

Who's on the menu : marsupial carnivore feeding ecology and extinction risk

Author:

Attard, Marie

Publication Date:

2013

DOI:

<https://doi.org/10.26190/unsworks/2512>

License:

<https://creativecommons.org/licenses/by-nc-nd/3.0/au/>

Link to license to see what you are allowed to do with this resource.

Downloaded from <http://hdl.handle.net/1959.4/53130> in <https://unsworks.unsw.edu.au> on 2024-05-03

WHO'S ON THE MENU: MARSUPIAL CARNIVORE FEEDING ECOLOGY AND EXTINCTION RISK



Thesis submitted by

MARIE ATTARD

For the Degree of Doctor of Philosophy

in the

School of Biological, Earth & Environmental Sciences

Faculty of Science

March 2013



UNSW
THE UNIVERSITY OF NEW SOUTH WALES

THE UNIVERSITY OF NEW SOUTH WALES

Thesis/Dissertation Sheet

Surname or Family name: Attard

First name: Marie

Other name/s: Rosanna Gabrielle

Abbreviation for degree as given in the University calendar: PhD

School: School of Biological, Earth and Environmental Sciences

Faculty: Faculty of Science

Title: Who's on the menu: marsupial carnivore feeding ecology and extinction risk

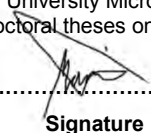
Abstract

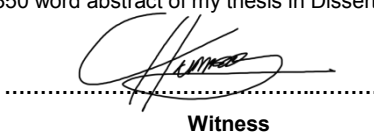
The aim of this thesis is to assess the role of diet in the extinction of Australia's iconic marsupial carnivore, the thylacine (*Thylacinus cynocephalus*) in Tasmania. Herein, we present two novel techniques to address fundamental questions regarding their maximum prey size and potential competition with sympatric predators. Three-dimensional computer models of the thylacine skull were used to assess their biomechanical limitations in prey size within a comparative context. This included living relatives from the family Dasyuridae as well as a recently recovered fossil, *Nimbacinus dickoni*, from the family Thylacinae. Stable isotope ratios of carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) of tissues from thylacine and potential prey species were used to assess the thylacine's dietary composition. Furthermore, we integrate historical and recent marsupial carnivore stable isotope data to assess long-term changes in the ecosystem in response to multiple human impacts following European settlement. Our biomechanical findings support the notion that solitary thylacines were limited to hunting prey weighing less than their body mass. This concurs with our stable isotope results, which suggest medium-sized mammals were a major element of thylacine subsistence in Tasmania. Prey species may have included the common wombat (*Vombatus ursinus*), red-necked wallaby (*Macropus rufogriseus*) and Tasmanian pademelon (*Thylogale billardierii*). These species fall within the expected size range of prey needed to sustain this large carnivore. Competition among thylacines and sympatric marsupial carnivores in Tasmania was suggested by similarities in their prey size and trophic niche. If interspecific competition had been a major limiting factor for the small thylacine population, encounters with smaller predators were possibly reduced by occupying different habitats, as indicated by differences in predator $\delta^{13}\text{C}$ values. The century-long change in Tasmanian devil and spotted-tailed quoll $\delta^{13}\text{C}$ values suggests a change in vegetation in the areas typically inhabited by these species, or more likely indicates the movement of these top predators away from traditional open grassland, mosaic habitats to more densely forested habitats due to habitat loss and fragmentation. These species also displayed temporal changes in their $\delta^{15}\text{N}$ values, which may be attributed changing prey availability or changes in the source nitrogen at the base of the food. This thesis provides a framework to test the resilience of marsupial carnivores to anthropogenic impacts in light of their biological traits and may be used to improve the conservation of large predators.

Declaration relating to disposition of project thesis/dissertation

I hereby grant to the University of New South Wales or its agents the right to archive and to make available my thesis or dissertation in whole or in part in the University libraries in all forms of media, now or here after known, subject to the provisions of the Copyright Act 1968. I retain all property rights, such as patent rights. I also retain the right to use in future works (such as articles or books) all or part of this thesis or dissertation.

I also authorise University Microfilms to use the 350 word abstract of my thesis in Dissertation Abstracts International (this is applicable to doctoral theses only).


.....
Signature


.....
Witness

28/03/2013
.....
Date

The University recognises that there may be exceptional circumstances requiring restrictions on copying or conditions on use. Requests for restriction for a period of up to 2 years must be made in writing. Requests for a longer period of restriction may be considered in exceptional circumstances and require the approval of the Dean of Graduate Research.

FOR OFFICE USE ONLY

Date of completion of requirements for Award:

COPYRIGHT STATEMENT

'I hereby grant the University of New South Wales or its agents the right to archive and to make available my thesis or dissertation in whole or part in the University libraries in all forms of media, now or here after known, subject to the provisions of the Copyright Act 1968. I retain all proprietary rights, such as patent rights. I also retain the right to use in future works (such as articles or books) all or part of this thesis or dissertation.

I also authorise University Microfilms to use the 350 word abstract of my thesis in Dissertation Abstract International (this is applicable to doctoral theses only).

I have either used no substantial portions of copyright material in my thesis or I have obtained permission to use copyright material; where permission has not been granted I have applied/will apply for a partial restriction of the digital copy of my thesis or dissertation.'

Signed



Date

03/02/2014

AUTHENTICITY STATEMENT

'I certify that the Library deposit digital copy is a direct equivalent of the final officially approved version of my thesis. No emendation of content has occurred and if there are any minor variations in formatting, they are the result of the conversion to digital format.'

Signed

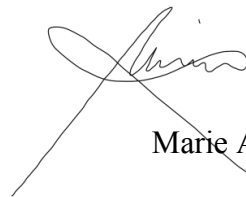


Date

03/02/2014

ORIGINALITY STATEMENT

I hereby declare that this submission is my own work and to the best of my knowledge it contains no materials previously published or written by another person, or substantial proportions of material which have been accepted for the award of any other degree or diploma at UNSW or any other educational institution, except where due acknowledgement is made in the thesis. Any contribution made to the research by others, with whom I have worked at UNSW or elsewhere, is explicitly acknowledged in the thesis. I also declare that the intellectual content of this thesis is the product of my own work, except to the extent that assistance from others in the project's design and conception or in style, presentation and linguistic expression is acknowledged.



Marie Attard
28 March 2013

ACKNOWLEDGEMENTS

Completing my PhD was truly a marathon event, and I would not have been able to accomplish this journey without the aid and support of countless people over the past four years. Foremost, I would like to express my sincere appreciation to my joint-supervisors, Tracey Rogers and Steve Wroe for their continuous support and excellent guidance on how to negotiate the academic world. Tracey's love for all things mammals, insightful ideas and skill in communicating science has been a great asset to the project and has helped develop my skills as a researcher. Steve's vast knowledge of marsupial carnivores and all-round ingenuity and kindness has been an inspiration to me that I will never forget.

Uphar Chamoli, William Parr, Toni Ferrara, Laura Wilson and Natalie Rogers from the Computational Biomechanics research group have provided many stimulating discussions and a never ending level of support. I would especially like to thank Uphar Chamoli for generously giving up his time to help me with various problems over the years relating to my biomechanics research. I have never seen anyone as skilled or quick to tackle problems as Uphar, and I know he will go a long way in his career. I would also like to thank my colleagues in the Rogers' research group, Marlee Tucker, Naysa Balcazar-Cabrera, Jeffery Fung, Jessica Meade, and earlier on Tiffanie Nelson, Nadine Constantinou, Lisa Steindler, Benjamin Jeggle, Tempe Adams and Michaela Ciaglia. They have provided me with many good memories and have brightened up our lab with their humour and fantastic baking skills.

I also thank my fellow postgraduate colleagues for sharing with me the battles of PhD life and joy in finally getting our research out there. Together we have proved that no matter how small (or dead) the research subject, our research matters. I would particularly like to thank Alan Kwok, Samantha Travers, Joanne Ocock, Celine Steinfeld and Stefani Daryanto for their friendship and providing me with a listening ear in a difficult situation.

I would like to acknowledge the generosity of various museums for opening the doors to their collection and providing tissues from their precious specimens. I hope my research is able to enlighten others about the value of museum collections, which is usually underestimated. I would like to personally thank Darrin Lunde (American Museum of Natural History), Gerald Legg (Booth Museum), Matt Lowe (Cambridge Zoological Museum), Clare Brown (Leeds City Museum), Lawrence Heaney and William Stanley (Field Museum of Natural History), Virginie Volpato (Forschungsinstitut & Schaumuseum Senckenberg), Göran Nilson (Gothenburg Natural History Museum), Maggie Reilly (Hunterian Museum and Art Gallery), Østyein Wiig and Fridtjof Mehlum (Natural History Museum), Vicki Humphrey and Nicki Smith (National Museum of Australia) Wayne Longmore (Museum Victoria), Vicki Humphrey and Nicki Smith (National Museum of Australia), Tammy Gordon (Queen Victoria Museum and Art Gallery), Kathryn Medlock and Nicole Zehntner (Tasmanian Museum and Art Gallery), Mikael Siversson (Western Australian Museum), Milly Farrell and Simon Chaplin (The Royal College of Surgeons of England) Clem Fisher and Tony Parker (World Liverpool Museum) for their kind permission and assistance with sampling of collections from their affiliated institution. Fresh tissue samples were also provided by Bronwyn Fancourt (University of Tasmania), Sarah Peck (Department of Primary Industries, Parks, Water and Environment), Andrew Palmer (Tasmanian Quality Meats) and Damien Stanioch. I cannot express my gratitude enough for your kind support of my research, and for believing in me. I would also like to thank all the museums that kindly considered my request for samples but were unable to provide tissues. The decision to provide tissues from rare and extinct species is extremely difficult, and must be carefully weighed against the benefits of the research.

Towards the start of the project, Stephen Sleightholme kindly helped me identify suitable thylacine material for the stable isotope component of this thesis. Michaela Ciaglia, Edith King, Larisa DeSantis and Tamsin O'Connell provided useful advice on the preparation of tissue samples and Anita Andrew analysed all samples for stable isotope research. Kirsty Collard, Ceiwen Pease and Marina Pizà provided technical

assistance in running the freeze dryer and were extremely generous with their time. Donald Phillips provided invaluable guidance with the application of Bayesian mixing models to the stable isotope data. For the biomechanics component of my research, I would like to thank makers of Digimorph for providing me with access to their CT scan data and Sandy Ingleby from the Australian Museum for loaning specimens to my research group and allowing me to visit their natural history collection.

This research was supported by several funding bodies: the Australian Research Council, the Strategic Initiatives and Performance Group and the School of Biological, Earth and Environmental Sciences at the University of New South Wales. Robert Beale from the UNSW science media office helped open my eyes to the infinite possibilities of getting my science 'out there' to the public.

I would especially like to thank my awesome partner Christopher for constantly reminding me that there is a life outside of my PhD. Thanks for letting me drag you out into the previously uncharted (at least by us) lands of Tasmania to help collect maggot-infested road kill. I would also like to thank the (anonymous) family who helped save our tent from imminent destruction during our first gusty night in Tassie.

My love and thanks to 'the girls' of the family — my mum, Cath and sisters, Catherine and Teresa — your critiques and comments will hopefully made this thesis a memorable one. Well done Catherine on achieving your doctorate in the same year as me — such an academic family! And finally, to my dog Toby for patiently sitting by my side at home for many hours while I wrote my thesis.

ABSTRACT

Australia has the highest risk of extinction in the world, with 22 mammals becoming extinct over the past 200 years. An understanding of biological species traits associated with increased extinction vulnerability will be necessary to help improve conservation management decisions. The thylacine (*Thylacinus cynocephalus*) was the largest marsupial carnivore to survive up to European settlement in Tasmania and became extinct in 1936. Factors contributing to their extinction have been debated and will benefit from an improved understanding of their ecology. A multidisciplinary approach was used to assess two biological traits in the thylacine that are commonly associated with high extinction risk among large mammalian predators: maximum prey size and competition. Firstly, a three-dimensional finite element model (FEM) of the thylacine skull was used to assess their biomechanical limitations in prey size within a comparative context. This included living relatives from the family Dasyuridae as well a recently recovered fossil, *Nimbacinus dickoni*, from the family Thylacindae. Secondly, stable isotope analysis of preserved tissues from thylacines, sympatric predators and potential prey species were used to assess the diet composition of the thylacine and test for niche overlap with other marsupial carnivores. Furthermore, we integrate historical and recent stable isotope data of marsupial carnivore tissues to assess long-term changes in the ecosystem in response to multiple human impacts following European arrival in Tasmania.

Our biomechanical findings support the notion that solitary thylacines were limited to hunting prey weighing less than their own body mass. This concurs with stable isotope analysis of preserved tissues that suggest medium-sized (13-30 kg) mammals were a major element of thylacine subsistence in Tasmania. Prey species most likely to be preyed upon include the common wombat (*Vombatus ursinus*), red-necked wallaby (*Macropus rufogriseus*) and Tasmanian pademelon (*Thylogale billardierii*). These species fall within the expected size range of prey needed to sustain large (>21.5 kg) terrestrial mammalian carnivores. This would suggest that the size of prey targeted by

thylacines was sufficient to meet their energetic requirements. Co-operative hunting may have permitted predation upon larger prey, such as the forester kangaroo (*Macropus giganteus*), but the stable isotope composition of this herbivore indicated that they formed a negligible component of the thylacine's diet.

