139.28983	male?	301	525	1	0	0	-	-
607	14/09/2019	11:25	-33.89028	139.28378	male?	300	450	1	1	0	-	-
822	14/09/2019	12:15	-33.89157	139.26630	female?	298	550	1	1	0	-	-
27132	14/09/2019	12:52	-33.89178	139.26294	female	306	550	1	0	0	-	-
7287	14/09/2019	13:45	-33.89427	139.22952	female?	303	575	0	0	0	-	-
10581	14/09/2019	14:10	-33.89423	139.23053	male?	310	600	1	0	0	-	-
6281 special	14/09/2019	14:56	-33.89445	139.22679	male?	302	550	0	1	0	-	-
27133	14/09/2019	15:38	-33.90330	139.21713	female?	281	500	0	1	0	-	-
27134	14/09/2019	16:57	-33.88646	139.33499	male?	322	750	1	0	0	-	-
10930	15/09/2019	9:15	-33.88516	139.35086	male?	313	575	1	0	0	-	-
31194?	15/09/2019	9:48	-33.88788	139.31561	female?	302	525	0	1	0	-	-
41274	15/09/2019	9:50	-33.88793	139.31580	female?	284	350	0	0	0	-	-
11480	15/09/2019	11:11	-33.88955	139.29333	male?	320	550	1	0	0	-	-
27019	21/09/2019	13:23	-33.88584	139.34706	female?	279	500	1	0	0	-	-
27065	21/09/2019	14:08	-33.88680	139.33081	female?	279	450	1	0	0	-	-
10839	21/09/2019	14:45	-33.88763	139.31946	female?	327	850	1	0	0	-	-
4952	22/09/2019	9:23	-33.89307	139.34261	female?	304	650	0	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27138	22/09/2019	10:21	-33.94607	139.25598	unknown	313	500	0	1	0	-	-
3381	22/09/2019	11:25	-33.95586	139.24147	male?	297	550	0	0	0	-	-
27135	22/09/2019	12:15	-33.95961	139.23582	female?	282	425	0	1	0	-	-
11656	22/09/2019	13:52	-33.89468	139.22348	female?	314	600	1	0	0	-	-
34402	22/09/2019	14:30	-33.89161	139.26633	female?	296	700	0	0	0	-	-
12855	23/09/2019	9:55	-33.91343	139.27034	male	308	650	0	1	0	-	-
27136	23/09/2019	9:56	-33.91343	139.27034	female	315	675	0	1	0	-	-
5690	23/09/2019	11:28	-33.89704	139.27042	male?	320	700	1	1	0	-	-
41747	23/09/2019	12:22	-33.88863	139.31099	juvenile	236	275	0	1	0	-	-
9372	23/09/2019	13:05	-33.88832	139.31128	male	315	575	1	1	0	-	-
2536	27/09/2019	17:15	-33.89812	139.42470	female	334	800	1	0	0	-	-
27137	28/09/2019	10:20	-33.88869	139.30592	male	316	750	1	0	0	-	-
40018	28/09/2019	11:21	-33.88803	139.31468	male	291	550	0	0	0	-	-
27139	5/10/2019	10:20	-33.92802	139.42682	female	302	700	1	0	0	-	-
27140	5/10/2019	16:15	-33.92641	139.43799	female	328	725	1	0	0	-	-
11900	6/10/2019	9:50	-33.92816	139.40091	female	318	600	1	0	0	-	-
27144	11/10/2019	11:20	-34.09177	139.17429	unknown	274	325	0	1	1	-	-
27142	11/10/2019	12:20	-33.96616	139.22601	female	251	300	0	0	0	-	-
27147	11/10/2019	12:20	-33.96616	139.22601	male	293	500	0	0	0	-	-
27063	11/10/2019	15:58	-33.88505	139.35391	juvenile	230	275	1	0	0	-	-
4876	11/10/2019	16:35	-33.88509	139.35335	female?	315	700	1	0	0	-	-
24190	11/10/2019	17:17	-33.88705	139.32718	male	314	625	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27143	12/10/2019	12:00	-33.92533	139.21712	male	301	600	0	1	0	-	-
27146	12/10/2019	12:50	-33.91323	139.21718	male?	278	450	0	1	0	-	-
27145	12/10/2019	12:50	-33.91323	139.21718	female?	258	425	0	1	0	-	-
12424	12/10/2019	14:24	-33.89410	139.23186	male	301	625	1	0	0	-	-
686	12/10/2019	14:24	-33.89410	139.23186	female	281	550	1	0	0	-	-
27148	12/10/2019	15:50	-33.89259	139.25217	male	274	425	1	0	0	-	-
11102	13/10/2019	10:38	-33.89002	139.28699	male	321	550	1	1	0	-	-
#01	17/10/2019	12:25	-34.01795	139.22052		289	550	0	1	0	-	-
#02	17/10/2019	14:10	-33.92792	139.42682	male	317	750	1	0	0	-	-
#03	17/10/2019	14:49	-33.95459	139.42638	female?	280	450	1	0	0	-	-
#04	17/10/2019	15:24	-33.97093	139.41905	female?	325	700	0	1	0	-	-
#05	17/10/2019	16:00	-33.97126	139.38280	male	330	700	0	1	0	-	-
#06	17/10/2019	16:45	-33.97134	139.37149	female?	313	550	0	1	0	-	-
27149	17/10/2019	18:28	-33.88587	139.35529	juvenile	202	200	1	0	0	-	-
#07	18/10/2019	10:20	-34.08719	139.18045	female?	275	475	0	1	0	-	-
#08	18/10/2019	11:20	-34.15963	139.08852	female?	257	350	0	0	0	-	-
#09	18/10/2019	14:20	-34.17785	139.09116		249	350	0	0	0	-	-
27150	25/10/2019	14:00	-33.88599	139.35510	juvenile	207	225	0	0	0	-	-
27151	25/10/2019	15:15	-33.88602	139.35509	juvenile	207	225	0	0	0	-	-
27152	26/10/2019	10:40	-33.89685	139.42465	male	300	625	1	1	0	-	-
27153	26/10/2019	11:45	-33.89133	139.42493	male?	287	550	1	0	0	-	-
27154	26/10/2019	12:39	-33.87741	139.42564	male	316	625	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27155	26/10/2019	15:21	-33.88860	139.42505	female?	310	575	1	0	0	-	-
27157	26/10/2019	16:21	-33.88390	139.42531		269	375	0	0	1	-	-
17293	2/11/2019	14:45	-33.88676	139.35298	female?	308	675	1	0	0	-	-
775	2/11/2019	15:26	-33.88987	139.34785	male	323	675	0	0	0	-	-
27156	2/11/2019	16:14	-33.89602	139.33791	male?	294	500	1	0	0	-	-
#10	3/11/2019	16:30	-34.15380	139.12439	male	285	475	0	1	0	-	-
4964	3/11/2019	18:00	-33.95561	139.24188	male	300	550	0	1	0	-	-
27158	4/11/2019	10:35	-33.92699	139.37273	female?	298	500	1	0	0	-	-
40722	4/11/2019	12:39	-33.88789	139.31564	male	285	550	1	0	0	-	-
27106	8/11/2019	14:25	-33.88512	139.35225	female	292	600	1	0	0	-	-
28104	8/11/2019	15:07	-33.88605	139.34021	male?	322	750	0	0	0	-	-
12737	9/11/2019	11:11	-33.88652	139.33401	male	304	625	0	0	0	-	-
846	9/11/2019	11:54	-33.88763	139.31935	female?	325	700	0	0	0	-	-
41204	9/11/2019	12:39	-33.88786	139.31548	female?	309	675	1	1	0	-	-
11885	9/11/2019	13:20	-33.88857	139.31099	male	316	750	1	1	0	-	-
12847	9/11/2019	14:00	-33.88857	139.31099	female?	301	650	1	1	0	-	-
11505	9/11/2019	15:50	-33.88877	139.31081	male	317	725	0	0	0	-	-