Competition among thylacines and sympatric marsupial carnivores in Tasmania was suggested by similarities in their prey size and trophic niche. If interspecific competition had been a major limiting factor for the small thylacine population, encounters with smaller predators were possibly reduced by occupying different habitats, as indicated by differences in predator $\delta^{13}\text{C}$ values. The century-long decrease in $\delta^{13}\text{C}$ values of extant marsupial carnivores in Tasmania reveals a dramatic change in vegetation in the areas typically inhabited by these species, or more likely indicates the movement of these species away from traditional open grassland, mosaic habitats to more densely forested habitats due to human persecution, habitat loss and fragmentation. This thesis provides a framework to test the resilience of marsupial carnivores to changing environments in light of their biological traits and may be used to improve the conservation of large predators.

PREFACE

This thesis compiles three stand-alone manuscripts that have been published (Chapter 2), recently submitted for publication (Chapter 3), or are intended for publication in peer-reviewed journals (Chapter 4). Some sections of the thesis introduction (Chapter 1) have been adapted from the Australasian Science Magazine article 'The thylacine myth' written by Marie Attard and Stephen Wroe. Each chapter is self-contained and subsequently, some repetition occurs. Tables and figures are sequentially numbered within each chapter and the appendices can be found at the end of the thesis. The appendices are divided into four sections, and are denoted by a letter. Tables and figures within each appendices section have been individually numbered (e.g. Appendix A1). A single reference list has been provided at the end of the dissertation to avoid unnecessary duplication.

This thesis is a compilation of my own work, with guidance from my joint-supervisors A/Prof Tracey Rogers and A/Prof Stephen Wroe. Apart from the contributions of co-authors outlined below, I conceptualised the research, conducted all field work and data analysis, interpreted the results and wrote all chapters included in this thesis. No other authors will be submitting this work as part of their thesis submission. Co-authors proof-read and edited the final manuscript versions as is required for publications. The contributions of each co-author are detailed below.

Chapter 2

Skull mechanics and implications for feeding behaviour in a large marsupial carnivore guild: the thylacine, Tasmanian devil and spotted-tailed quoll

Authors: Marie R.G. Attard ^{1*}, Uphar Chamoli ¹, Toni L. Ferrara ¹, Tracey L. Rogers ¹ and Stephen Wroe^{1,2}

¹Evolution and Ecology Research Centre, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia.

²Evolution of Earth and Life Sciences Research Centre, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia

Marie Attard can claim credit to over 80% of the work since she gathered the literature, built the finite element models, performed the data analysis and wrote the manuscript. Steve Wroe conceived the project that formed the basis of the manuscript and created the 3D digital images in Mimics software. Marie Attard was trained by Steve Wroe, Uphar Chamoli and Toni Ferrara to produce, solve and analyse the finite element models. Steve Wroe, Uphar Chamoli and Tracey Rogers assisted in the interpretation of results. All co-authors critically reviewed the manuscript.

Chapter 3

Virtual Reconstruction and prey size preference of the Mid Cenozoic Thylacinid, *Nimbacinus dicksoni* (Thylacinidae, Marsupialia)

Authors: Marie R.G. Attard ^{1*}, William Parr ¹, Michael Archer ², Suzanne J. Hand ², Laura A. B. Wilson ¹, Tracey L. Rogers ¹ and Stephen Wroe^{1,2}

¹Evolution and Ecology Research Centre, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia

²Evolution of Earth and Life Sciences Research Centre, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia

Marie Attard can claim credit to over 90% of the work since she gathered the literature, built the finite element models, performed the data analysis and wrote the manuscript. Steve Wroe and Marie Attard jointly conceptualised the project. William Parr digitally reconstructed the fossil carnivorous marsupial, *Nimbacinus dicksoni*, discovered by Mike Archer and Sue Hand. All co-authors critically reviewed the manuscript.

Chapter 4

New insight from the old: Using stable isotopes to assess marsupial carnivore vulnerability to anthropogenic impacts

Authors: Marie R.G. Attard ^{1*}, Stephen Wroe¹ and Tracey L. Rogers ¹

¹Evolution and Ecology Research Centre, School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, New South Wales 2052, Australia

Marie Attard can claim credit to more than 95% of the work since she gathered the literature, organised the collection of most samples, performed all fieldwork and data analysis and wrote the manuscript. Tracey Rogers and Marie Attard jointly conceived the project that formed the basis of the manuscript. Tracey Rogers co-ordinated with and collected samples from the Leeds Museum, American Museum of Natural History and Cambridge University. All co-authors assisted in the interpretation of the data and critically reviewed the manuscript.

A number of the studies reported in this thesis have been published in journals, magazine and feature articles or presented at conference proceedings as outlined below.

Chapter 1

Attard, M. R. G. and Wroe, S. (2012) –The thylacine myth” *Australasian Science Magazine*, **6**, 19-22.

Chapter 2

Attard, M. R. G., Chamoli, U., Ferrara, T., Rogers, T. L. and Wroe, S. (2011). Skull biomechanics and implications for feeding behaviour in a large marsupial carnivore guild: the thylacine, Tasmanian devil and spotted-tailed quoll. *Journal of Zoology*, **285**, 292-300.

Attard, M. R. G. and Wroe, S. (2012) –The thylacine myth” *Australasian Science Magazine*, **6**, 19-22.

Attard, M. R. G., Chamoli, U. and Wroe, S. (April, 2011) Skull mechanics and implications for feeding behaviour in a large marsupial carnivore guild. Oral presentation at the 13th Conference on Australian Vertebrate Evolution, Palaeontology and Systematics (CAVEPS), Perth, Western Australia.

Attard, M. R. G., Chamoli, U. and Wroe, S. (December, 2011) Skull mechanics and its implications for feeding behaviour in a large marsupial carnivore guild. Oral presentation at 4th International Conference on the Mechanics of Biomaterials and Tissues, Kona, Hawaii.

Attard, M. R. G., Chamoli, U., Ferrara, T., Rogers, T. L. and Wroe, S. (December, 2011) Thylacine's weak jaws linked to extinction. Poster presented at 4th

International Conference on the Mechanics of Biomaterials and Tissues, Kona, Hawaii.
 Attard, M. R. G., Chamoli, U., Ferrara, T., Rogers, T. L. and Wroe, S. (September, 2012) Diet and link to extinction in Australia's iconic thylacine. Guest speaker at workshop, “Using Virtual Reconstruction and Computational Biomechanics to study form and function in biology”, University of New South Wales, Sydney, Australia.

Chapter 3

Attard, M.R.G., Parr, W., Archer, M., Hand, S. J., Rogers, T. L. and Wroe, S. (In review) Virtual reconstruction, biomechanical modeling and the prediction of diet of the Australian mid Cenozoic thylacinid, *Nimbacinus dicksoni* (Thylacinidae, Marsupialia). *Plos One*.
 Attard, M.R.G., Wroe, S. and Rogers, T. L. (December, 2012) The thylacine myth: stable isotopes and skull biomechanics reveal their actual diet and extinction risk, oral presentation at the Ecological Society of Australia 2012 Annual Conference, Melbourne, Australia.

Chapter 4

Attard, M.R.G., Wroe, S. and Rogers, T. L. (December, 2012) The thylacine myth: stable isotopes and skull biomechanics reveal their actual diet and extinction risk, oral presentation at the Ecological Society of Australia 2012 Annual Conference, Melbourne, Australia.

ABBREVIATIONS AND SYMBOLS

Terms and symbols

ANOVA: Analysis of Variance

CT: Computer Tomography

FEA: Finite Element Analysis

FEM: Finite Element Model

mya: Million years ago

TMJ: Temporomandibular joint

VM: Von-Mises

$\delta^{15}\text{N}$: Stable isotope ratio of ^{15}N to ^{14}N

$\delta^{13}\text{C}$: Stable isotope ratio of ^{13}C to ^{12}C

Δ : Difference

Units

‰: Parts per thousand (per mil)

KPa: Kilo-Pascal

GPa: Giga-Pascal

g: Gram

Kg: Kilogram

mm: Millimetre

cm: Centimetre

m: Metre

N: Newton

Species

D. maculatus: *Dasyurus maculatus*, Spotted-tailed quoll

D. viverrinus: *Dasyurus viverrinus*, Eastern quoll

N. dicksoni: *Nimbacinus dicksoni*

S. harrisii: *Sarcophilus harrisii*, Tasmanian devil

T. cynocephalus: *Thylacinus cynocephalus*, Thylacine

TABLE OF CONTENTS

Originality statement	i
Acknowledgements	ii
Abstract	v
Preface	vii
Abbreviations and symbols	xi
Table of contents	xii
List of figures	xvi
List of tables	xxi
List of appendices	xxiii

CHAPTER 1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

1.1	Chapter outline	2
1.2	Rationale	2
1.3	The link between mammal body size, competition and vulnerability	3
	1.3.1 Body size	3
	1.3.2 Competition	5
1.4	Consequences of apex predator extinctions	6
1.5	History of Thylacinidae	8
1.6	Extinction risk in the thylacine	10
	1.6.1 Diet and prey size	10
	1.6.2 Intraguild competition and predation	13
	1.6.3 Home range	13
	1.6.4 Disease and genetic diversity	14
	1.6.5 Human population expansion	14
	1.6.6 Hunting	15
	1.6.7 Habitat loss and modification	16
1.7	Aims of the program of research	17
1.8	Thesis overview	18

CHAPTER 2. SKULL MECHANICS AND IMPLICATIONS FOR FEEDING BEHAVIOUR IN A LARGE MARSUPIAL CARNIVORE GUILD: THE THYLACINE, TASMANIAN DEVIL AND SPOTTED-TAILED QUOLL

2.1	Abstract	20
2.2	Introduction	21
2.3	Materials and methods	24
2.3.1	Specimens	24
2.3.2	Model assembly	24
2.3.3	Load cases	28
2.4	Results	31
2.5	Discussion	38
2.6	Acknowledgements	41

CHAPTER 3. VIRTUAL RECONSTRUCTION AND PREY SIZE PREFERENCE OF THE MID CENOZOIC THYLACINID, *NIMBACINUS DICKSONI* (THYLACINIDAE, MARSUPIALIA)

3.1	Abstract	44
3.2	Introduction	45
3.3	Materials and methods	50
3.3.1	Specimens	50
3.3.2	Digital reconstruction of <i>Nimbacinus dicksoni</i>	50
3.3.3	Finite element models	54
3.3.4	Modeling masticatory muscle forces	54
3.3.5	Restraints, loading conditions and scaling	55
3.4	Results	57
3.5	Discussion	65
3.6	Acknowledgements	67

**CHAPTER 4. NEW INSIGHT FROM THE OLD: USING STABLE ISOTOPES
TO ASSESS MARSUPIAL CARNIVORE VULNERABILITY TO
ANTHROPOGENIC IMPACTS**

4.1	Abstract	70
4.2	Introduction	72
4.3	Materials and methods	75
4.3.1	Specimens and tissues	75
4.3.1.1	Marsupial carnivores	75
4.3.1.2	Potential prey species	75
4.3.2	Sample acquisition	78
4.3.3	Sample preparation	78
4.3.3.1	Bone	78
4.3.3.2	Hair and vibrissae	79
4.3.3.3	Skin	79
4.3.4	Sample analysis	80
4.3.5	Controlling animal $\delta^{13}\text{C}$ values for the Suess effect	80
4.3.6	Inter-tissue isotopic spacing	81
4.3.7	Niche overlap and sex-biased variation	81
4.3.8	Diet reconstruction and prey size	82
4.3.8.1	Diet-tissue fractionation	84
4.3.9	Long-term trends in marsupial carnivore stable isotope values	85
4.4	Results	86
4.5	Discussion	97
4.5.1	Diet and prey size of the thylacine	98
4.5.2	Niche overlap among sympatric marsupial carnivores	100
4.5.3	Sexual dimorphism and resource use	103
4.5.4	Long-term change in stable isotope ratio of thylacines	104
4.5.5	Long-term change in $\delta^{13}\text{C}$ of extant marsupial carnivores	104
4.5.5.1	Movement of Tasmanian devils from open to	

	closed environments	105
	4.5.5.2 Woody plant invasion	106
4.5.6	Long-term change in $\delta^{15}\text{N}$ of extant marsupial carnivores	107
4.5.6.1	Diet/trophic variability	107
4.5.6.2	Environmental influences	109
4.5.6.3	Verifying cause of temporal $\delta^{15}\text{N}$ shift	110
4.6	Conclusion	110
4.7	Acknowledgements	111

CHAPTER 5. CONCLUSION: THE INTERACTION BETWEEN DIET AND EXTINCTION RISK AND THE FUTURE OF AUSTRALIA'S CARNIVOROUS MARSUPIALS

5.1	Key findings of the thesis	114
5.1.1	Thylacine prey size and extinction risk	114
5.1.2	Predatory behaviour of <i>Nimbacinus dicksoni</i>	116
5.1.3	A case for intraspecific competition	116
5.1.4	Ecological responses to prolonged human disturbances	117
5.2	Recommendations for future work	119
5.2.1	Were thylacines individual specialists?	119
5.2.2	Temporal change in predator $\delta^{15}\text{N}$ values: is it a real 'trophic' shift?	120
5.2.3	Caveats to stable isotope studies	120
5.2.4	Looking into the future: the management of critically endangered carnivores	122
	Appendix A	125
	Appendix B	141
	Appendix C	146
	References	155

LIST OF FIGURES

Figure 2.1 Muscle origin and insertion areas and lines of action of muscle forces for skulls of (a) spotted-tailed quoll (b) Tasmanian devil, and (c) thylacine constructed using finite element software (Strand 7, Vers 2.4). 27

Figure 2.2 Bite force reaction in the spotted-tailed quoll, Tasmanian devil and thylacine during a bilateral bite and unilateral carnassial bite for (a) unscaled models and (b) models scaled to the same surface area and muscle force as the thylacine. 32

Figure 2.3 Muscle recruitment of the spotted-tailed quoll and Tasmanian devil to produce the same bite force as the thylacine in models scaled to a uniform surface area. 33

Figure 2.4 Dorsal view of the stress distribution in scaled heterogeneous models with a uniform bite force during a bilateral canine bite (a–c) and a unilateral carnassial bite (e, f). Dorsal views of models are displayed for the spotted-tailed quoll (a, d), Tasmanian devil (b, e) and thylacine (c, f). von Mises (VM) stress was measured from the anterior to posterior along the mid-sagittal plane for a bilateral canine bite (g) and a carnassial bite (h). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle). 35

Figure 2.5 Lateral view of the stress distribution in heterogeneous models with a uniform bite force in scaled models during a bilateral canine bite (a–c) and a unilateral carnassial bite (d–f). von Mises (VM) stress was measured from the anterior to posterior along the jaw for a bilateral canine bite (g) and a carnassial bite (h). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle). 36

Figure 2.6 Stress distribution for extrinsic loads. Comparison of the distribution of von Mises (VM) stresses in MPa in models scaled to the same surface area during four extrinsic loads: an axial twist (a–c), pullback (d–f), lateral shake (g–i) and dorsoventral (j–l). Species from left to right are the spotted-tailed quoll (a, d,g, j), Tasmanian devil (b, e, h, k) and thylacine (c, f, i, l). VM stress was measured from the anterior to the posterior along the mid-sagittal plane for each species during an axial twist (m), pullback (n), lateral shake (o) and dorsoventral (p). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle). White areas indicate VM stress exceeds the scale maximum (2.5 MPa) in those areas. 37

Figure 2.7 Volume ratio of cortical to cancellous bone in the mandible and cranium of the spotted-tailed quoll, Tasmanian devil and thylacine. 38

Figure 3.1 Digital reconstruction of *Nimbacinus dicksoni*. Original (grey) and reconstructed 3D (yellow) in (a), lateral view; (b), dorsal view; (c), Pre-processed FE model of *N. dicksoni*, showing jaw musculature represented by trusses. 53

Figure 3.2 Position of nodes selected at equidistant points along the (a) mid-sagittal plane, (b) zygomatic arch, and (c) mandible to measure distribution of VM stress for each loading case. 57

Figure 3.3 Stress distribution along mandible during a bilateral canine bite. Stress distribution in scaled homogeneous models with bite force scaled based on theoretical body mass during a bilateral canine bite for (a) northern quoll, (b) spotted-tailed quoll, (c) Tasmanian devil, (d) *Nimbacinus dicksonim*, and (e) thylacine. (f) Distribution of von Mises (VM) stress was measured from anterior to posterior along the jaw. 60

Figure 3.4 Stress distribution along mid-sagittal crest during a bilateral canine bite. Stress distribution in homogeneous models with bite forces scaled based on predicted body mass during a bilateral canine bite for (a) nothern quoll, (b) spotted-tailed quoll,

(c) Tasmanian devil, (d) *Nimbacinus dicksoni*, and (e) thylacine. (f) Distribution of von Mises (VM) stress was measured from anterior to posterior along the mid-sagittal plane.

61

Figure 3.5 Stress distribution along mandible for extrinsic loads. Stress distribution in scaled homogeneous models with bite force scaled based on predicted body mass during a (a), (e), (i), (m), (q) lateral shake, (b), (f), (j), (n), (r) axial twist, (c), (g), (k), (o), (s) pullback, and (d), (h), (l), (p), (t) dorsoventral. Species compared were (a-d) northern quoll, (e-h) spotted-tailed quoll, (i-l) Tasmanian devil, (m-p) *Nimbacinus dicksoni*, and (q-t) thylacine. Distribution of von Mises (VM) stress was measured from anterior to posterior along the jaw for a (u) lateral shake, (v) axial twist, (w) pullback, and (x) dorsoventral.

62

Figure 3.6 Stress distribution along mid-sagittal crest for extrinsic loads. Stress distribution in scaled homogeneous models with bite force scaled based on predicted body mass during a (a), (e), (i), (m), (q) lateral shake, (b), (f), (j), (n), (r) axial twist, (c), (g), (k), (o), (s) pullback, and (d), (h), (l), (p), (t) dorsoventral. Species compared were (a-d) northern quoll, (e-h) spotted-tailed quoll, (i-l) Tasmanian devil, (m-p) *Nimbacinus dicksoni*, and (q-t) thylacine. Distribution of von Mises (VM) stress was measured from anterior to posterior along the jaw for a (u) lateral shake, (v) axial twist, (w) pullback, and (x) dorsoventral.

63

Figure 4.1 Relationship between stable isotope signatures of thylacines (circle symbol; black = bone collagen, white = keratin) and potential prey species (square symbol). Mean ($\pm$ SD) stable isotope values are present for each species sampled between 1835 and 1964 (sample size in brackets). The stable isotope values have not been adjusted for trophic enrichment. Refer to Appendix A4 for average C/N ratios of each potential prey species.

77

Figure 4.2 Bivariate plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of the thylacine ($n=34$; red), Tasmanian devil ($n=12$; dark blue), spotted-tailed quoll ($n=10$; light blue) and eastern

quoll ($n=8$; yellow) dating from 1803 to 1936 in Tasmania. Symbols represent tissue analysed; bone collagen (square), hair (circle) and vibrissa (triangle) samples. 87

Figure 4.3 Bivariate plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ($\pm\text{SD}$) of thylacine bone collagen ($n=33$; black circle symbol) and skin collagen of potential prey species (square symbol) grouped into four isotopically distinguished categories; Carnivores (blue), Insectivores (purple), Herbivores A (red square) and Herbivores B (yellow square). The polygon describes the mixing space of potential prey groups. Prey stable isotope values have been enriched by $1.13\pm0.60\text{‰}$ for $\delta^{13}\text{C}$ and $3.89\pm0.80\text{‰}$ for $\delta^{15}\text{N}$. The proximity of the mixture (thylacine) to the source (prey groups) indicates the greatest contribution to the diet. Posterior contributions are measured for each prey group from the Bayesian mixing model, MixSIR. 88

Figure 4.4 Comparison of the $\delta^{13}\text{C}$ values for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll from 1830 to 2012, Tasmania. Values were corrected with an atmospheric model calculation (Long *et al.* 2005). Blue squares with no dashed treadline and pink triangles with dashed treadline indicate values for collagen (bone) and keratin (hair and vibrissa) from specimens, respectively. 91

Figure 4.5 Comparison of the $\delta^{15}\text{N}$ values for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll from 1830 to 2012, Tasmania. Circles and triangles indicate values for collagen (bone) and keratin (hair and vibrissa) from specimens, respectively. 92

Figure 4.6 Box and whisker plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of (a, b) Tasmanian devil, (d, e) spotted-tailed quoll and (g, h) eastern quoll specimens collected between 1930 and 2012 in Tasmania. Specimen $\delta^{13}\text{C}$ values were combined for years dating 1803-1939, 1940-1959, 1980-1999 and 2000-2012. Values were corrected for the Suess effect and inter-tissue differences (see section 4.3.5 and 4.3.6, respectively). Box shows median,

lower and upper quartiles, whiskers cover 10th±90th percentile range, and circles are statistical outliers. Overall specimen sample size is displayed in brackets. For tissue type see Appendix A1. Significant differences between groups are denoted in *, where *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$ for a 1. Geographic distribution of tissue samples collected of (c) Tasmanian devil, (f) spotted-tailed quoll and (i) eastern quoll based on museum records. Samples are categorised for each species by year of collection. Locality records were unavailable for most historic samples. 95

LIST OF TABLES

Table 2.1 Values used to assign eight material properties to brick elements in heterogeneous models.	26
Table 2.2 Force used for each jaw muscle division in unscaled models of the spotted-tailed quoll, Tasmanian devil and thylacine. These were calculated using muscle mass proportions from a dissected Virginia opossum (Turnbull 1970).	27
Table 2.3 Volume (mm ³) and percentage volume (%) of each material property in the skull of the spotted-tailed quoll, Tasmanian devil and thylacine.	31
Table 3.1 Muscle forces used for each jaw muscle division in unscaled intrinsic models of the northern quoll, spotted-tailed quoll, Tasmanian devil, <i>Nimbacinus dicksoni</i> and thylacine. These were calculated using muscle mass proportions from dissected Virginia opossum (Turnbull 1970). Muscle forces were scaled for a bilateral canine bite by multiplying the muscle force by the ratio between bite force estimated using body mass regressions and maximum bite force estimated from the unscaled FEM.	55
Table 3.2 Predicted body mass (kg) calculated using the regression equation for dasyuromorphians provided by Myers (2001) based on lower molar row length. Temporalis and masseteric muscle forces (N) were calculated based on cross-sectional area (Thomason 1991).	57
Table 3.3 Maximum bite forces (N) for unscaled homogeneous models during a bilateral canine for the northern quoll, spotted-tailed quoll, Tasmanian devil, <i>Nimbacinus dicksoni</i> and the thylacine.	58

Table 4.1 Mann-Whitney *U*-test for comparisons in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between sexes (M = male, F = female) for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll. All specimens were wild adults collected from Tasmania at any time following European settlement. Mean ($\pm$ SD) $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for each sex are provided. Comparisons between sexes were completed for bone collagen, keratin (hair and vibrissae) and both tissue types combined (corrected for inter-tissue differences between vibrissae and bone collagen) for species where there was a sufficient sample size. 88

Table 4.2 Spearman correlation coefficient for $\delta^{13}\text{C}$ values for thylacines (1852-1930), Tasmanian devil (1921-2012), spotted-tailed quoll (1914-2008) and eastern quoll (1921-2012) in Tasmania that were uncorrected and uncorrected with an atmospheric model (Long et al. 2005). 93

Table 4.3 Spearman correlation coefficient for $\delta^{15}\text{N}$ for the thylacine (1852-1930), Tasmanian devil (1921-2012), spotted-tailed quoll (1914-2008) and eastern quoll (1921-2012) in Tasmania. 93

Table 4.4 Post-hoc Tukey Test comparing $\delta^{13}\text{C}$ values for Tasmanian devil, spotted-tailed quoll and eastern quoll specimens collected in Tasmania during four time periods: 1900-1959, 1960-1979, 1980-1999 and 2000-2012. The $\delta^{13}\text{C}$ values for one Tasmanian devil specimen (QVM:1984:1:0260) and two eastern quoll specimens (65344 and A1516) were identified as outliers based on box and histogram plots and were excluded from the analysis. *P* values and sample sizes are listed for each comparison between grouped decades. 96

Table 4.5 Post-hoc Tukey Test comparing $\delta^{15}\text{N}$ values of Tasmanian devil, spotted-tailed quoll and eastern quoll specimens collected in Tasmania at four time periods: 1900-1959, 1960-1979, 1980-1999 and 2000-2012. *P* values and sample sizes are listed for each comparison between grouped decades. 96

LIST OF APPENDICES

Appendix A1 Specimen records for all carnivorous marsupials included in the study, including the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll.

Information is provided on the sex (M = male, F = female, U = unknown), year of death/acquisition, locality and institution where each specimen. 125

Appendix A2 List of specimens representing potential prey species of the thylacine.

Skin was analysed for all prey specimens. The list includes specimen records including sex (male=M; female=F, unknown=U), year of death/acquisition and locality. All specimens were held at the Queen Victoria Museum and Art Gallery. 137

Appendix A3 Diet and body mass of potential prey species of the thylacine included in this study. Average body mass are given for males (M) and females (F) where available.

139

Appendix A4 Sample size, mean $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ($\pm\text{SE}$), and C/N ratios (mean $\pm$ SD) of museum-preserved skin of potential prey species. All specimens were collected between 1835 and 1964. 140

Appendix B1 Raw stable isotope data for bone collagen, skin collagen, vibrissa and hair samples collected opportunistically from deceased wild adult Tasmanian devils (n=10). Specimen sex (M = male, F = female, U = unknown), stable isotope ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) and C/N ratios are provided. 142

Appendix B2 Stable carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) isotope ratios of paired (a) bone and skin, (b) bone and whisker, (c) bone and hair, (d) skin and whisker, (e) skin and hair, and (f) vibrissa and hair from ten wild adult Tasmanian devils. Lines are drawn between tissue stable isotope values from the same individual. 144

Appendix B3 Wilcoxon matched pairs signed rank test comparing inter-tissue isotopic spacing for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ from wild Tasmanian devils ($n=10$). Valid N excludes paired tests that have the same stable isotope values. 145