12935	9/11/2019	16:47	-33.88920	139.29756	male	325	700	1	0	0	-	-
41274	10/11/2019	10:13	-33.88779	139.31651	female?	297	525	1	0	0	-	-
11819	10/11/2019	11:22	-33.88943	139.29488	female?	318	725	1	0	0	-	-
27159	10/11/2019	12:12	-33.88639	139.33571	male	291	575	1	0	0	-	-
881	15/11/2019	15:46	-33.88608	139.34019	male	296	575	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
27160	15/11/2019	15:46	-33.88608	139.34019	female	303	625	0	0	0	-	-
17127	15/11/2019	15:59	-33.88666	139.33180	female?	307	725	1	1	0	-	-
40077	15/11/2019	16:37	-33.88786	139.31589	female?	306	625	0	1	0	-	-
32727	15/11/2019	17:25	-33.88683	139.32970	female?	309	700	1	0	0	-	-
9372	16/11/2019	13:10	-33.88820	139.31165	male	319	675	0	1	0	-	-
1588	16/11/2019	14:18	-33.92164	139.29570	male	315	600	1	1	0	-	-
120833 special	16/11/2019	15:32	-33.88861	139.31113	male	305	600	0	0	0	-	-
17125	16/11/2019	16:22	-33.88668	139.33168	male	321	850	1	0	0	-	-
12754	16/11/2019	16:58	-33.88597	139.34163	female?	308	700	1	0	0	-	-
3024	17/11/2019	10:18	-33.93129	139.21712	female?	292	550	0	0	0	-	-
8775 special	17/11/2019	10:55	-33.92812	139.21712	female?	289	575	0	1	1	-	-
5444	17/11/2019	12:01	-33.89422	139.23048	male	307	675	0	1	0	-	-
7455	17/11/2019	12:42	-33.89218	139.25751	female?	316	550	1	0	0	-	-
27052	17/11/2019	13:36	-33.88976	139.29018	male?	302	575	1	0	0	-	-
27161	17/11/2019	14:34	-33.88615	139.33916	female?	309	700	0	0	0	-	-
27162	18/11/2019	9:20	-33.94603	139.25595	female?	299	575	0	1	0	-	-
7906	18/11/2019	9:22	-33.94599	139.25601	female?	311	700	0	1	0	-	-
5231	18/11/2019	9:23	-33.94606	139.25606	male	297	550	0	1	0	-	-
6186	18/11/2019	10:55	-33.94630	139.25623	male?	310	575	0	1	0	-	-
10658	18/11/2019	12:21	-33.88840	139.31088	male	304	600	0	0	0	-	-
27163	23/11/2019	12:15	-33.92830	139.28366	female?	297	600	1	1	0	-	-
27164	23/11/2019	13:23	-33.97145	139.31032	male	313	550	0	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
7754	23/11/2019	17:42	-33.97427	139.35658	male?	301	550	0	1	0	-	-
27165	30/11/2019	12:52	-33.96856	139.22243	female?	288	475	0	0	0	-	-
27166	30/11/2019	13:27	-33.96736	139.22417	female?	287	375	0	0	1	-	-
14367	30/11/2019	14:42	-33.92202	139.29514	female?	299		0	1	0	-	-
14426	30/11/2019	15:20	-33.91044	139.31407	female?	318	600	1	0	0	-	-
27167	30/11/2019	16:16	-33.89529	139.33902	female?	273	450	1	1	0	-	-
854	1/12/2019	10:33	-33.88514	139.35291	female?	315	725	1	0	0	-	-
27168	6/12/2019	17:35	-33.95973	139.23570	female?	264	400	0	0	0	-	-
4796	6/12/2019	18:24	-33.91287	139.31013	female?	320	500	1	0	0	-	-
6103	6/12/2019	18:33	-33.90305	139.32622	male?	317	650	1	0	0	-	-
12153	6/12/2019	18:42	-33.88945	139.34854	female?	316	500	1	0	0	-	-
17017	7/12/2019	11:24	-33.89154	139.26587	male?	317	650	1	0	0	-	-
10447	7/12/2019	12:36	-33.90027	139.33093	male	314	650	1	0	0	-	-
1041	7/12/2019	18:52	-33.88772	139.31726	female?	332	800	0	0	0	-	-
4831	8/12/2019	8:33	-33.89368	139.34177	male?	305	500	0	0	0	-	-
14729	8/12/2019	9:15	-33.89495	139.33955	male?	302	575	1	0	0	-	-
17391	8/12/2019	9:51	-33.89895	139.33308	female?	285	525	1	0	0	-	-
11572	8/12/2019	10:30	-33.90078	139.33011	female?	322	775	1	0	0	-	-
27169	8/12/2019	11:26	-33.95249	139.24664	female?	281	525	0	1	0	-	-
12153	15/12/2019	8:30	-33.88943	139.34857	female?	323	500	1	0	0	-	-
8012	15/12/2019	9:15	-33.89846	139.33379	female?	309	575	1	0	0	-	-
5845	15/12/2019	10:00	-33.90018	139.33090	female?	312	875	1	0	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	Sex	SVL	BM	<i>Amblyomma</i>	<i>Bothriocroton</i>	<i>Ornithodoros</i>	<i>Ophionyssus</i>	Chigger
								<i>limbatum</i>	<i>hydrosauri</i>	<i>gurneyi</i>	<i>natricis</i>	
11545	15/12/2019	15:39	-33.91998	139.29729	male	304	550	0	1	0	-	-
27170	16/12/2019	9:21	-33.89156	139.26642	male?	320	625	0	0	0	-	-
27171	16/12/2019	10:14	-33.94204	139.21722	female?	298	475	0	1	1	-	-
4880	16/12/2019	11:30	-33.91996	139.29729	female?	307	750	1	1	0	-	-

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros</i>	<i>Oochoristica</i>	<i>Hemolivia</i>	IIT 1	IIT 2	IIT 3	Note
					<i>trachysauri</i>	<i>trachysauri</i>	<i>mariae</i>				
623	31/08/2017	13:13	-33.91101	139.35620	0	-	0	-	-	1	WG*
17904	31/08/2017	14:52	-33.90908	139.31631	-	-	-	-	-	-	
8040	1/09/2017	12:20	-33.90418	139.32449	-	-	-	-	-	-	
9389	1/09/2017	14:35	-33.92007	139.35612	-	-	-	-	-	-	
9373	1/09/2017	14:35	-33.92007	139.35612	-	-	-	-	-	-	
17442	4/09/2017	15:45	-33.92007	139.35612	-	-	-	-	-	-	
9373	6/09/2017	12:43	-33.92038	139.35619	-	-	-	-	-	-	
5463	6/09/2017	13:10	-33.99239	139.35664	1	-	-	-	-	-	
12962	13/09/2017	12:30	-33.89448	139.35608	-	-	0	1	0	1	G
501	13/09/2017	12:35	-34.11604	139.35690	-	-	-	-	-	-	
27000	13/09/2017	14:23	-33.89598	139.35616	-	-	0	1	1	1	G*
8506	14/09/2017	13:42	-33.91063	139.31390	1	-	-	-	-	-	
10392	14/09/2017	13:42	-33.91063	139.31390	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27001	15/09/2017	9:50	-33.88664	139.35323	-	-	0	1	1	1	G*
11701	15/09/2017	12:00	-33.88707	139.35251	1	-	-	-	-	-	
8973	19/09/2017	14:10	-33.90754	139.35620	-	-	0	1	0	0	G
27003	19/09/2017	15:39	-33.92817	139.30365	-	-	0	1	0	0	WG*
27002	19/09/2017	15:39	-33.92817	139.30365	1	-	0	1	1	0	G*
27004	20/09/2017	11:00	-33.92824	139.30377	-	-	0	-	-	0	WG*
14937	20/09/2017	11:54	-33.95774	139.23865	-	-	-	-	-	-	
12749	25/09/2017	13:40	-33.88597	139.35535	-	-	-	-	-	-	