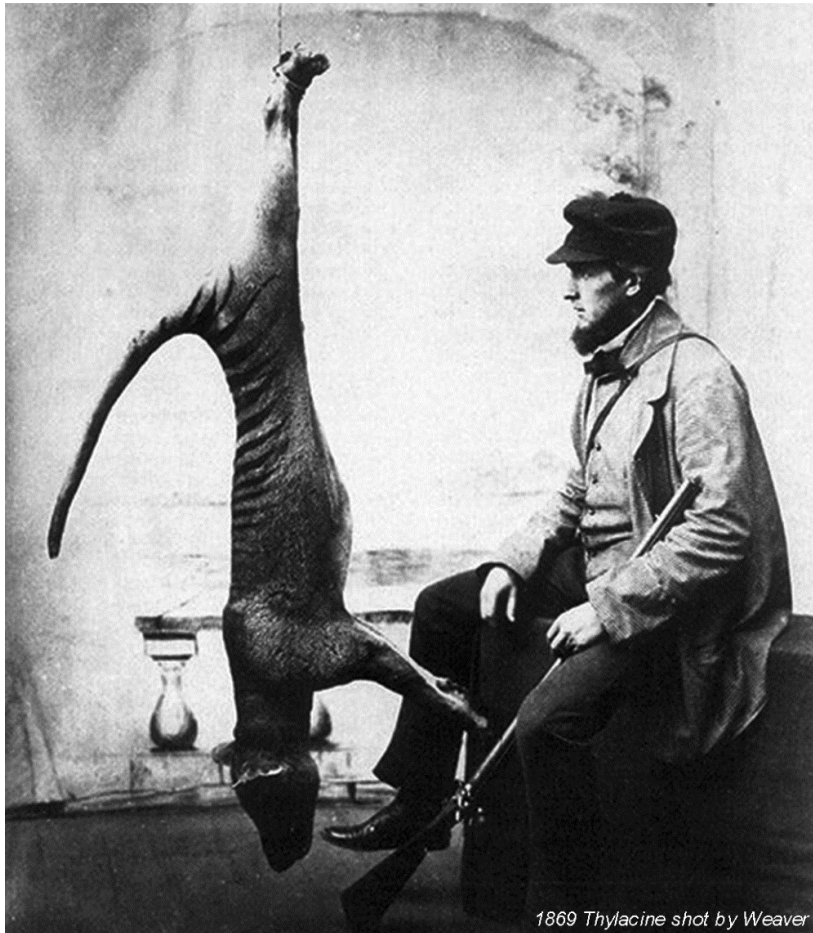
Appendix C1 Literature search for specimen-specific plasma fractionation factors in mammals, sorted by environment, infraclass and trophic level. The animal section describes the species common and scientific name, environment (T = terrestrial, M = marine), Infraclass (M = marsupial, P = placental) and tissue. The discrimination factor section describes the discrimination factors ($\Delta^{13}\text{C}$ and $\Delta^{15}\text{N}$) calculated in each case. Adapted from Caut *et al.* (2009). 146

Appendix C2 Literature search for specimen-specific blood and red blood cell fractionation factors in mammals, sorted by environment, infraclass and trophic level. The animal section describes the species common and scientific name, environment (T = terrestrial, M = marine), Infraclass (M = marsupial, P = placental) and tissue (red blood cells, blood). The discrimination factor section describes the discrimination factors ($\Delta\delta^{13}\text{C}$ and $\Delta\delta^{15}\text{N}$) calculated in each case. Adapted from Caut *et al.* (2009). 149

Appendix C3 Previously published isotopic fractionation between bone collagen and diet of terrestrial carnivorous mammal communities and species. 152

Chapter 1

INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS



"The history of a species can invariably be interpreted from different angles to reveal new facets and is always more complicated."

D.M.J.S. Bowman, 2001

1.1 CHAPTER OUTLINE

This thesis assesses the diet of Australia's iconic marsupial carnivore, the thylacine (*Thylacinus cynocephalus*) and its implications for their extinction in Tasmania. Much of our understanding of their feeding and predatory behaviour comes from anecdotal accounts, which may not fully represent the behaviour of the species. Chapter 1 will present a review of causal factors associated with large mammal extinctions, followed by a review of the history of the family Thylacinidae. Conflicting evidence regarding the feeding ecology of the thylacine will be identified and provides the incentive for the research program described in the following chapters of this thesis. A brief summary of each chapter is provided and its relevance in identifying biological traits of the thylacine and other large carnivores that are associated with high extinction risk.

1.2 RATIONALE

Not all species are at an equal risk of extinction. Predicting the susceptibility of a population or species to extinction is extremely complicated and requires consideration of their internal biological attributes (intrinsic traits e.g. body size, diet, reproductive output) and exposure to external threats (extrinsic traits e.g. land-use, over-exploitation, introduced species, climate change) (Diamond 1984; Thomas *et al.* 2004; Schwartz *et al.* 2006; Hannah *et al.* 2007; Pressey *et al.* 2007). Identifying traits attributing to increased extinction risk in a given species can be difficult to untangle as (i) different lineages are threatened by different external threats, (ii) different biological attributes are linked to each external threat, and (iii) compounding effects can occur when multiple factors are involved (Owens and Bennett 2000). For proactive conservation planning to be successful and protect biodiversity, we need a better grasp of how specific intrinsic traits of a species may influence their response to anthropogenic impacts (Murray *et al.* 2011).

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

Palaeontologists and ecologists are working towards deciphering the cause of species declines and extinctions, and with this knowledge, facilitate the development of a predictive conservation science (McKinney 1997). The inclusion of historic data to achieve this goal relies on the assumption that the sensitivity of species to extinction (whether taxonomically or biologically) is consistent through time (Bromham *et al.* 2012). However, the driving forces for past extinctions may not necessarily relate to present species loss due to the changing nature of threatening processes or increased resilience of community assemblages to previously conceived threats (Bromham *et al.* 2012). The decline of a species is thus context-dependant and relies on their unique suite of intrinsic traits (Olden *et al.* 2008; Davidson *et al.* 2009). By focusing on contemporary extinctions to learn more about present-day extinction risk factors, ecologists limit potential threats to a narrow time window that may not fully capture changing patterns of extinction risk into the future (Chiba and Roy 2011; Ezard *et al.* 2011). A combined understanding of historic and contemporary extinctions that consider both intrinsic traits and extrinsic threats will be necessary to describe the complete picture of extinction risk and overcome present challenges for species conservation (McKinney 1997; Murray *et al.* 2011).

1.3 THE LINK BETWEEN MAMMAL BODY SIZE, COMPETITION AND VULNERABILITY

1.3.1 Body size

A mounting body of evidence reveals that body size is directly linked to extinction probability (Belovsky 1987; Baillie *et al.* 2004). The accumulation of multiple biological and anthropogenic processes has a greater impact on larger mammals (Cardillo *et al.* 2005). As home range size increases with body mass, deforestation increases the likelihood of large mammal extinctions due to habitat forestation and increased encounters with humans (Woodroffe and Ginsberg 1998; Cullen Jr *et al.* 2000; Bodmer *et al.* 2002). People are usually less tolerant of large-bodied species, and

these species are more likely targets for persecution and exploitation (Bodmer *et al.* 2002; Jerozolinski and Peres 2003; Cardillo *et al.* 2005). The recovery of large-bodied mammal populations from human hunting is slow if they possess biological traits associated with large size, such as low population density, slow life history and small geographic range size (Damuth 1981; Purvis *et al.* 2000; Bodmer *et al.* 2002). These traits explain close to 50% of variation in extinction risk for large mammalian carnivores (Purvis *et al.* 2000).

Considering the high extinction risk among large mammal predators, many ecologists have asked themselves "What selective pressure influences predator body size?" Optimal foraging theory predicts that animals should forage in a way that maximises their net energy intake per time (Schoener 1971). The body size of terrestrial vertebrate predators is typically associated with increases in mean prey size (Rosenzweig 1966; Gittleman 1985; Vézina 1985; Carbone *et al.* 1999), maximum prey size (Woodward *et al.* 2005) and prey diversity (Schoener 1969; Wilson 1975; Gittleman 1985; Cohen *et al.* 1993). Small carnivores tend to specialise in small prey relative to their body size as there is greater difficulty, time and energy expended in finding, pursuing and subduing larger animals (Griffiths 1980). Specialising in smaller prey also reduces the risk of predator injury, the prey escaping or a combination of these (Griffiths 1980). As the energy requirements of an individual increases with body mass (Hemmingsen 1960; Kleiber 1961), large predators require large prey relative to their own body size to achieve a higher net energy gain (Carbone *et al.* 2007). To maintain high rates of energy intake, terrestrial mammalian carnivores exceeding 21.5 kg in body mass need to rely on prey approaching or exceeding their own body mass (Carbone *et al.* 1999). When large prey are difficult to catch, weaker individuals may be selectively hunted by large mammalian predators (Temple 1987). Pack hunting allows larger prey to be caught compared with solitary hunting (Zimen 1976). This remarkable relationship in body size between predator and prey species has important implications in the evolution, function and structure of food webs (Brose *et al.* 2006).

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

The feeding ecology of a population or species can be described as a specialist or generalist. Specialisation is the use of a relatively narrow subset of resources (food and habitat) compared with other individuals, populations or species (Colles *et al.* 2009). Specialists are able to use resources more efficiently but this benefit comes at a cost; they are more limited in their capacity to adapt to changes in the environment than generalists (Colles *et al.* 2009). If alternative prey is available, a generalist predator might switch to a more profitable resource and experience higher levels of fitness than if it captured the original prey (Brodie III and Brodie Jr 1999). The extinction of many large carnivores has been attributed to their highly specialised diet (Van Valkenburgh and Hertel 1998; Leonard *et al.* 2007). The correlation between specialisation and greater risk of extinction is relevant for ecosystems today that are under excessive anthropogenic pressure.

1.3.2 Competition

Competition is an individual's use of a resource that thereby limits the availability of the resource to others (exploitation competition) or an individual's agnostic behaviour that excludes others from a resource (interference competition) (Glen and Dickman 2005). This can occur within or between species (Begon *et al.* 1990). Competition is most likely to influence species interactions at a guild level (Begon *et al.* 1990) and may lead to reduced densities (Mac Nally 1983; Korpimäki 1987; Petren and Case 1996) or extinction (Pimm 1992) of subordinate species. A guild is a group of species that exploits the same subset of environmental resources in a similar way (Root 1967) and are both sympatric and synchronous in foraging activity (Mac Nally 1983; Mac Nally and Doolan 1986). Large predators more easily co-exist in the same area than smaller predators as they are better able to avoid other predators and have access to a wider food and habitat range due to their ecological dominance, large geographic range and greater mobility (Brown and Maurer 1986).

Adaptive shifts in prey anti-predator defenses and predator preferences enhance community-level stability and may help facilitate the coexistence of predators (Kondoh 2007). For example, prey may more actively avoid predators that are present in high abundance, allowing low density predators to have greater access to 'defense switching' prey (Sih *et al.* 1998). Similarly, generalist predators tend to prefer more abundant prey species thereby preventing prey populations from becoming drastically reduced or overly abundant (Murdoch 1969). Prey switching has been documented in a range of predators (Greenwood and Elton 1979; Gendron 1987; Allen and Greenwood 1988) and allows large complex communities to form when multiple prey species are involved (Drossel *et al.* 2001; Kondoh 2007).

1.4 CONSEQUENCES OF APEX PREDATOR EXTINCTIONS

Apex predators, also known as top predators, occupy the top trophic position in a community and are often large-bodied, specialised hunters (Ritchie and Johnson 2009). They have been ubiquitous worldwide for many millions of years and are one of the most influential drivers of trophic structure and biodiversity in terrestrial ecosystems (Sergio *et al.* 2008). Apex predators provide resources to other species that would otherwise be difficult or impossible to obtain. For example, apex predators provide carrion for scavengers (Wilmers *et al.* 2003a; Wilmers *et al.* 2003b) and support safer breeding grounds for other species by deterring predators that pose a greater threat (Bogliani *et al.* 1999; Haemig 2001; Quinn and Kokorev 2002). Large mammalian predators exert a controlling influence over species at the next lower trophic level by regulating or limiting the number of prey, with indirect cascading effects down the trophic ladder (Paine 1980). They can also influence the behaviour of prey and competing species (Fortin *et al.* 2005; Ritchie and Johnson 2009). Conservation strategies commonly prioritise the preservation of native apex predators to achieve broader ecosystem-level benefits (Sergio *et al.* 2006). Additionally, the association of apex predators with biologically diverse environments makes them a good indicator species (Sergio *et al.* 2008).

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

The decline of apex predators is particularly detrimental; their loss carries broader implications on ecosystem structure and biodiversity due to indirect effects down the food chain. Secondary extinctions may follow the loss of an apex predator through two mechanisms: (i) mesopredator release, defined as a dramatic increase in the abundance of smaller predators (mesopredators) normally controlled by the top predator, causing sudden changes to the structure of ecosystems (Prugh *et al.* 2009; Wallach *et al.* 2009), or (ii) prey release, defined as a dramatic increase in herbivore communities, leading to overexploitation of vegetation (Berger *et al.* 2001a; Terborgh *et al.* 2001). Mesopredator release has been documented in over 60 systems worldwide (Ritchie and Johnson 2009) and results in higher predation of small prey species (Crooks and Soulé 1999). Island communities are particularly sensitive to outbreaks of mesopredator populations (Courchamp *et al.* 2001). The removal of large predators may also allow smaller predators to switch their diet towards larger prey (Dickman 1988). Additionally, nutrient cycling is indirectly affected by the loss of top predators by altering prey abundance, composition and foraging efficiency (Power 1990; Werner and Peacor 2003).

The recent rapid decline of fauna marks the beginning of the sixth mass extinction over Earth's history (Wake and Vredenburg 2008). Human pressures, both direct and indirect, threaten the survival of apex predators in many regions of the world; from wild dogs (*Lycaon pictus*) and lions (*Panthera leo*) in Africa, wolves (*Canis lupus*) in Asia, North America and Europe (Berger *et al.* 2001a; Gittleman *et al.* 2001) to Tasmanian devils (*Sarcophilus harrisii*), spotted-tailed quolls (*Dasyurus maculatus*) and eastern quolls (*Dasyurus viverrinus*) in Australia (Jones *et al.* 2003). Astonishingly, Australia accounts for one third of all contemporary mammal extinctions over the past 200 years, despite the relatively low human population density (Groombridge 1993). Marsupial species have been lost at a greater rate than any other mammal in Australia. At least ten species and six subspecies of Australian marsupials have become extinct following European settlement and many more are now at high risk of extinction (Morris 1996). Insight into the history of mammalian predator extinctions in Australia will be critical to

understand present environmental problems and learn from past experiences and events (Dovers 2000).