10773	25/09/2017	13:50	-33.88596	139.35506	-	-	-	-	-	-	
854B	25/09/2017	14:53	-33.88598	139.35448	-	-	0	1	1	1	G*
11670	25/09/2017	15:18	-33.88818	139.35075	-	-	0	-	-	0	G*
4054	25/09/2017	16:03	-33.92031	139.29790	-	-	-	-	-	-	
27005	26/09/2017	12:15	-34.04074	139.35666	-	-	0	1	0	1	G*
27006	26/09/2017	12:45	-33.99982	139.35658	-	-	0	1	1	0	G*
27007	26/09/2017	13:05	-33.99848	139.35664	-	-	0	1	0	0	G
2864	26/09/2017	13:45	-33.96051	139.35632	-	-	-	-	-	-	
11500	26/09/2017	14:10	-33.93203	139.35629	-	-	0	1	0	0	G
11829	26/09/2017	15:55	-33.89765	139.33521	-	-	0	-	-	0	G
8012	27/09/2017	10:40	-33.89876	139.33330	1	-	0	1	0	1	G*
8743	27/09/2017	10:40	-33.89876	139.33330	-	-	-	-	-	-	
1595	27/09/2017	10:42	-33.89867	139.33308	-	-	0	1	0	0	WG
12225	27/09/2017	11:40	-33.90499	139.32306	-	-	0	-	-	0	G

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27008	27/09/2017	11:45	-33.90571	139.32195	-	-	-	-	-	-	
2726	27/09/2017	12:30	-33.89570	139.33847	1	-	0	1	0	1	G*
11051	27/09/2017	12:45	-33.89383	139.34131	1	-	0	1	0	1	G
3787	1/10/2017	16:55	-34.06472	139.35670	-	-	0	-	-	0	G
17067	2/10/2017	11:00	-33.96201	139.23235	-	-	0	1	1	0	WG
27018	2/10/2017	11:33	-33.96750	139.22385	-	-	-	-	-	-	
1100	2/10/2017	11:33	-33.96750	139.22385	0	-	-	-	-	-	
27009	2/10/2017	13:38	-33.92201	139.29503	-	-	0	1	0	1	WG*
11256	2/10/2017	13:38	-33.92201	139.29503	1	-	-	-	-	-	
29000	2/10/2017	13:45	-33.96818	139.22295	-	-	0	1	0	1	WG
525	2/10/2017	17:41	-33.88966	139.34819	-	-	0	-	-	0	G*
1525	3/10/2017	9:38	-33.89817	139.33453	-	-	-	-	-	-	
14160	3/10/2017	9:38	-33.89817	139.33453	1	-	-	-	-	-	
1609	3/10/2017	10:40	-33.90333	139.32593	-	-	0	1	0	0	G
4855	3/10/2017	11:10	-33.90834	139.31758	-	-	-	-	-	-	
27010	3/10/2017	11:37	-33.90546	139.32239	-	-	0	1	1	1	G
27011	16/10/2017	11:23	-33.90189	139.35617	-	-	0	0	1	1	WG
17153	16/10/2017	11:25	-33.90213	139.35619	1	-	-	-	-	-	
27012	16/10/2017	14:33	-33.95138	139.24835	-	-	0	1	0	1	G*
27014	16/10/2017	16:42	-33.92823	139.31145	-	-	0	-	-	1	WG
27013	16/10/2017	17:10	-33.92822	139.30217	1	-	-	-	-	-	
27020	17/10/2017	9:03	-33.88656	139.35335	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
2010	17/10/2017	9:23	-33.88834	139.35046	-	-	0	-	-	0	G*
5530	17/10/2017	10:00	-33.89622	139.33742	-	-	-	-	-	-	
5561	17/10/2017	10:38	-33.89821	139.33432	1	-	-	-	-	-	
2342	17/10/2017	10:38	-33.89821	139.33432	1	-	-	-	-	-	
27015	17/10/2017	10:42	-33.89801	139.33463	1	-	0	1	1	1	WG
27016a	17/10/2017	12:33	-33.90925	139.31607	-	-	0	1	1	0	WG
27016b	24/10/2017	11:33	-33.96729	139.22433	-	-	0	1	1	1	WG
27017	24/10/2017	11:35	-33.88680	139.33049	-	-	0	1	0	1	WG
27019	24/10/2017	14:51	-33.88984	139.34793	-	-	0	1	1	0	G*
5561	24/10/2017	15:38	-33.89792	139.33475	-	-	0	1	1	0	G
2342	24/10/2017	15:38	-33.89792	139.33475	-	-	0	-	-	0	G
27021	24/10/2017	16:30	-33.93909	139.26704	-	1	0	-	-	0	G*
17164	25/10/2017	10:42	-33.88599	139.35530	-	-	0	-	-	1	WG
27022	25/10/2017	10:42	-33.88599	139.35530	-	-	-	-	-	-	
27024	25/10/2017	10:42	-33.88599	139.35530	-	-	-	-	-	-	
27023	25/10/2017	10:42	-33.88599	139.35530	0	-	-	-	-	-	
27001	30/10/2017	13:07	-33.88699	139.35271	-	-	0	1	1	1	G
12331	30/10/2017	13:29	-33.88793	139.35106	-	-	-	-	-	-	
11833	30/10/2017	14:11	-33.91151	139.31244	-	-	0	1	0	1	WG
27025	30/10/2017	15:14	-33.95469	139.24330	-	-	-	-	-	-	
553	30/10/2017	15:52	-33.97160	139.22134	-	-	-	-	-	-	
7655	30/10/2017	16:26	-33.95656	139.24039	-	-	0	1	1	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27026	31/10/2017	11:18	-33.96742	139.22385	1	-	0	1	1	1	WG
27027	31/10/2017	12:18	-33.91376	139.30862	-	-	0	1	1	1	WG
10860	5/11/2017	14:35	-33.90684	139.32005	-	-	0	1	0	0	G
826	5/11/2017	14:35	-33.90684	139.32005	-	-	0	-	-	1	G
27026	5/11/2017	16:00	-33.96700	139.22469	1	-	-	-	-	-	
11592	5/11/2017	16:50	-33.89764	139.33516	-	-	0	1	0	0	G
27028	6/11/2017	18:28	-33.99656	139.35664	-	-	0	-	-	0	WG
837	7/11/2017	13:23	-33.95368	139.24469	-	-	-	-	-	-	
528	7/11/2017	14:15	-33.91185	139.31194	-	-	0	0	1	0	WG*
27029	7/11/2017	15:05	-33.88552	139.35510	-	-	-	-	-	-	
2208	7/11/2017	16:55	-33.97150	139.27895	-	-	0	-	-	1	WG
10692	7/11/2017	18:02	-33.92822	139.29881	1	-	0	1	1	1	WG
11391 special	8/11/2017	10:10	-33.89453	139.34019	-	-	0	-	-	1	WG
11256	8/11/2017	11:08	-33.92200	139.29518	-	-	0	1	1	1	WG
27030	8/11/2017	12:00	-33.95950	139.23596	-	-	-	-	-	-	
27031	8/11/2017	12:50	-33.90360	139.32536	-	-	-	-	-	-	
12853	13/11/2017	17:26	-33.93372	139.35625	-	-	-	-	-	-	
10508	13/11/2017	18:14	-33.92822	139.30714	-	-	0	-	-	0	WG*
525	14/11/2017	8:31	-33.88966	139.34824	-	-	-	-	-	-	
8855	14/11/2017	9:02	-33.89320	139.34254	-	-	0	-	-	0	WG*
4952	14/11/2017	9:02	-33.89320	139.34254	1	-	-	-	-	-	
8707	14/11/2017	15:13	-33.90332	139.32571	-	-	0	-	-	0	WG*

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
14426	20/11/2017	15:01	-33.91010	139.31442	1	-	0	-	-	0	WG*
5530	20/11/2017	17:53	-33.89666	139.33664	1	-	0	-	-	0	WG*
3053	20/11/2017	18:23	-33.89216	139.34412	-	-	0	-	-	0	WG*
854	21/11/2017	8:35	-33.88604	139.35403	-	-	0	0	1	1	WG*
854B	21/11/2017	9:02	-33.88599	139.35446	1	-	0	1	1	1	WG
17183	21/11/2017	10:09	-33.90087	139.32990	-	-	-	-	-	-	
12795	21/11/2017	10:50	-33.91385	139.30867	-	-	0	1	0	0	WG
27032	21/11/2017	12:21	-33.97142	139.33258	-	-	-	-	-	-	
2728	21/11/2017	17:55	-33.88682	139.32999	-	-	0	-	-	0	WG*
27033	21/11/2017	18:33	-33.88984	139.29002	-	-	-	-	-	-	
17011	22/11/2017	8:58	-33.88640	139.35353	1	-	-	-	-	-	
27034	22/11/2017	9:25	-33.88678	139.35304	1	-	-	-	-	-	
9910	22/11/2017	10:10	-33.90113	139.32951	-	-	0	-	-	0	WG*
27035	22/11/2017	10:55	-33.93241	139.27808	1	-	0	-	-	0	WG*
10075	27/11/2017	9:48	-33.97163	139.26550	-	-	-	-	-	-	
1186 special	27/11/2017	10:40	-33.97156	139.27986	-	-	0	1	1	0	G
12458	27/11/2017	11:20	-33.97152	139.27592	1	-	0	1	1	0	WG
27037	27/11/2017	12:32	-33.91307	139.30978	1	-	-	-	-	-	
8878	27/11/2017	13:00	-33.91044	139.31406	1	1	-	-	-	-	
12364	27/11/2017	13:40	-33.90088	139.32980	1	-	0	-	-	0	WG*
17530	27/11/2017	17:30	-33.97165	139.31567	1	-	-	-	-	-	
11460	28/11/2017	9:23	-33.88509	139.35341	1	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
Stumpy	28/11/2017	9:23	-33.88507	139.35358	1	-	-	-	-	-	
27038	28/11/2017	10:28	-33.88509	139.35323	1	-	0	-	-	0	WG*
27039	28/11/2017	11:15	-33.88997	139.28804	1	-	-	-	-	-	
8177	3/12/2017	16:12	-34.00756	139.35664	-	-	0	-	-	0	WG*
11284	4/12/2017	9:12	-33.92826	139.29788	-	-	-	-	-	-	
17137	4/12/2017	10:53	-33.97152	139.24487	-	-	0	-	-	1	WG
2391	4/12/2017	11:55	-33.92107	139.29655	-	-	0	1	1	0	WG
7511	4/12/2017	12:24	-33.92067	139.29726	-	-	-	-	-	-	
10844	4/12/2017	13:09	-33.90141	139.32896	-	-	-	-	-	-	
3053	4/12/2017	13:52	-33.89279	139.34323	-	-	0	-	-	0	WG*
10773	4/12/2017	14:20	-33.88605	139.35432	-	-	0	-	-	0	G
12749	4/12/2017	14:45	-33.88609	139.35522	-	-	0	1	1	0	G
12860	4/12/2017	16:00	-33.88680	139.35529	-	-	-	-	-	-	
27040	4/12/2017	17:10	-33.91686	139.27037	-	-	0	-	-	0	G*
555	5/12/2017	9:55	-33.97171	139.23102	-	-	-	-	-	-	
12442	5/12/2017	10:23	-33.97165	139.23404	-	-	-	-	-	-	
11644	5/12/2017	10:51	-33.97160	139.23033	-	-	0	-	-	0	WG
27041	5/12/2017	11:23	-33.96433	139.22884	-	-	-	-	-	-	
27042	5/12/2017	12:17	-33.94436	139.25891	-	-	-	-	-	-	
6027	5/12/2017	12:54	-33.93084	139.28072	1	-	0	-	-	0	WG*
5034	5/12/2017	13:38	-33.90923	139.31609	-	-	0	1	0	1	WG*
41804	11/12/2017	11:50	-33.88768	139.30411	1	-	-	-	-	-	missing

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
14533	11/12/2017	16:54	-33.90125	139.27026	-	-	0	0	1	0	WG*
27043	11/12/2017	17:20	-33.89796	139.27016	1	-	0	1	0	1	WG
5692	11/12/2017	18:05	-33.89706	139.27026	1	1	-	-	-	-	
4870	11/12/2017	18:58	-33.90958	139.31564	-	-	-	-	-	-	
27044	11/03/2018	11:22	-33.93372	139.35626	1	-	0	-	-	0	WG*
8567	11/03/2018	11:22	-33.93372	139.35626	-	-	0	1	0	0	WG*
27045	11/03/2018	17:45	-33.89696	139.35619	-	-	0	1	0	0	WG
27046	23/03/2018	13:20	-34.01046	139.35664	-	-	0	1	1	0	WG
1922	24/03/2018	11:40	-33.98855	139.35664	-	-	0	0	1	1	WG
17220	15/04/2018	13:32	-33.88531	139.35068	-	-	0	-	-	1	WG
14611	20/05/2018	13:14	-33.91306	139.30994	-	-	0	1	1	0	WG
10510	13/08/2018	12:15	-33.88736	139.32291	-	-	0	1	1	1	WG
9367	13/08/2018	12:56	-33.88811	139.31337	1	-	-	-	-	-	
12598	13/08/2018	13:29	-33.88889	139.30186	-	-	-	-	-	-	
27047	27/08/2018	12:30	-33.89104	139.27368	-	-	0	1	1	0	WG
9511	27/08/2018	13:19	-33.89066	139.27916	-	-	-	-	-	-	
876	27/08/2018	13:45	-33.89009	139.28583	-	-	0	-	-	1	WG
14138	1/09/2018	13:12	-33.89444	139.22823	-	-	0	-	-	1	WG
10839	1/09/2018	13:37	-33.88756	139.31955	-	-	0	-	-	0	WG
27048	2/09/2018	12:00	-33.88625	139.35515	-	-	0	1	1	1	WG
27049	2/09/2018	12:57	-33.88654	139.33377	-	-	-	-	-	-	
11833	2/09/2018	14:27	-33.91184	139.31194	-	-	0	1	0	1	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
1000	9/09/2018	13:07	-33.88696	139.35272	-	-	0	1	1	0	WG
4876	9/09/2018	18:07	-33.88655	139.35347	-	-	0	1	1	0	WG
11701	9/09/2018	18:07	-33.88655	139.35347	1	-	1	1	1	1	WG
9910	9/09/2018	14:12	-33.90109	139.32951	-	-	-	-	-	-	
854B	9/09/2018	12:25	-33.88590	139.35439	-	-	0	1	1	1	WG
3101	9/09/2018	16:39	-33.89359	139.34192	-	-	0	-	-	0	WG
5040	9/09/2018	15:42	-33.91272	139.31053	1	-	-	-	-	-	
22849 special	9/09/2018	13:45	-33.89744	139.33543	-	-	0	1	1	1	WG
11833	9/09/2018	14:59	-33.91175	139.31221	-	-	0	1	0	0	WG
12606	10/09/2018	12:05	-33.89136	139.26961	-	-	0	-	-	1	WG
27051	10/09/2018	13:39	-33.88876	139.30437	1	-	0	1	0	1	WG
27050	10/09/2018	12:46	-33.88923	139.29744	-	-	-	-	-	-	
27727	15/09/2018	13:07	-33.88587	139.34320	-	-	0	1	1	1	WG
27052	15/09/2018	14:09	-33.88974	139.29060	-	-	0	-	-	0	WG
11612	15/09/2018	15:21	-33.89297	139.27023	-	-	0	-	-	1	WG
17013	15/09/2018	12:25	-33.88565	139.35506	-	-	0	-	-	1	WG
876	15/09/2018	14:46	-33.89009	139.28581	-	-	0	-	-	0	WG
7839	16/09/2018	10:08	-33.90083	139.33002	-	-	0	-	-	0	WG
27053	16/09/2018	14:05	-33.88786	139.31584	1	-	0	-	-	0	WG
4880	16/09/2018	12:15	-33.91988	139.29860	-	-	-	-	-	-	
5820	16/09/2018	11:27	-33.91213	139.31133	-	-	0	-	-	0	WG
6103	16/09/2018	10:54	-33.90280	139.32658	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
12456	16/09/2018	12:57	-33.92239	139.29449	-	-	0	-	-	0	WG
11724	23/09/2018	15:47	-33.89076	139.27579	-	-	0	-	-	0	WG
27054	24/09/2018	11:24	-33.88546	139.34825	1	-	0	1	1	1	WG
32494 special	24/09/2018	10:32	-33.88792	139.31583	-	-	0	1	0	0	WG
27055	29/09/2018	13:07	-33.89168	139.34492	-	-	0	1	1	1	WG
8743	29/09/2018	14:27	-33.89852	139.33365	-	-	0	1	1	1	WG
27009	29/09/2018	15:31	-33.92266	139.29402	-	-	0	1	1	1	WG
8770	29/09/2018	13:46	-33.89260	139.34341	-	-	0	1	1	1	WG
37072	30/09/2018	13:14	-34.00706	139.21974	-	-	-	-	-	-	
(duplicated)											
8949	30/09/2018	16:15	-33.89801	139.35617	-	-	0	1	1	1	WG
41111	30/09/2018	10:33	-34.03645	139.29517	-	-	1	0	1	1	WG
(duplicated)											