1.5 HISTORY OF THYLACINIDAE

For the purposes of this study, marsupial carnivores refer to species of the lineage Dasyuroidea, including the families Dasyuridae and Thylacinidae, which predominantly consume vertebrate prey. This definition excludes all small insectivorous species. The extinct marsupial carnivore, the thylacine (*Thylacinus cynocephalus*) is the most well known representative of Thylacinidae. It was "an animal of a truly singular and novel (novel) description" (Paterson 1805), being the only marsupial other than the water opossum (*Chironectes minimus*) to have a pouch for both sexes (Beddard 1891; Galliez *et al.* 2009). The species is a classical example of convergent evolution due to its close resemblance to placental predators such as tigers and wolves (Paddle 2000). This unusual wolf-like marsupial has become a topic of great scientific curiosity and is now a household name due to the tragic history of its extinction.

The fossil history of Thylacinidae in Australasia dates back some 23 million years, and has revealed surprising diversity (Muirhead and Wroe 1998). Twelve fossil species from this family are now known and are separated into nine genera; *Badjcinus*, *Maximucinus*, *Muribacinus*, *Mutpuracinus*, *Ngamalacinus*, *Nimbacinus*, *Thylacinus*, *Tjarrpecinus* and *Wabulacinus*. The smallest known thylacinid, *Mutpuracinus archibaldi* weighed 1.1 kg and the largest, *Thylacinus megiriani* weighed over 57 kg (Wroe 2001; 2002).

Differences in body size among thylacinids may have facilitated partitioning of prey species in regions where they likely co-existed. All members of Thylacinidae, except the thylacine, became extinct by the Pleistocene (Archer *et al.* 1999). Mummified thylacine remains found in caves of Western Australia reveal the first traces of modern thylacines, dated between four and five million years old (Lowry and Merrilees 1969). The species was once widely distributed across continental Australia but was wiped out from the mainland around 3, 000 years ago and possibly earlier in New Guinea (Smith

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

1982). This coincides with the local extinction of Tasmanian devils, which persisted on mainland Australia until around 3, 000 to 4, 000 years ago (Brown 2006).

Aboriginal hunting, changed land use patterns, climate change and the presence of dingoes (*Canis lupus dingo*) have been linked to the extinction of the thylacine on the continent's mainland (Wroe *et al.* 2007; Letnic *et al.* 2012). Dingoes were introduced into Australia by Aboriginal people around 5, 000 years ago (Savolainen *et al.* 2004; Wroe *et al.* 2007; Letnic *et al.* 2012). The highly generalised diet of dingoes gave them an edge over mainland thylacines and they likely outcompeted thylacines for shared food resources (Wroe *et al.* 2007). Dingoes and male thylacines were similarly sized, while female thylacines were much smaller (Letnic *et al.* 2012). Potential killing of female thylacines by dingoes would have reduced the reproductive output of thylacine populations. Expansion of the human population on the mainland about 4, 000 years ago, together with improved hunting technology, increased hunting pressure on large vertebrates including Tasmanian devils and thylacines (Johnson and Wroe 2003). A small population of thylacines persisted on the remote island of Tasmania where there were no dingoes. No more than 3, 000 thylacines were estimated to be present in Tasmania by the time the British colonised Australia in 1789 (Guiler and Godard 1998).

At the time of European settlement, Tasmania's marsupial carnivore guild consisted of the thylacine and three dasyurids: the Tasmanian devil, spotted-tailed quoll and eastern quoll (Jones and Barmuta 2000). Thylacines were distributed throughout the island, apart from the southwest (Paddle 2000). Their numbers were thought to have declined rapidly after 1905, though reports as far back as the 1850's suggest the population was in decline. Despite the thylacine's obvious decrease in abundance, the species did not receive official protection from the Tasmanian government until two months before the last known captive animal died in 1936. Other marsupial carnivores face extinction threats. The Tasmanian devil experienced dramatic population declines in the 1850's, 1920's and late 1990's (Guiler 1992; Bradshaw and Brook 2005). The most recent collapse in Tasmanian devil numbers has been attributed to a fatal contagious cancer

that has spread across their range (Jones *et al.* 2004a). The last known mainland population of eastern quolls were sighted in the 1960's and are believed to be extinct (Nathan 1975; Morris 1996). The range of Eastern quolls has reduced by 50-90% since European colonisation and currently has a widespread but patchy distribution in Tasmania (Maxwell *et al.* 1996). A few small pockets of spotted-tailed quolls are present in high rainforest areas of Victoria and northern Queensland (Mansergh 1983). The decline of quoll species is slower in Tasmania due to the absence of red foxes (*Vulpes vulpes*) until recently in the region (Oakwood 1997; Jones *et al.* 2003). Understanding the ecology of the thylacine and driving forces behind their extinction may aid in the management of remaining marsupial carnivores that are in danger of extinction.

1.6 EXTINCTION RISK IN THE THYLACINE

Growing scientific evidence reveals a complex tapestry of forces involved in the decline and extinction of the thylacine in Tasmania, yet further investigation is required to tease apart these different influences. Assessing extinction vulnerability in the thylacine has faced several difficulties, including incomplete or inaccurate information about their ecology (Guiler 1985) and phylogeny (Thomas *et al.* 1989; Krajewski *et al.* 1997; Muirhead and Wroe 1998; Miller *et al.* 2009). Despite these issues, it is generally agreed that multiple factors have been involved in their decline (Guiler 1985; Paddle 2000; Prowse *et al.* 2013). Evidence for potential risk factors relating to the extinction of the thylacine in Tasmania is detailed in the following section.

1.6.1 Diet and prey size

Thylacines were by far the largest marsupial carnivore to survive up to recent times; with a head to tail length of up to 2.7 metres (Heberle 2004) and average body weight of 30 kg (Watts 1993; Paddle 2000). Based on their dentition and biochemical information, thylacines were most certainly hypercarnivorous (Nedin 1991; Wroe *et al.* 1998; Wroe

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

et al. 2008; Figueirido and Janis 2011), which means having a diet primarily consisting of meat (Holliday and Stepan 2004). They do not have the dental features associated with bone consumption and scavenging. A striking transition in prey selection occurs at a predator mass of about 21.5 kg, with those species below the threshold taking prey of <45% their mass and those above taking prey of >45% their mass (Carbone *et al.* 1999). As a large-bodied hypercarnivore, relying on small prey would have been energetically constraining for thylacines: their food may have been inadequate to support them unless small prey were abundant. There were very few large (>13 kg) native animals in Tasmania that would have allowed thylacines to meet their metabolic requirements. This included the forester kangaroo (*Macropus giganteus*; 25-66 kg, Strahan 1995), common wombat (*Vombatus ursinus*; 20-40 kg, Cronin 1991), red-necked wallaby (*Macropus rufogriseus*; 11-27 kg, Strahan 1995) and extinct Tasmanian emu (*Dromaius novaehollandiae diemenensis*; ~40 kg, Heupink *et al.* 2011). Sheep (*Ovis aries*) were another large herbivore that may have also been preyed upon by thylacines (Paddle 2000). Whether or not thylacines were capable of taking down large prey species remains a contentious subject. Evidence of thylacine diet are based on historical accounts, morphology and fossil remains of associated prey.

Anecdotal accounts: Anecdotal evidence (Paddle 2000) and the only known Aboriginal story of the thylacine (Owen 2003) suggest that thylacines may have taken large prey species such as the forester kangaroo and Tasmanian emu. But few naturalists were present to record the thylacine's foraging behaviour in Tasmania and were very difficult to study due to their low densities, nocturnal activity and wariness of humans (Paddle 2000). Many of the accounts that do exist are thought to be derived from unreliable or biased sources formed from prejudices towards the species.

Around 30 sheep were introduced into Tasmania in 1803, and their numbers rapidly increased to 172, 000 by 1819 (Paddle 2000). Official reports by the Tasmanian sheep industry did not consider thylacines as a threat to livestock (Paddle 2000). There are only six published accounts of thylacines preying on adult sheep, the last of which was

reported 15 years prior to the government bounty scheme initiated in 1888 (Paddle 2000). The intention of the bounty was to provide predator-free environments for livestock and native game used for hunting (e.g. wallabies, wild ducks, quail and common pheasants) (Paddle 2000). Thylacines that were killed as part of the government bounty scheme were mostly from areas outside of grazing lands (Davies 1965). Prowse *et al.* (2013) suggested that most thylacines were unlikely to have been contact with sheep during the late 1800's and possibly earlier.

Stomach contents: The only direct evidence of the thylacine's diet in the wild is based on the stomach contents of thylacines killed. In 1805, Governor William Paterson described the stomach contents of a dissected male thylacine as "filled with a quantity of kangaroo weighing 5 lbs" (Paterson 1805). A few years later, another dissected male thylacine revealed partly digested remains of a short-beaked echidna (*Tachyglossus aculeatus*) (Harris 1808) but the absence of oral accounts of predation on monotremes puts to question the dietary contribution of these small insectivorous mammals (Paddle 2000).

Morphology: Features such as low rates of canine tooth wear and fracture suggest that thylacines relied on prey ranging from <1 kg to 5 kg (Jones and Stoddart 1998). Though, their large body size and wide gape may have allowed them to catch larger prey species (Johnson and Wroe 2003). The long narrow rostrum of thylacines (Jones 2003) may be a morphological disadvantage by eliciting size-selective prey selection (Slater *et al.* 2009). Larger prey can be killed when hunted communally and thereby counteract the morphological constraints of a predator (McKenzie 1990; Husseman *et al.* 2003).

Cave deposits: Thylacines were thought to use caves as lairs and have been associated with prey found in sub-fossil cave deposits in mainland Australia (Case 1985). These prey ranged in size from 1 kg to 5 kg. As only smaller prey species may be brought back to a den, they may not represent the full range of prey species killed by thylacines.

1.6.2 Intraguild competition and predation

Competition, intraguild predation or a combination of these may have occurred in Tasmania between thylacines and sympatric marsupial carnivores. Intersexual and interspecific differences in the body size and habitat of these apex predators possibly facilitated prey partitioning during early settlement as is seen in the present-day food web (Jones and Barmuta 1998). The diet of adult Tasmanian devils (mean weight males 8.4 kg, females 5.4 kg) and adult male spotted-tailed quolls (mean weight 3.2 kg) currently consist of larger prey species (Jones and Barmuta 1998). However, the ability of spotted-tailed quolls to climb trees and rock faces allow them to eat a higher proportion of arboreal species than other marsupial carnivores (Jones and Barmuta 2000). Young Tasmanian devils can climb trees and have more avian material in their stomachs than adults (Guiler 1970). Eastern quolls (mean weight males 1.1 kg, females 0.7 kg) and female spotted-tailed quolls (mean weight 1.7 kg) specialise in smaller prey species due to their smaller body mass (Jones and Barmuta 1998). As the largest and most dominant of the three extant marsupial carnivores, adult Tasmanian devils are able to displace both quoll species from carcasses (Jones 1998; Jones and Barmuta 2000). It is unknown whether Tasmanian devils kill quolls (Jones 1998). Current hypotheses suggest that the thylacine preyed on juvenile Tasmanian devils, and adult Tasmanian devils ate thylacine young and scavenged from kills made by the thylacine (Jones 1998), though no anecdotal evidence exist to substantiate these claims. Cranium measures of male thylacines are 13% to 86% larger than that of females in Tasmania and may have allowed them to catch larger animals (Jones 1997).

1.6.3 Home range

The home range of thylacines is estimated to be 50-60 km² based on bounty and northwest Tasmania station records (Guiler and Godard 1998). Prowse (2013) estimated a home range area for thylacines of 25.7 km² based on the allometric relationship between body mass and home range for mammalian carnivores (Kelt and Van Vuren 2001). Species with large home ranges, including large carnivores, are particularly

vulnerable to degradation and edge effects (Woodroffe and Ginsberg 1998). Human-induced environmental changes may have resulted in a reduction in the thylacine's home range size and access to food resources.

1.6.4 Disease and genetic diversity

A debilitating disease originating in the thylacine population on the east coast of Tasmania was first reported in the late 1800's. Paddle (2000) estimated that the disease would have spread to the west coast of Tasmania within six years. The disease was not fatal, but infected animals were likely easier to bait and kill due to their weakened state. Surprisingly, there is no definitive proof of the disease in the wild population (Prowse *et al.* 2013). Mathematical models have identified that disease alone could not account for the extinction of the thylacine in Tasmania (Prowse *et al.* 2013). The genetic variability of the Tasmanian thylacines is thought to be low and therefore may have increased their risk of extinction due to decreased capacity to adapt to environmental change (Menzies *et al.* 2012). Though, this was only based on a small fragment of the mitochondrial DNA control region and 12 museum specimens.

1.6.5 Human population expansion

In carnivores, biological traits explain 80% of variation in extinction risk with high exposure to humans, compared to 45% with limited exposure to humans (Cardillo *et al.* 2004). The biological traits of the thylacine would thus become a more critical determinant of risk as human populations expanded. The small Aboriginal tribes present in Tasmania prior to European settlement likely posed little threat to the thylacine population. The Aboriginal people did not fear thylacines, nor did they hunt thylacines as they did not form part of their diet (Owen 2003). Conversely, the early-colonial generation of Europeans (1826-1846) underwent a dramatic expansion over a 10 year period, increasing from 5, 519 people in 1820 to 24, 279 people by 1830 (Australian Bureau of Statistics 2008; Reynolds 2012). There were 230, 104 people in Tasmania in

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

1936 when the thylacine was officially declared extinct (Australian Bureau of Statistics 2008).