27056	30/09/2018	15:32	-33.88635	139.33644	-	-	0	-	-	0	WG
27070	30/09/2018	11:38	-34.03690	139.22453	-	-	0	-	-	1	WG
27071	30/09/2018	12:37	-34.01770	139.22050	-	-	-	-	-	-	
12298	30/09/2018	14:58	-33.88625	139.33797	-	-	-	-	-	-	
27057	1/10/2018	10:20	-33.89031	139.34726	-	-	-	-	-	-	
846	1/10/2018	9:28	-33.88760	139.31972	-	-	0	-	-	0	WG
27058	1/10/2018	11:44	-33.92815	139.28494	-	-	0	-	-	0	WG
10391	6/10/2018	14:34	-33.89412	139.34094	-	-	0	0	1	1	WG
11622	6/10/2018	15:35	-33.88678	139.35533	-	-	0	-	-	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27072	7/10/2018	10:00	-33.86629	139.42618	-	-	0	-	-	0	WG
27082	7/10/2018	17:00	-33.90784	139.42412	-	-	0	1	1	0	WG
27080	7/10/2018	16:07	-33.92692	139.42566	-	-	0	-	-	0	WG
27073	7/10/2018	10:20	-33.86725	139.42615	-	-	0	0	1	1	WG
27076	7/10/2018	11:20	-33.86856	139.42613	-	-	-	-	-	-	
461	7/10/2018	18:20	-33.88432	139.35849	-	-	0	0	1	1	WG
27079	7/10/2018	15:24	-33.89278	139.42503	-	-	0	1	0	1	WG
27077	7/10/2018	11:35	-33.87015	139.42603	-	-	0	-	-	1	WG
27074	7/10/2018	10:45	-33.86725	139.42615	-	-	0	1	1	0	WG
27078	7/10/2018	12:45	-33.88506	139.42532	-	-	0	1	0	0	WG
27075	7/10/2018	10:50	-33.86725	139.42615	-	-	0	1	0	1	WG
27081	7/10/2018	16:45	-33.90933	139.42403	-	-	1	1	0	1	WG
7027	8/10/2018	12:00	-33.86535	139.42633	-	-	0	1	1	1	WG
2451	8/10/2018	10:20	-33.88950	139.42506	-	-	0	-	-	0	WG
27086	8/10/2018	11:35	-33.89378	139.42480	-	-	0	1	1	1	WG
27083	8/10/2018	9:38	-33.86562	139.42630	-	-	0	1	1	1	WG
27085	8/10/2018	10:40	-33.89933	139.42455	-	-	0	-	-	1	WG
4516	8/10/2018	12:40	-33.86152	139.42645	-	-	0	1	1	1	WG
27084	8/10/2018	10:45	-33.90269	139.42447	-	-	1	-	-	0	WG
4803	13/10/2018	17:03	-33.91394	139.30832	-	-	-	-	-	-	
27009	13/10/2018	16:04	-33.92237	139.29451	-	-	-	-	-	-	
11701	13/10/2018	18:15	-33.88770	139.35152	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
610	13/10/2018	14:25	-33.90513	139.32294	-	-	-	-	-	-	
11545	13/10/2018	16:31	-33.92032	139.29791	-	-	-	-	-	-	
8807	13/10/2018	17:35	-33.90373	139.32507	-	-	-	-	-	-	
9372	14/10/2018		-33.88839	139.31082	-	-	-	-	-	-	
11885	14/10/2018		-33.88839	139.31082	-	-	-	-	-	-	
27061	14/10/2018		-33.88715	139.31635	-	-	-	-	-	-	
12723	14/10/2018	11:00	-33.88794	139.31572	-	-	-	-	-	-	
41827	14/10/2018	9:05	-33.88829	139.31215	-	-	-	-	-	-	
8040	14/10/2018	16:13	-33.90407	139.32466	-	-	-	-	-	-	
27059	14/10/2018	11:30	-33.88783	139.31633	-	-	0	0	1	1	WG*
41840	14/10/2018	10:40	-33.88547	139.31035	-	-	-	-	-	-	
4952	14/10/2018	15:40	-33.89307	139.34276	-	-	-	-	-	-	
50935	14/10/2018	16:58	-33.91796	139.30168	-	-	-	-	-	-	
27090	15/10/2018	12:00	-34.03644	139.31398	-	-	0	1	0	0	WG
27088	15/10/2018	11:25	-34.03639	139.31458	-	-	-	-	-	-	
14750	15/10/2018	13:37	-33.92104	139.29668	-	-	0	1	0	0	WG
27089	15/10/2018	11:45	-34.03643	139.31514	-	-	0	-	-	0	WG
27087	15/10/2018	10:47	-34.03814	139.32904	-	-	0	-	-	1	WG
27060	20/10/2018	14:03	-33.88945	139.34859	-	-	1	1	0	1	WG
27062	20/10/2018	16:19	-33.97292	139.21729	-	-	0	-	-	0	WG
875	20/10/2018	14:40	-33.89401	139.34109	-	-	0	-	-	1	WG
27092	21/10/2018	12:25	-34.09363	139.17078	-	-	0	1	1	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27091	21/10/2018	11:38	-34.07679	139.19922	-	-	0	-	-	0	WG
10391	22/10/2018	9:13	-33.89405	139.34102	-	-	-	-	-	-	
6451	22/10/2018	8:25	-33.89030	139.34666	-	-	-	-	-	-	
1769	22/10/2018	9:33	-33.89934	139.33231	1	-	0	1	1	1	WG
7097	22/10/2018	9:33	-33.89927	139.33241	1	-	0	1	1	1	WG
8012	22/10/2018	9:33	-33.89925	139.33263	1	-	-	-	-	-	
27093	28/10/2018	14:11	-34.03622	139.34077	-	-	-	-	-	-	
27094	28/10/2018	15:11	-34.03659	139.28075	-	-	0	-	-	0	WG
27095	28/10/2018	15:46	-34.03664	139.26997	-	-	0	1	0	0	WG
144000 special	28/10/2018	16:48	-34.06903	139.23355	-	-	-	-	-	-	
27096	28/10/2018	17:55	-34.03630	139.31635	-	-	0	-	-	0	WG
27064	29/10/2018	10:18	-33.89245	139.25371	-	-	-	-	-	-	
5034	29/10/2018	17:41	-33.90942	139.31572	-	-	0	1	0	1	WG*
17099	29/10/2018	11:53	-33.89447	139.22757	-	-	-	-	-	-	
1190	30/10/2018	9:34	-33.90669	139.32033	1	-	-	-	-	-	
17169	30/10/2018	9:45	-33.90825	139.31769	-	-	1	-	-	1	WG
27097	2/11/2018	18:03	-34.06862	139.23183	-	-	0	0	1	0	WG
27098	2/11/2018	18:25	-34.06881	139.23248	-	-	0	0	1	1	WG
2771	3/11/2018	9:15	-33.93378	139.35695	-	-	0	1	1	0	WG
27099	3/11/2018	12:19	-34.03630	139.31909	-	-	0	1	0	1	WG
27100	3/11/2018	12:19	-34.03630	139.31909	-	-	0	1	0	0	WG
27063	9/11/2018	16:00	-33.88502	139.35411	-	-	0	1	0	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
854	9/11/2018	16:05	-33.88507	139.35330	-	-	0	1	1	1	WG*
27065	9/11/2018	18:21	-33.88681	139.33017	-	-	0	0	1	1	WG
4997	9/11/2018	17:33	-33.88789	139.31567	-	-	0	1	1	0	WG
27067	10/11/2018	11:04	-33.88758	139.31969	-	-	0	0	1	0	WG
659	10/11/2018	10:19	-33.88544	139.34834	1	-	-	-	-	-	
534	10/11/2018	16:15	-33.91616	139.30467	-	-	-	-	-	-	
27068	10/11/2018	16:20	-33.91575	139.30550	1	-	0	1	1	1	WG
27066	10/11/2018	9:30	-33.88528	139.35088	1	-	0	1	1	1	WG
41274	10/11/2018	11:40	-33.88782	139.31595	1	-	0	0	1	1	WG
11690	10/11/2018	12:45	-33.89041	139.28171	-	-	0	1	1	1	WG
2939	10/11/2018	13:51	-33.88594	139.34209	1	-	0	-	-	1	WG
3396	11/11/2018	9:16	-33.89819	139.33429	-	-	0	1	1	1	WG
27069	11/11/2018	9:17	-33.89828	139.33431	-	-	0	0	1	0	WG
10452	11/11/2018	11:28	-33.92822	139.34590	-	-	0	1	0	0	WG
5350	11/11/2018	10:32	-33.93093	139.28040	1	1	0	-	-	1	WG*