1.6.6 Hunting

The bounty system introduced by the Van Diemen's Land Company, a large pastoral company, in 1830 was designed to remove thylacines on their 40, 500 hectare property in north-west Tasmania (Bowman 2001). In 1888, the Tasmanian government paid £1 for every adult and 10 shillings for every juvenile thylacine. At the time, the award would have been equivalent to half a week's wage (Bulte *et al.* 2003). The government paid out 2, 184 bounties, but many more animals were likely killed than was claimed in the bounty records (Guiler 1961). There were also 3, 482 thylacine pelts sold until 1896, which were a valuable commodity among Europeans (Bulte *et al.* 2003). Live specimens or mothers with pouched young were in high demand by zoos and museums worldwide, and offered greater remuneration than the bounty scheme. The last thylacine was captured in 1933 and sold to Hobart Zoo (Paddle 2000). The government bounty scheme was terminated in 1909, by which time the species was exceedingly rare. Furthermore, the *Animals and Birds Protection Act* was amended in 1928 (Animals and Birds Protection Act Amendment Act 1928) to prevent overexploitation of native animals for the fur trade.

Scientific data relating to the decline of thylacines in Tasmania was first published in 1961 based on bounty payments made between 1874 and 1901 by the Van Diemen's Land Company and payments made between 1888 and 1912 by the Tasmanian government for the rest of the state. Both records show a reduced number of thylacines presented for bounty payment over time, revealing a species increasing in rarity and quickly heading towards extinction (Bowman 2001). Increased hunting pressure would appear to be the obvious extinction culprit but an evaluation of bounty records revealed that hunting for bounty and pelts alone could not account for their extinction (Bulte *et al.* 2003).

Macropod harvesting by humans appeared to have little effect on the thylacine population using population viability analysis (Prowse *et al.* 2013). Prowse *et al.* (2013) argued that their extinction was only possible under extreme circumstances, including high human harvest, low maximum population growth rate and small starting population size.

1.6.7 Habitat loss and modification

The earliest vegetation maps of Tasmania reveal wet sclerophyll forests (tall, open forests) dominating east Tasmania, with temperate rainforests (closed forest) and open grasslands mainly found in western Tasmania (Davis 1940). Coastal heaths extended along the east of the north coast and at the north-west tip of Tasmania. Thylacines were originally found in north-western, central, eastern and south-eastern parts of Tasmania, but not in the south-west (Guiler 1961; Paddle 2000). Based on limited observations and bounty information, thylacines preferred open forests, open woodlands, heathlands and coastal environments. The habitat size of the thylacine population in 1803 is estimated to have been 56, 051 km² (Prowse *et al.* 2013).

Habitat destruction and fragmentation directly reduces niche availability and are particularly threatening to species that are ecologically specialised (Owens and Bennett 2000). From 1820 to 1830, over two million acres were granted for use by the European settlers, including 50, 000 acres devoted to cultivation (Reynolds 2012). By 1935 the total habitat range available to thylacines and potential prey is estimated to have decreased by 17.2% (Prowse *et al.* 2013), including large areas in southeast and northwest Tasmania that had been modified or cleared for homes, agriculture and livestock (Davis 1940). Persistent hunting and land clearing eventually confined the thylacine population to dense rainforests, which are generally considered an unsuitable thylacine habitat (Guiler and Godard 1998; Owen 2003).

1. INTRODUCTION: RATIONALE AND SCOPE OF THE THESIS

Changes in landscape burning practices following European settlement has been proposed as influential in the decline of thylacines (Guiler 1985). Aboriginal people used frequent, low-intensity burning regimes that were usually initiated under conditions when the vegetation was too wet to burn intensely (Marsden-Smedley 1998; Marsden-Smedley and Kirkpatrick 2000; Enright and Thomas 2008). There are occasional accounts of large and high-intensity fires lit by Aboriginal people in Tasmania (Peron 1809). The fire regime shifted following the displacement of Aboriginal people by Europeans to having fewer fires, and occasional regional scaled fires (Marsden-Smedley and Kirkpatrick 2000; Enright and Thomas 2008). There are also extensive historical records up to the 1930's of settlers starting fires when exploring regions and building access tracks (Gowlland and Gowlland 1976; Gilbert 1979; Binks 1980). Changes to traditional Aboriginal burning practices in Australia have resulted in mass vegetation change and the reduction or disappearance of wildlife (Brown 1988; Pemberton 1988; 1989; Peterson 1990; Service 1990; Robertson and Duncan 1991).

1.7 AIMS OF THE PROGRAM OF RESEARCH

The primary aim of the research described in this thesis is to investigate two biological traits in the Tasmanian thylacine that are associated with high extinction risk among large carnivores: prey body size and resource competition. The specific aims of each research component were to:

- (i) Determine if the thylacine's diet was energetically constraining by assessing their maximum prey size (Chapter 2 and 3) and prey composition post-settlement (Chapter 4).
- (ii) Evaluate the likelihood of competition between the thylacine and sympatric marsupial carnivores by quantifying the degree of overlap in their prey size (Chapter 2 and 3) and trophic niche (Chapter 4).

(iii) Assess long-term trends in the trophic structure and habitat of the thylacine and extant marsupial carnivores in Tasmania post-European settlement (Chapter 4). A critical evaluation of the ecology of these apex predators will provide the first step to comprehending the full extent of their vulnerability to anthropogenic impacts. This knowledge will also provide the foundation for more meaningful conservation strategies to be adopted for remaining marsupial carnivores.

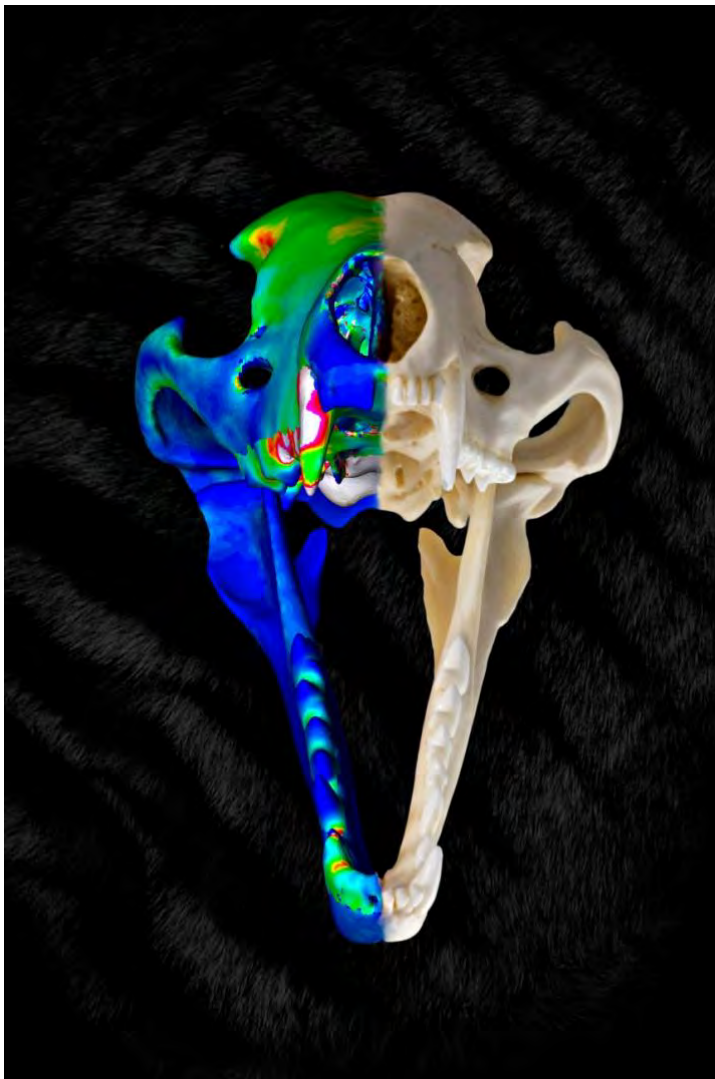
1.8 THESIS OVERVIEW

Interdisciplinary techniques involving skull biomechanics and stable isotope analysis will be used to inform us of ecological and behavioural traits of the thylacine within the context of extant and extinct carnivorous marsupial species. Both approaches are frequently used in palaeoecological and ecological studies (West *et al.* 2006), yet to my knowledge have never been used in combination to reconstruct the ecology of a species. Niche partitioning among marsupial carnivore species and maximum prey size were investigated by comparing the biomechanics and associated feeding limitations of the thylacine skull relative to that of extant marsupial carnivores (Chapter 2 and 3) and one fossil thylacinid (Chapter 3). Niche overlap and potential for intersexual and interspecific resource competition was further explored by comparing the trophic and habitat niche space of marsupial carnivore specimens collected in Tasmania prior to the extinction of the thylacine using a stable isotope approach (Chapter 4). The contribution of different prey species to the diet of Tasmanian thylacines following European settlement was determined using a dual isotope mixing model of preserved predator and potential prey tissues (Chapter 4). The response of the thylacine and extant marsupial carnivores to long-term changes in the Tasmanian ecosystem following European settlement was determined using stable isotope analysis of tissues from historic and modern specimens (Chapter 4). The concluding chapter provides an overview of the findings of this thesis and discusses their potential implications for the management of native species and communities in Tasmania.

Chapter 2

SKULL MECHANICS AND IMPLICATIONS FOR FEEDING BEHAVIOUR IN A LARGE MARSUPIAL CARNIVORE GUILD: THE THYLACINE, TASMANIAN DEVIL AND SPOTTED-TAILED QUOLL

Published in Journal of Zoology (2011), **285**, 292-300



2.1 ABSTRACT

Extinction risk varies across species and is influenced by key ecological parameters, such as diet specialisation. For predictive conservation science to be effective, we need to understand extinction risk factors that may have implicated recent species extinctions. Diet and feeding behaviour of the large extinct marsupial carnivore, the thylacine (*Thylacinus cynocephalus*), have long been debated. Improved understanding of the skull's biomechanical performance and its limitations in a comparative context may yield important insights. Here, we use three dimensional (3D) finite element analysis (FEA) to assess aspects of biomechanical performance in the skull of the thylacine relative to those of two extant marsupial carnivores with known diets that occurred sympatrically with the thylacine: the Tasmanian devil (*Sarcophilus harrisii*) and spotted-tailed quoll (*Dasyurus maculatus*). Together, these three species comprised the large mammalian carnivore guild in Tasmania at the time of European settlement. The bone crunching Tasmanian devil produced high bite forces for its size as expected, but the stresses induced were surprisingly high. A higher proportion of cancellous bone in the skull of this osteophage may act to absorb shock but decrease rigidity and hence raise stress. A relatively high bite force and rigid skull characterised the spotted-tailed quoll, which may allow them to target prey of variable sizes. Compared with the Tasmanian devil and spotted-tailed quoll, we found that the skull of the thylacine was least well adapted to withstand forces driven solely by its jaw-closing musculature, as well as to simulations of struggling prey. Our findings suggest that the thylacine likely consumed smaller prey relative to its size, which may have had implications for their survival.

2.2 INTRODUCTION

The idea that some species are intrinsically at a greater risk of extinction than others is a well-established concept, and recent efforts have focused on identifying species susceptible to extinction (McKinney 1997). Species traits, such as body size and degree of diet and habitat specialisation, are key predictors used to prioritise species in conservation and management plans (Purvis *et al.* 2000; Cardillo and Bromham 2001; Kamilar and Paciulli 2008). Top predators, which have less stable food supplies than species from lower trophic levels, are thought to be particularly vulnerable (Davies *et al.* 2000).

The thylacine (*Thylacinus cynocephalus*) was a large apex predator and the only species of marsupial to become extinct in Tasmania following European settlement. The factors involved in its extinction in Tasmania have been debated (see review Paddle 2000), and yet the underlying cause of their decline remains incomplete. Dental and biogeochemical evidence suggests that the thylacine was a hypercarnivore restricted to eating vertebrate flesh (Nedin 1991; Wroe *et al.* 1998; Wroe *et al.* 2008). The total energetic expenditure and time spent hunting by carnivorous mammals increases with body mass (Carbone *et al.* 1999). Given its large body mass and carnivorous diet, any limitations in the capacity of thylacines to kill large-bodied species may have made them particularly vulnerable unless small prey was available in high abundance.

There is almost no direct evidence regarding the diet of the thylacine (Paddle 2000). The range of potential prey taxa in 19th century Tasmania included the forester kangaroo (*Macropus giganteus*) and now extinct Tasmanian emu (*Dromaius novaehollandiae diemenensis*) and that exceeded the marsupial carnivore's body mass. Whether or not thylacines were well adapted to kill prey approaching or exceeding its own body mass remains a contentious issue. Previous morphological studies on this species suggest that they targeted prey considerably smaller (1–5 kg) than themselves (Jones and Stoddart 1998). Most data are restricted to Tasmania, whereas in Pleistocene and early Holocene

times, the species ranged across Australia and New Guinea (Plane 1976). At that time, still larger, now extinct megafauna were theoretically available and diet may have varied in different parts of the species range.

We can infer the diet of extinct species such as the thylacine, including likely prey size, through comparisons with related species with known diets. The thylacine is a member of the now wholly extinct family Thylacinidae. Its closest living relatives are the dasyurids (Dasyuridae), a now diverse taxon comprising over 60 species. Together with the monotypic numbat (Myrmecobiidae), these three families constitute the order Dasyuromorphia (Wroe and Musser 2001). First appearing in the fossil record in the late Oligocene to early Miocene, dasyurid diversity appears to have been initially low, but increased through the late Miocene to ultimately dominate most insectivore–carnivore niches. In contrast, the Thylacinidae were once more diversely represented by 11 species spanning a considerable range of body sizes (Wroe 1996; Muirhead and Wroe 1998; Wroe and Musser 2001).