6166	11/11/2018	12:34	-33.90950	139.31583	1	-	0	-	-	1	WG
14020	17/11/2018	17:13	-33.89214	139.25810	-	-	0	0	1	1	WG
839	17/11/2018	18:26	-33.89283	139.24866	1	-	-	-	-	-	
12298	17/11/2018	15:29	-33.88624	139.33833	1	-	0	0	1	1	WG
27101	17/11/2018	16:31	-33.89180	139.26318	-	-	0	-	-	1	WG
12296	17/11/2018	17:41	-33.89280	139.24956	-	-	0	0	1	1	WG
609	18/11/2018	12:00	-33.90118	139.32928	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
9910	18/11/2018	12:00	-33.90118	139.32928	-	-	0	1	0	1	WG
878	18/11/2018	9:13	-33.88755	139.35141	1	-	0	0	1	1	WG*
11701	18/11/2018	9:15	-33.88762	139.35150	1	-	1	1	1	1	WG
8927	18/11/2018	10:20	-33.89251	139.34372	-	-	0	1	0	0	WG
2370	18/11/2018	10:21	-33.89262	139.34351	-	-	0	1	1	0	WG*
27027	18/11/2018	13:20	-33.91394	139.30838	1	-	0	1	1	0	WG
27102	18/11/2018	13:27	-33.91475	139.30707	1	1	0	0	1	0	WG
4952	18/11/2018	10:51	-33.89286	139.34305	1	-	0	1	1	1	WG*
27103	18/11/2018	15:47	-33.94680	139.25531	1	-	0	-	-	0	WG
27104	23/11/2018	16:00	-33.95548	139.21710	1	-	0	1	0	1	WG
27105	23/11/2018	17:04	-33.89441	139.22769	-	-	0	1	0	0	WG
12043	23/11/2018	13:32	-33.96766	139.22369	-	-	0	1	0	0	WG
14086	24/11/2018	12:01	-33.89707	139.27022	-	-	1	1	1	1	WG
12244	24/11/2018	10:12	-33.89363	139.27023	1	1	0	1	0	1	WG
8944	24/11/2018	11:27	-33.89428	139.27022	-	-	0	1	1	1	WG
8786	25/11/2018	9:15	-33.92029	139.29782	1	-	0	1	1	1	WG*
12094	30/11/2018	17:08	-33.92193	139.29516	1	-	0	-	-	0	WG
12153	30/11/2018	16:11	-33.88943	139.34869	-	-	-	-	-	-	
763	1/12/2018	10:11	-33.88566	139.34544	-	-	0	-	-	0	WG*
8770	1/12/2018	18:06	-33.89323	139.34219	1	-	0	-	-	1	WG
8855	1/12/2018	18:07	-33.89322	139.34248	1	-	-	-	-	-	
27106	1/12/2018	9:28	-33.88514	139.35260	-	-	0	-	-	1	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
11460	1/12/2018	8:54	-33.88511	139.35323	-	-	-	-	-	-	
12889	1/12/2018	10:57	-33.88621	139.33800	1	-	-	-	-	-	
6451	1/12/2018	17:53	-33.89095	139.34608	1	-	0	1	0	0	WG*
8373	1/12/2018	17:57	-33.89285	139.34302	-	-	-	-	-	-	
17108	2/12/2018	10:09	-33.92834	139.24544	-	-	0	1	0	0	WG
17159	2/12/2018	13:15	-33.89193	139.26158	-	-	0	1	0	0	WG
27108	2/12/2018	13:18	-33.89185	139.26253	-	-	-	-	-	-	
27056	2/12/2018	16:23	-33.88633	139.33675	-	-	-	-	-	-	
7349	2/12/2018	12:32	-33.89257	139.25243	-	-	0	1	1	1	WG
11656	2/12/2018	11:33	-33.89469	139.22336	1	1	0	-	-	0	WG
14111	2/12/2018	15:31	-33.88876	139.30386	-	-	-	-	-	-	
11798	2/12/2018	14:38	-33.88980	139.29002	1	-	0	-	-	1	WG
27107	2/12/2018	10:46	-33.92070	139.21722	-	-	0	-	-	1	WG
8623	3/12/2018	11:17	-33.91834	139.30115	1	-	0	1	1	1	WG
1521	3/12/2018	10:26	-33.90946	139.31586	-	-	-	-	-	-	
11151	3/12/2018	9:53	-33.90408	139.32460	-	-	-	-	-	-	
11597	3/12/2018	11:52	-33.90282	139.32661	-	-	0	-	-	0	WG
17029	8/12/2018	16:07	-33.88749	139.32097	1	-	0	1	1	1	WG
8117	8/12/2018	8:17	-33.90480	139.32346	-	-	0	1	0	1	WG
27109	8/12/2018	8:17	-33.90480	139.32346	-	-	-	-	-	-	
11584	8/12/2018	9:20	-33.90944	139.31586	-	-	-	-	-	-	
27110	8/12/2018	9:20	-33.90944	139.31586	-	-	0	1	1	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
5040	8/12/2018	10:25	-33.91311	139.30906	1	1	-	-	-	-	
27111	8/12/2018	11:33	-33.94623	139.25601	-	-	0	0	1	1	WG
10839	8/12/2018	16:43	-33.88764	139.31935	-	-	0	1	1	1	WG
12933	9/12/2018	10:10	-33.89407	139.23093	-	-	0	0	1	1	WG
12656	9/12/2018	11:37	-33.89167	139.26430	1	-	-	-	-	-	
12201	9/12/2018	10:52	-33.89226	139.25644	-	-	0	1	0	1	WG
11209 special	16/12/2018	17:10	-33.89314	139.24496	-	-	-	-	-	-	
4579	16/12/2018	16:20	-33.89355	139.23914	-	-	0	1	1	0	WG
40745	16/12/2018	15:27	-33.88814	139.31085	1	1	0	-	-	1	WG
1988	17/12/2018	11:00	-33.92818	139.28488	1	-	-	-	-	-	
4092	17/12/2018	10:27	-33.92835	139.27007	1	-	-	-	-	-	
12364	17/12/2018	11:54	-33.90166	139.32869	-	-	0	-	-	1	WG
27114	22/12/2018	17:19	-33.90714	139.31956	-	-	1	1	1	1	WG
27112	22/12/2018	11:30	-33.95748	139.23911	-	-	0	1	0	1	WG
27113	22/12/2018	12:35	-33.92117	139.29640	-	-	0	1	1	0	WG
27115	22/12/2018	18:35	-33.88565	139.34579	-	-	0	0	1	0	WG
12760	22/12/2018	18:37	-33.88561	139.34616	-	-	-	-	-	-	
3101	22/12/2018	17:58	-33.89354	139.34198	-	-	0	1	1	0	WG
10562	23/12/2018	10:12	-33.88631	139.33728	-	-	0	-	-	1	WG
5040	23/12/2018	11:15	-33.91309	139.30972	-	-	0	1	1	1	WG*
673	23/12/2018	9:30	-33.88514	139.35283	-	-	1	0	1	0	WG
4827	23/12/2018	11:44	-33.91335	139.30940	-	-	0	1	1	1	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27116	5/01/2019	11:03	-33.88759	139.32001	-	-	0	0	1	1	WG
12153	5/01/2019	17:05	-33.88956	139.34865	-	-	0	1	0	0	WG
27117	5/01/2019	17:34	-33.89042	139.34695	-	-	-	-	-	-	
12760	5/01/2019	11:42	-33.88575	139.34554	-	-	0	1	0	1	WG
11480	5/01/2019	9:48	-33.88957	139.29337	-	-	-	-	-	-	
10545	6/01/2019	10:20	-33.92836	139.35429	-	-	0	1	0	1	WG
27118	6/01/2019	10:20	-33.92836	139.35429	-	-	0	-	-	1	WG
8117	6/01/2019	11:38	-33.90445	139.32391	-	-	0	1	0	1	WG
100	19/01/2019	12:45	-33.90443	139.32390	-	-	0	0	1	1	WG
11689	19/01/2019	13:45	-33.88583	139.35501	-	-	0	1	1	1	WG
12759	19/01/2019	13:45	-33.88589	139.35504	-	-	0	1	1	1	WG
27120	9/02/2019	16:08	-34.06642	139.19992	-	-	0	1	1	1	WG
11955	9/02/2019	11:18	-33.92839	139.28473	-	-	0	-	-	0	WG
526	9/02/2019	14:20	-33.89025	139.34732	-	-	0	0	1	1	WG
4829	9/02/2019	14:45	-33.90872	139.31691	-	-	0	-	-	0	WG
27119	9/02/2019	10:50	-34.07676	139.19202	-	-	0	0	1	0	WG
27121	10/02/2019	12:29	-33.89312	139.24504	-	-	0	1	0	1	WG