The two largest extant dasyurids are the Tasmanian devil (*Sarcophilus harrisii*; average body mass 6–13 kg) and the spotted-tailed quoll (*Dasyurus maculatus*; average body mass 1–4 kg) (Jones and Barmuta 2000). The dietary preferences of these two species have been extensively studied. The Tasmanian devil can generate the highest maximum bite force, adjusted for body mass, of any extant mammalian species studied to date (Wroe *et al.* 2005). This feature may assist them in catching relatively large mammals, up to 30 kg (Jones and Barmuta 1998). Approximately 60% of the prey biomass consumed by adult male spotted-tailed quolls consists of medium- to large-sized mammals including bandicoots, pademelons and possums that can weigh up to three times its own body mass, although invertebrates form a larger proportion of the diet of female spotted-tailed quolls (Belcher 1995; Jones 1997). Known size relationships between predator and prey in the Tasmanian devil and spotted-tailed quoll make them ideal candidates to compare with the thylacine to determine prey size range.

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

The maximum prey size that can be taken by a predator is strongly influenced by biomechanical limitations. Skull shape and bite force adjusted for body mass correlate with prey size and feeding ecology in many terrestrial carnivores (Meers 2002; Wroe *et al.* 2005; McHenry *et al.* 2006; Wroe and Milne 2007). Animals that need stronger jaw-closing muscles to catch large prey or process hard materials may develop larger surface areas for muscle attachment, such as the sagittal crest along the midline of the top of the skull, and zygomatic arches extending along the side of the skull beneath the orbit (Tanner *et al.* 2008). The skull must also be rigid enough to withstand forces applied by jaw-closing muscles, along with feeding and other forces. Different cranial shapes among extant marsupials have been correlated with diet (Wroe and Milne 2007), and yet little is known about how the biomechanical performance of these structures may reflect dietary functions.

Unlike most engineering structures, the geometry of biological structures such as the skull is extremely complex. Problems involving complicated geometries, loadings, and multiple material properties are difficult to solve using analytical methods. Instead, numerical methods such as Finite Element Analysis (FEA) are required (Geng *et al.* 2001). FEA is able to obtain a solution to complex geometries by dividing the structure into a collection of much smaller and simpler domains called elements. Solutions are formulated for each individual element and are then combined to obtain a solution for the entire structure. As a non-invasive technique, FEA is particularly attractive for paleontologists and has been increasingly used over the past two decades to investigate the skull mechanics of extinct species (Walmsley *et al.* 2013).

Testing hypotheses regarding the relationship between the form and function of skulls from extinct species requires an understanding of this relationship in living animals (Ross 2005). A comparative biomechanics approach involving living analogues are thus used to predict the feeding behaviour of extinct species (McHenry *et al.* 2007; Wroe *et al.* 2007; Tseng 2008; Bell *et al.* 2009; Porro 2009; Wroe *et al.* 2010; Young *et al.* 2012). A recent study by Wroe *et al.* (2007) examined convergence with respect to

biomechanical performance in the skulls of the thylacine and the placental grey wolf subspecies, the dingo (*Canis lupus*). The results suggested that relative to *the* dingo, thylacines may have been restricted to smaller prey, but closely related species were not included in this analysis.

In the present study, we apply FEA to examine differences in biomechanical performance between the skulls of the thylacine and two extant marsupial carnivore relatives: the Tasmanian devil and spotted-tailed quoll. Our objectives are twofold. Firstly, fundamental structural differences between marsupial and placental skulls (Wroe and Milne 2007) make comparisons between these taxa difficult because differences may be phylogenetically constrained (Wroe *et al.* 2007; Goswami *et al.* 2011). Additional insights into the feeding ecology of the thylacine may be derived from comparisons with closely related, extant marsupial carnivores. Secondly, we will examine whether variation in the biomechanical performance of these sympatric marsupial carnivores reflects differences in the feeding ecology of each species.

2.3 MATERIALS AND METHODS

2.3.1 Specimens

Spotted-tailed quoll (UNSW Z20; University of New South Wales), Tasmanian devil (AM10756; Australian Museum) and thylacine (AM1821; Australian Museum). The skull length of each specimen was 101, 135 and 232 mm, respectively. Marsupial carnivores display substantial sexual dimorphism (Jones 1997). Data on sex were unavailable; however, because each is within the upper size range for their species, it is likely that all were adult males.

2.3.2 Model assembly

The assembly of finite element models (FEMs) largely follows the procedures applied by Wroe *et al.* (2007; 2010). Computed tomography (CT) scanning was conducted at

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

the Mater Hospital, Newcastle, NSW, using a Toshiba Aquilon 16 scanner (Toshiba Medical Systems Corporation, Otawara, Tachigi, Japan). Slices were 1mm thick, with an inter-slice distance of 0.8 mm and a field of view of 240 mm. Surface meshes were generated from the CT data using Mimics 13.2 and converted to FEMs in STRAND7 (ver. 2.4).

Heterogeneous FEMs were composed of 3D four-noded tetrahedral ‘brick’ elements. Brick elements were assigned one of eight material properties based on CT density values (Rho *et al.* 1995; Schneider *et al.* 1996) ranging from low-density tissue [Young’s modulus (Y)=1.5GPa; Poisson’s ratio=0.4; density=251 kgm⁻³] to enamel (Y)=38.6GPa; Poisson’s ratio=0.4, density=28.61 kgm⁻³) and see McHenry *et al.* (2007). Full data for the eight material properties are available in Table 2.1. Jaw elevators were modelled as seven muscle subdivisions: *temporalis superficialis*, *temporalis profundus*, *masseter superficialis*, *masseter profundus*, *zygomaticomandibularis*, *pterygoideus internus* and *pterygoideus externus* (Wroe *et al.* 2007). Maximum contractile forces were estimated using the dry skull method (Thomason 1991) on the basis of estimated maximal cross-sectional areas with an estimated force of 300 kPa for vertebrate striated muscle (Weijs and Hillen 1985).

Table 2.1 Values used to assign eight material properties to brick elements in heterogeneous models.

Material property type	Poisson's ratio	Young's modulus (MPa)	Density (T/mm ³)
1	0.40	1527.00	2.51E-10
2	0.40	1868.60	2.92E-10
3	0.40	2223.40	3.33E-10
4	0.40	10786.80	1.09E-10
5	0.40	21734.20	1.86E-09
6	0.40	27082.20	2.19E-09
7	0.40	32704.30	2.53E-09
8	0.40	38575.40	2.86E-09

The protocols used for the 3D modelling of muscle architecture following Wroe *et al.* (2007) were consistent in all FEMs and therefore any variations in von Mises (VM) stresses observed are likely a result of morphological differences. Muscle forces were distributed in each model based on the percentage contribution of each muscle division to the total jaw muscle mass in the Virginia opossum (*Didelphis virginiana*) (Turnbull 1970; Wroe *et al.* 2007). The muscles themselves were simulated using pre-tensioned trusses that connected origin and insertion areas. Figure 2.1 displays muscle origin and insertion areas and lines of action of muscle forces. The number of trusses allocated to each muscle division for each species was based on the muscle origin and insertion areas of the thylacine (Wroe *et al.* 2007) for ease of comparison. Truss elements were evenly distributed in each muscle subdivision and positioned in the approximate vectors of the contracting musculature. Pretention values for each truss element were determined by dividing the total muscle force for each muscle subdivision by the number of trusses allocated to that group (Table 2.2). Truss diameters were calculated from cross-sectional areas of temporalis and masseteric muscles.

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

Models were restrained following procedures applied by Wroe *et al.* (2007). A rigid link framework at the occipital condyle was used to restrain the cranium in all simulations. Static loading cases were set to an optimum gape angle of 35°, which is consistent with the optimum gape angle found in the dingo (Bourke *et al.* 2008).

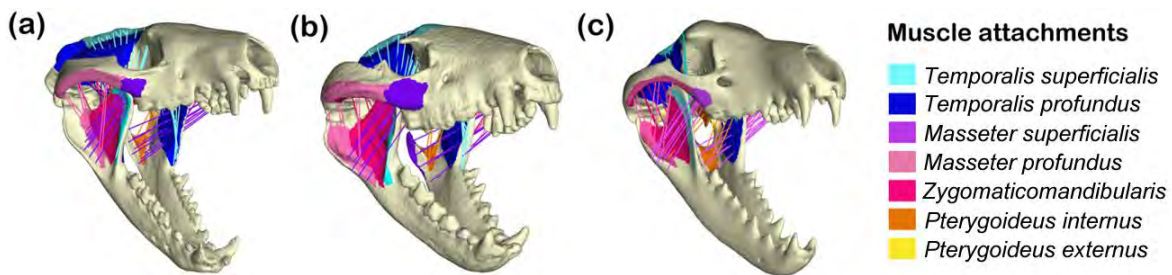


Figure 2.1 Muscle origin and insertion areas and lines of action of muscle forces for skulls of (a) spotted-tailed quoll (b) Tasmanian devil, and (c) thylacine constructed using finite element software (Strand 7, Vers 2.4).

Table 2.2 Force used for each jaw muscle division in unscaled models of the spotted-tailed quoll, Tasmanian devil and thylacine. These were calculated using muscle mass proportions from a dissected Virginia opossum (Turnbull 1970).

	Muscle force distribution (in Newtons) for different muscle groups		
	Spotted-tailed quoll	Tasmanian devil	Thylacine
<i>Temporalis superficialis</i>	76.00	176.00	274.00
<i>Temporalis profundus</i>	132.00	307.00	478.00
<i>Masseter superficialis</i>	55.00	128.00	199.00
<i>Masseter profundus</i>	36.00	83.00	129.00
<i>Zygomaticomandibularis</i>	34.00	79.00	123.00
<i>Pterygoideus internus</i>	27.00	62.00	97.00
<i>Pterygoideus externus</i>	6.00	14.00	21.00

2.3.3 Load cases

We applied six loading cases to each model: two intrinsic loading cases to simulate bites driven solely by jaw musculature and four extrinsic loading cases to test skull performance under the influence of unrestrained prey. The intrinsic cases comprised a bilateral bite at the canines and a unilateral bite at the fourth molar, or ‘carnassial’. Constraints were applied at the tip of the upper and lower canines on both sides of the skull for the bilateral bite and at the upper and lower fourth molar on the right side of the skull for the unilateral bite.

The maximum bite force of each specimen was measured for both intrinsic cases using unscaled models with specimen-specific estimated muscle forces (Table 2.2). We also solved two sets of load cases with models scaled to the same total surface area of the thylacine to remove the effects of size where (1) the same muscle forces were applied to each specimen to quantify differences in bite forces after adjusting for size differences; (2) the same bite reaction force as the thylacine (747 N at canines and 1659 N at carnassial) was allocated to each model to determine how well adapted each species was to withstand the stresses generated in the production of a given bite reaction force. Scaled models were also used to measure the muscle recruitment required to produce the same bite force as the thylacine.

Extrinsic loading cases were applied only to scaled models, and consisted of (1) a lateral ‘shake’ (50 N); (2) axial twist (5000 Nmm); (3) pull back (50 N); (4) dorsoventral force (50 N). These extrinsic loading cases simulated different behaviours used by carnivores during prey capture and killing. Axial twisting and lateral shaking in particular are used by dasyurids to disorient unrestrained prey and assist in canine penetration into the skull of restrained prey (Pellis and Officer 1987). All extrinsic forces were applied using an H-frame that linked the upper and lower jaws, with forces applied at the centre of the frame (and see Wroe *et al.* 2007). The extrinsic forces applied were based on the estimated body mass of the thylacine taken from Wroe *et al.* (2007) and are low relative

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

to the size of the predators. However, as this is a comparative analysis using scaled models, the actual value used is immaterial, provided that it is held constant across species (Dumont *et al.* 2009).

In routine mastication in mammals, not all muscles are recruited at the same time, or maximally on both working and balancing sides (Thexton and Hiiemae 1977). The activity patterns in masticatory muscles are also highly variable between species (Crompton *et al.* 2008). However, in this study, as in most other comparable studies, we predicted the maximum bite force in each species where all jaw adductor muscles have been activated as we wanted to assess the maximal bite forces. The overarching objective here is to determine mechanical performance limits, as these are most likely to put a ceiling on potential killing behaviours and prey size. The actual bite force used by these species in reality will likely not be as high as our values because animals will vary their bite force depending on the task and context.

The general approaches applied here have been validated against experimental data from other taxa (Tsafnat and Wroe 2010; Wroe *et al.* 2010), but not yet using FEMs of marsupial carnivores. In the absence of validated data for material properties and forces generated in the despatch of large prey, actual mechanical limits on maximal prey size cannot be directly predicted. We apply a comparative approach wherein the results must be interpreted in relative context and not as predictions of absolute stress magnitudes (Wroe *et al.* 2010).

Bone fails under a ductile model of fracture and von Mises stress is a good predictor of failure in ductile materials (Nalla *et al.* 2003; Tsafnat and Wroe 2010). Therefore, VM stress was used here as a metric to compare the structural strength of FEMs under different loading conditions. Brick elements were selected at set points along the mid-sagittal plane, zygomatic arch and mandible to assess changes in stress magnitudes and distributions under different loadings. At each point, values were calculated by averaging VM stress of the bricks surrounding each node, with between five and nine

bricks joined at any given point. Nodes were equally distributed and VM stress was measured using the same node points in each simulation. For mid-sagittal sampling, stresses were sampled from the anterior edge of the nasal bone to the posteriormost point of the sagittal crest. At the zygomatic arch, nodes were selected midway between the top and the bottom of the arch from the most anterior extreme of the jugal to the posterior extreme of the zygomatic arch. Nodes were also selected along the right side of the jaw from the mid-point between the first incisors and ventral surface of the jaw to the temporomandibular joint.