41401	10/02/2019	13:28	-33.88822	139.31081	-	-	0	0	1	1	WG
2041	16/02/2019	13:15	-33.90864	139.31717	-	-	-	-	-	-	
8743	16/02/2019	15:15	-33.89827	139.33408	-	-	-	-	-	-	
29122	16/02/2019	11:51	-34.11162	139.15598	-	-	-	-	-	-	
10930	17/02/2019	9:07	-33.88841	139.35031	-	-	1	-	-	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
4908 special	17/02/2019	10:09	-33.89319	139.34242	-	-	0	1	0	1	WG
12153	17/02/2019	9:30	-33.88945	139.34854	-	-	-	-	-	-	
11872	17/02/2019	11:29	-33.92946	139.28287	-	-	-	-	-	-	
2620	17/02/2019	10:46	-33.90598	139.32150	-	-	-	-	-	-	
7110	16/03/2019	17:34	-33.94334	139.26062	-	-	-	-	-	-	
27122	17/03/2019	10:10	-33.89021	139.34737	-	-	0	1	1	0	WG
3101	31/03/2019	15:43	-33.89365	139.34180	-	-	-	-	-	-	
12501	8/06/2019	16:23	-33.88629	139.35521	-	-	0	1	0	1	WG
27022	8/06/2019	16:23	-33.88629	139.35521	-	-	0	-	-	0	WG
27123	8/06/2019	16:23	-33.88629	139.35521	-	-	0	0	1	1	WG
12761	8/06/2019	12:45	-33.90057	139.33047	-	-	-	-	-	-	
27124	17/08/2019	11:16	-33.90707	139.31955	-	-	0	-	-	1	WG
10662	17/08/2019	14:15	-33.88984	139.28938	-	-	0	1	0	1	WG
10658	17/08/2019	13:21	-33.88845	139.31065	-	-	0	-	-	1	WG
14611	17/08/2019	10:25	-33.91315	139.30978	-	-	-	-	-	-	
14029	24/08/2019	9:09	-33.89507	139.33957	-	-	0	-	-	0	WG
17276	24/08/2019	12:16	-33.88691	139.32870	-	-	-	-	-	-	
12889	24/08/2019	12:20	-33.88619	139.33871	-	-	1	-	-	0	WG
47110	24/08/2019	11:28	-33.88776	139.31776	-	-	0	1	1	1	WG
27125	30/08/2019	16:35	-33.88603	139.35507	-	-	-	-	-	-	
123	30/08/2019	17:47	-33.92809	139.34030	-	-	0	0	1	1	G*
6137	30/08/2019	17:47	-33.92809	139.34030	-	-	0	1	0	1	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
27127	31/08/2019	12:10	-33.88760	139.31938	-	-	0	1	0	1	WG
12298	31/08/2019	13:23	-33.88625	139.33813	-	-	-	-	-	-	
9390	31/08/2019	16:30	-33.88821	139.30977	-	-	-	-	-	-	
12129	31/08/2019	9:44	-33.92780	139.34055	-	-	0	1	1	1	WG
27126	31/08/2019	9:44	-33.92780	139.34055	-	-	0	1	1	1	WG
11485	31/08/2019	12:43	-33.88709	139.32576	-	-	-	-	-	-	
12138	31/08/2019	15:45	-33.88811	139.31224	-	-	0	-	-	1	WG
27129	1/09/2019	11:05	-33.94617	139.25616	-	-	0	-	-	1	WG
27128	1/09/2019	10:25	-33.94599	139.25600	-	-	0	1	0	1	WG
2686	1/09/2019	9:36	-33.91874	139.30063	-	-	0	-	-	1	WG
27012	1/09/2019	11:55	-33.95166	139.24800	-	-	0	1	0	1	WG
27130	7/09/2019	10:10	-33.95943	139.22981	-	-	0	1	0	1	WG
26000	7/09/2019	9:25	-33.95288	139.24599	-	-	0	1	1	0	WG
17183	7/09/2019	14:30	-33.90100	139.32967	-	-	-	-	-	-	
877	8/09/2019	12:25	-33.88870	139.30496	-	-	0	-	-	1	WG
27131	8/09/2019	11:38	-33.88633	139.33644	-	-	-	-	-	-	
41207	13/09/2019	16:15	-33.88836	139.31049	-	-	-	-	-	-	
41722	13/09/2019	14:50	-33.88781	139.31595	-	-	0	1	1	0	WG
27053	13/09/2019	14:55	-33.88781	139.31604	-	-	0	1	0	1	WG
10581	14/09/2019	14:10	-33.89423	139.23053	-	-	0	-	-	0	WG
822	14/09/2019	12:15	-33.89157	139.26630	-	-	-	-	-	-	
607	14/09/2019	11:25	-33.89028	139.28378	-	-	0	-	-	0	WG*

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
31800	14/09/2019	10:35	-33.88980	139.28983	-	-	0	1	0	1	WG
27133	14/09/2019	15:38	-33.90330	139.21713	-	-	0	-	-	1	WG
7287	14/09/2019	13:45	-33.89427	139.22952	-	-	0	1	1	1	WG
27132	14/09/2019	12:52	-33.89178	139.26294	-	-	0	-	-	0	WG
6281 special	14/09/2019	14:56	-33.89445	139.22679	-	-	0	1	0	1	WG*
27134	14/09/2019	16:57	-33.88646	139.33499	-	-	0	1	1	1	WG
11480	15/09/2019	11:11	-33.88955	139.29333	-	-	-	-	-	-	
10930	15/09/2019	9:15	-33.88516	139.35086	-	-	1	1	0	1	WG
31194?	15/09/2019	9:48	-33.88788	139.31561	-	-	-	-	-	-	
41274	15/09/2019	9:50	-33.88793	139.31580	-	-	0	1	1	1	WG
27065	21/09/2019	14:08	-33.88680	139.33081	-	-	-	-	-	-	
27019	21/09/2019	13:23	-33.88584	139.34706	-	-	0	-	-	1	WG
10839	21/09/2019	14:45	-33.88763	139.31946	-	-	0	-	-	1	WG
27135	22/09/2019	12:15	-33.95961	139.23582	-	-	-	-	-	-	
27138	22/09/2019	10:21	-33.94607	139.25598	-	-	0	1	0	1	WG
4952	22/09/2019	9:23	-33.89307	139.34261	-	-	-	-	-	-	
3381	22/09/2019	11:25	-33.95586	139.24147	-	-	-	-	-	-	
34402	22/09/2019	14:30	-33.89161	139.26633	-	-	-	-	-	-	
11656	22/09/2019	13:52	-33.89468	139.22348	-	-	-	-	-	-	
9372	23/09/2019	13:05	-33.88832	139.31128	-	-	0	-	-	1	WG
41747	23/09/2019	12:22	-33.88863	139.31099	-	-	0	-	-	1	WG
5690	23/09/2019	11:28	-33.89704	139.27042	-	-	0	1	1	1	WG*

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
12855	23/09/2019	9:55	-33.91343	139.27034	-	-	0	-	-	0	WG
27136	23/09/2019	9:56	-33.91343	139.27034	-	-	-	-	-	-	
2536	27/09/2019	17:15	-33.89812	139.42470	-	-	-	-	-	-	
27137	28/09/2019	10:20	-33.88869	139.30592	-	-	0	1	1	1	WG
40018	28/09/2019	11:21	-33.88803	139.31468	-	-	0	-	-	1	WG
27139	5/10/2019	10:20	-33.92802	139.42682	-	-	1	1	0	1	WG
27140	5/10/2019	16:15	-33.92641	139.43799	-	-	0	1	0	1	WG
11900	6/10/2019	9:50	-33.92816	139.40091	-	-	-	-	-	-	
27144	11/10/2019	11:20	-34.09177	139.17429	-	-	0	1	0	1	WG
27142	11/10/2019	12:20	-33.96616	139.22601	-	-	0	1	1	1	WG
27147	11/10/2019	12:20	-33.96616	139.22601	-	-	-	-	-	-	
24190	11/10/2019	17:17	-33.88705	139.32718	-	-	0	1	0	1	WG
4876	11/10/2019	16:35	-33.88509	139.35335	-	-	0	1	1	1	WG
27063	11/10/2019	15:58	-33.88505	139.35391	-	-	0	1	1	1	WG
27143	12/10/2019	12:00	-33.92533	139.21712	-	-	0	-	-	1	WG
686	12/10/2019	14:24	-33.89410	139.23186	-	-	-	-	-	-	
12424	12/10/2019	14:24	-33.89410	139.23186	-	-	0	1	1	1	WG
27145	12/10/2019	12:50	-33.91323	139.21718	-	-	0	1	0	1	WG
27146	12/10/2019	12:50	-33.91323	139.21718	-	-	0	1	0	1	WG
27148	12/10/2019	15:50	-33.89259	139.25217	-	-	0	1	1	1	WG
11102	13/10/2019	10:38	-33.89002	139.28699	-	-	0	-	-	1	WG
#01	17/10/2019	12:25	-34.01795	139.22052	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
#02	17/10/2019	14:10	-33.92792	139.42682	-	-	0	1	0	1	WG
#03	17/10/2019	14:49	-33.95459	139.42638	-	-	0	1	0	1	WG
#04	17/10/2019	15:24	-33.97093	139.41905	-	-	0	1	0	1	WG*
#05	17/10/2019	16:00	-33.97126	139.38280	-	-	0	1	0	1	WG
#06	17/10/2019	16:45	-33.97134	139.37149	-	-	0	1	0	1	WG
27149	17/10/2019	18:28	-33.88587	139.35529	-	-	-	-	-	-	
#07	18/10/2019	10:20	-34.08719	139.18045	-	-	0	1	0	1	WG
#08	18/10/2019	11:20	-34.15963	139.08852	-	-	0	-	-	1	WG
#09	18/10/2019	14:20	-34.17785	139.09116	-	-	0	1	0	1	WG
27150	25/10/2019	14:00	-33.88599	139.35510	-	-	0	1	0	0	WG
27151	25/10/2019	15:15	-33.88602	139.35509	-	-	0	-	-	0	WG
27155	26/10/2019	15:21	-33.88860	139.42505	-	-	0	1	1	1	WG
27157	26/10/2019	16:21	-33.88390	139.42531	-	-	0	1	1	1	WG
27152	26/10/2019	10:40	-33.89685	139.42465	-	-	0	1	0	1	WG
27154	26/10/2019	12:39	-33.87741	139.42564	-	-	0	1	1	1	WG
27153	26/10/2019	11:45	-33.89133	139.42493	-	-	0	1	0	0	WG
27156	2/11/2019	16:14	-33.89602	139.33791	-	-	-	-	-	-	
775	2/11/2019	15:26	-33.88987	139.34785	-	-	0	-	-	1	WG
17293	2/11/2019	14:45	-33.88676	139.35298	-	-	0	1	1	1	WG*
4964	3/11/2019	18:00	-33.95561	139.24188	-	-	0	1	1	1	WG
#10	3/11/2019	16:30	-34.15380	139.12439	-	-	0	1	0	1	WG
27158	4/11/2019	10:35	-33.92699	139.37273	-	-	0	1	0	0	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
40722	4/11/2019	12:39	-33.88789	139.31564	-	-	0	-	-	1	WG
28104	8/11/2019	15:07	-33.88605	139.34021	-	-	0	-	-	1	WG
27106	8/11/2019	14:25	-33.88512	139.35225	-	-	0	1	0	1	WG
12847	9/11/2019	14:00	-33.88857	139.31099	-	-	0	1	0	1	WG
12737	9/11/2019	11:11	-33.88652	139.33401	-	-	0	1	0	1	WG
11885	9/11/2019	13:20	-33.88857	139.31099	-	-	0	1	0	1	WG
41204	9/11/2019	12:39	-33.88786	139.31548	-	-	1	1	1	1	WG
12935	9/11/2019	16:47	-33.88920	139.29756	-	-	0	1	0	0	WG
846	9/11/2019	11:54	-33.88763	139.31935	-	-	0	1	1	1	WG
11505	9/11/2019	15:50	-33.88877	139.31081	-	-	0	-	-	1	WG
41274	10/11/2019	10:13	-33.88779	139.31651	-	-	0	1	1	1	WG
27159	10/11/2019	12:12	-33.88639	139.33571	-	-	0	1	1	1	WG
11819	10/11/2019	11:22	-33.88943	139.29488	-	-	0	1	0	1	WG
32727	15/11/2019	17:25	-33.88683	139.32970	-	-	-	-	-	-	
40077	15/11/2019	16:37	-33.88786	139.31589	-	-	0	-	-	1	WG
881	15/11/2019	15:46	-33.88608	139.34019	-	-	0	-	-	1	WG
27160	15/11/2019	15:46	-33.88608	139.34019	-	-	0	-	-	0	WG
17127	15/11/2019	15:59	-33.88666	139.33180	-	-	0	-	-	1	WG
9372	16/11/2019	13:10	-33.88820	139.31165	-	-	0	-	-	0	WG
1588	16/11/2019	14:18	-33.92164	139.29570	-	-	0	1	0	1	WG
17125	16/11/2019	16:22	-33.88668	139.33168	-	-	0	1	0	0	WG
120833 special	16/11/2019	15:32	-33.88861	139.31113	-	-	0	1	1	1	WG

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
12754	16/11/2019	16:58	-33.88597	139.34163	-	-	0	1	0	1	WG
5444	17/11/2019	12:01	-33.89422	139.23048	-	-	0	1	0	1	WG*
3024	17/11/2019	10:18	-33.93129	139.21712	-	-	0	1	1	1	WG
27161	17/11/2019	14:34	-33.88615	139.33916	-	-	0	-	-	0	WG
27052	17/11/2019	13:36	-33.88976	139.29018	-	-	0	-	-	1	WG
7455	17/11/2019	12:42	-33.89218	139.25751	-	-	0	1	0	1	WG*
8775 special	17/11/2019	10:55	-33.92812	139.21712	-	-	0	1	1	0	WG
27162	18/11/2019	9:20	-33.94603	139.25595	-	-	0	-	-	0	WG
7906	18/11/2019	9:22	-33.94599	139.25601	-	-	-	-	-	-	
5231	18/11/2019	9:23	-33.94606	139.25606	-	-	-	-	-	-	
10658	18/11/2019	12:21	-33.88840	139.31088	-	-	0	1	1	1	WG
6186	18/11/2019	10:55	-33.94630	139.25623	-	-	0	1	1	1	WG
27163	23/11/2019	12:15	-33.92830	139.28366	-	-	0	1	0	1	WG*
27164	23/11/2019	13:23	-33.97145	139.31032	-	-	0	1	0	0	WG
7754	23/11/2019	17:42	-33.97427	139.35658	-	-	0	-	-	0	WG*
27167	30/11/2019	16:16	-33.89529	139.33902	-	-	0	1	0	1	WG
14426	30/11/2019	15:20	-33.91044	139.31407	-	-	0	1	0	1	WG
27166	30/11/2019	13:27	-33.96736	139.22417	-	-	0	1	0	0	WG
14367	30/11/2019	14:42	-33.92202	139.29514	-	-	0	1	0	1	WG
27165	30/11/2019	12:52	-33.96856	139.22243	-	-	0	1	0	1	WG
854	1/12/2019	10:33	-33.88514	139.35291	-	-	0	1	1	1	WG
4796	6/12/2019	18:24	-33.91287	139.31013	-	-	-	-	-	-	

Appendix 7.6 continued

ID	Date	Time	GPS (S)	GPS (E)	<i>Thelandros trachysauri</i>	<i>Oochoristica trachysauri</i>	<i>Hemolivia mariae</i>	IIT 1	IIT 2	IIT 3	Note
6103	6/12/2019	18:33	-33.90305	139.32622	-	-	-	-	-	-	
27168	6/12/2019	17:35	-33.95973	139.23570	-	-	0	1	1	1	WG
12153	6/12/2019	18:42	-33.88945	139.34854	-	-	0	-	-	1	WG
17017	7/12/2019	11:24	-33.89154	139.26587	-	-	-	-	-	-	
10447	7/12/2019	12:36	-33.90027	139.33093	-	-	0	1	1	0	WG
1041	7/12/2019	18:52	-33.88772	139.31726	-	-	0	1	0	0	WG
14729	8/12/2019	9:15	-33.89495	139.33955	-	-	0	1	0	1	WG
27169	8/12/2019	11:26	-33.95249	139.24664	-	-	0	1	0	0	WG
11572	8/12/2019	10:30	-33.90078	139.33011	-	-	0	1	0	1	WG
4831	8/12/2019	8:33	-33.89368	139.34177	-	-	0	1	0	1	WG
17391	8/12/2019	9:51	-33.89895	139.33308	-	-	1	1	0	1	WG
5845	15/12/2019	10:00	-33.90018	139.33090	-	-	0	1	0	0	WG*
8012	15/12/2019	9:15	-33.89846	139.33379	-	-	0	-	-	1	WG
12153	15/12/2019	8:30	-33.88943	139.34857	-	-	0	-	-	0	WG
11545	15/12/2019	15:39	-33.91998	139.29729	-	-	0	1	0	1	WG
27171	16/12/2019	10:14	-33.94204	139.21722	-	-	0	-	-	1	WG
27170	16/12/2019	9:21	-33.89156	139.26642	-	-	0	-	-	1	WG
4880	16/12/2019	11:30	-33.91996	139.29729	-	-	0	-	-	0	WG