Our FEMs were not of sufficient resolution to accurately incorporate material property distributions within teeth. Enamel thickness and distribution may play a major role in the processing of hard materials (Koenigswald *et al.* 1987). Differences in enamel microstructure between marsupial carnivores has not been clearly correlated with diet, however it may reflect the function of different tooth types and the loading conditions of specific sites within the teeth (Stefen 1999). The Tasmanian devil and thylacine have complex decussation in the middle and lower section of their teeth which may help prevent cracking due to vertical tensile stresses.

The proportion of cortical to cancellous bone in the mandible and cranium was estimated based on volume ratios (Table 2.3). Brick elements with material properties 1–4 were allocated as cancellous bone, while 5 (i.e., density of 1855.9 kg m^{-3}) and higher were considered cortical bone.

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

Table 2.3 Volume (mm³) and percentage volume (%) of each material property in the skull of the spotted-tailed quoll, Tasmanian devil and thylacine.

Material property type	Combined volume (%) of cranium and mandible		
	Spotted-tailed quoll	Tasmanian devil	Thylacine
1	8.80	10.48	17.33
2	6.65	9.53	13.01
3	7.72	11.21	11.32
4	18.16	33.51	21.14
5	8.49	11.75	10.04
6	9.44	9.92	10.41
7	11.30	6.39	9.37
8	29.43	7.23	7.38

2.4 RESULTS

Bite forces were higher at the carnassial than at the canines for each model and were positively correlated with body size (Figure 2.2a). Despite having an estimated body size only 44% of the thylacine, the Tasmanian devil has a modeled canine and ‘carnassial’ bit force of 89% and 87% of the thylacine in unscaled models, respectively. However, among models scaled to a uniform surface area and muscle recruitment force, the Tasmanian devil had the lowest bite force for both intrinsic cases (Figure 2.2b) and required the highest muscle recruitment to produce the same bite force (Figure 2.3). Overall, the magnitudes and distributions of VM stress in Tasmanian devil loading simulations are broadly similar to those generated in the thylacine and spotted-tailed quoll. The spotted-tailed quoll produced the highest bite forces in scaled models for both loading cases (Figure 2.2b), which suggests that the geometry of its lever system is the most efficient.

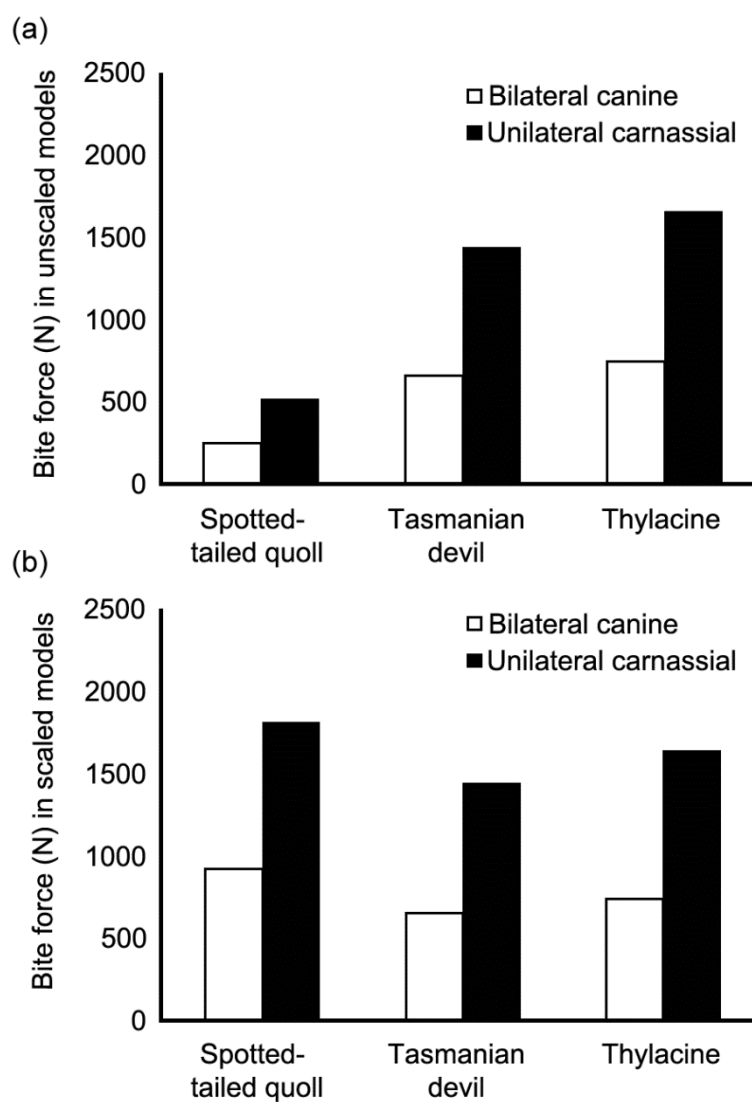


Figure 2.2 Bite force reaction in the spotted-tailed quoll, Tasmanian devil and thylacine during a bilateral bite and unilateral carnassial bite for (a) unscaled models and (b) models scaled to the same surface area and muscle force as the thylacine.

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

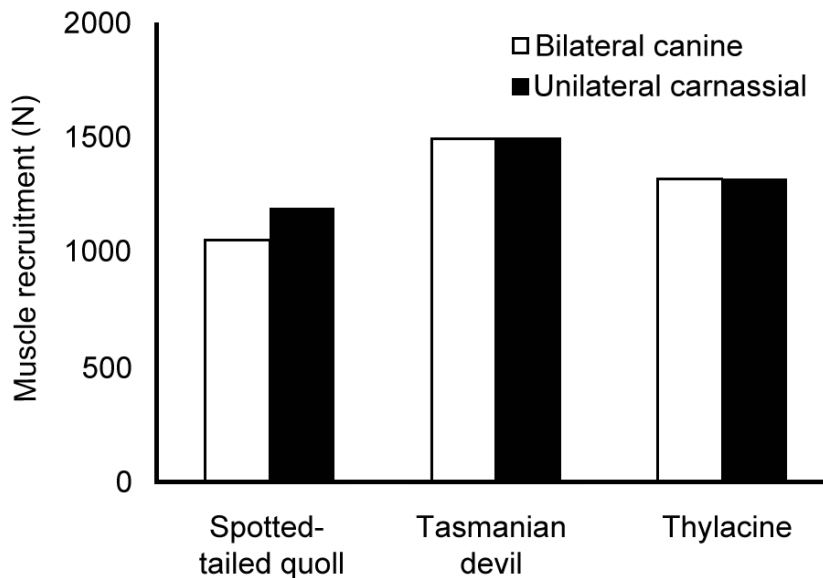


Figure 2.3 Muscle recruitment of the spotted-tailed quoll and Tasmanian devil to produce the same bite force as the thylacine in models scaled to a uniform surface area.

The distribution of stress in all models was similar on both sides of the skull for bilateral loads (Figure 2.4a–c) and was largely concentrated on the working side for unilateral loads (Figure 2.4d–f). Peak VM stresses in the thylacine during a canine bite were the highest at the nasal and temporal line (Figure 2.4c, g), which may be attributed to its relatively long rostrum. Unilateral carnassial loading on the thylacine resulted in localised stress at the temporal line, with higher peak stresses than other species (Figure 2.4h). In the cranium, VM stresses were greatest at the frontal line of the Tasmanian devil and at the temporal line for the spotted-tailed quoll in both intrinsic loads (Figure 2.4g, h). The marsupial carnivores studied here all showed a similar distribution of stress along the zygomatic arch that peaked posteriorly. The highest stresses were located along the mandible in scaled models during intrinsic loads (Figure 2.5), a finding consistent with studies in other taxa (Wroe 2008). The mandible may be a more reliable indicator of feeding behaviour than the cranium because selective pressure on its morphology is less likely to be influenced by competing demands such as olfaction, hearing and vision (Figueirido *et al.* 2008; Wroe *et al.* 2010).

The ability to resist extrinsic forces may be a better indicator of a predator's ability to subdue relatively large prey (Preuschoft and Witzel 2005). Differences between species undergoing extrinsic loads were greatest in the cranium (Figure 2.6). Lateral shaking caused the highest stresses along the zygomatic arches and mid-sagittal planes of each species. Stresses were higher for the thylacine than the other two species in all extrinsic loading cases. The highest VM stresses accumulated at the parietal of the thylacine during a pullback (Figure 2.6f, h) and dorsoventral (Figure 2.6l, p) loading and at the anterior point of the rostrum and parietal during a lateral shake (Figure 2.6i, o).

The proportion of cortical and cancellous bone in the skull varied considerably between species (Figure 2.7). The spotted-tailed quoll had the highest proportions of cortical bone in the cranium and mandible and the Tasmanian devil had the highest proportion of cancellous bone.

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

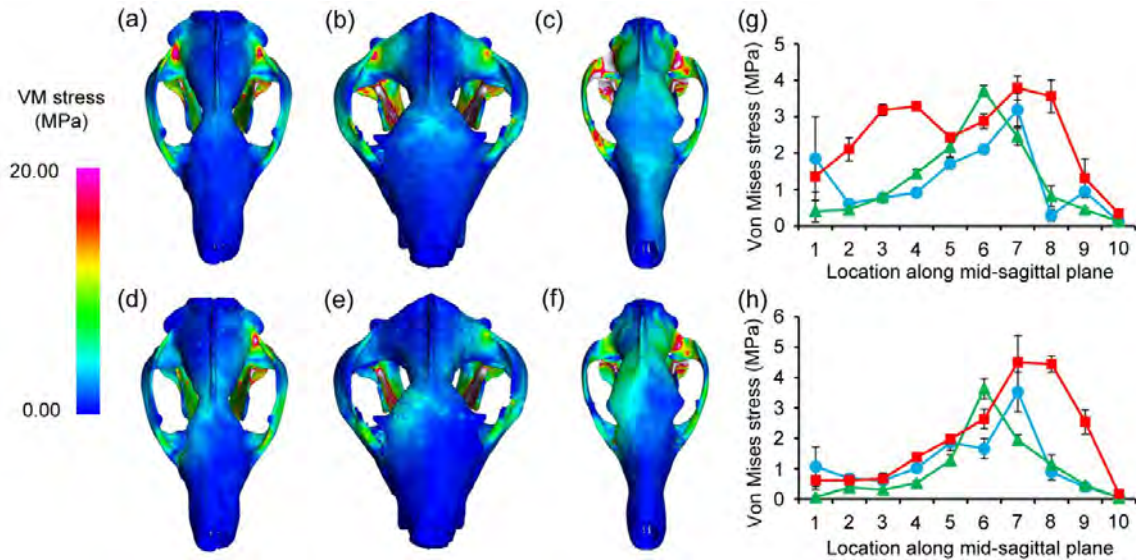


Figure 2.4 Dorsal view of the stress distribution in scaled heterogeneous models with a uniform bite force during a bilateral canine bite (a–c) and a unilateral carnassial bite (e, f). Dorsal views of models are displayed for the spotted-tailed quoll (a, d), Tasmanian devil (b, e) and thylacine (c, f). von Mises (VM) stress was measured from the anterior to posterior along the mid-sagittal plane for a bilateral canine bite (g) and a carnassial bite (h). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle).

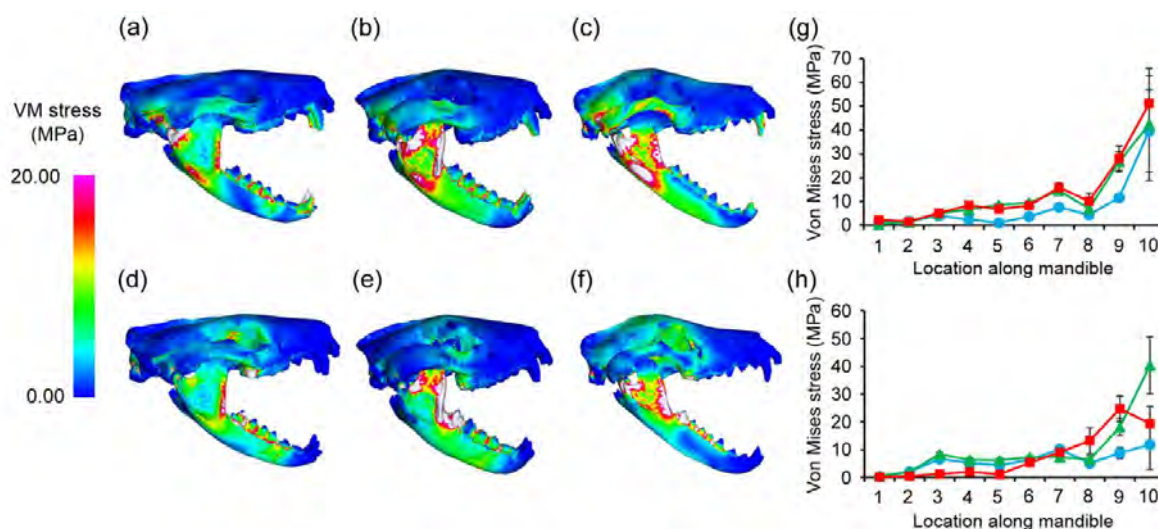


Figure 2.5 Lateral view of the stress distribution in heterogeneous models with a uniform bite force in scaled models during a bilateral canine bite (a–c) and a unilateral carnassial bite (d–f). von Mises (VM) stress was measured from the anterior to posterior along the jaw for a bilateral canine bite (g) and a carnassial bite (h). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle).

2. SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

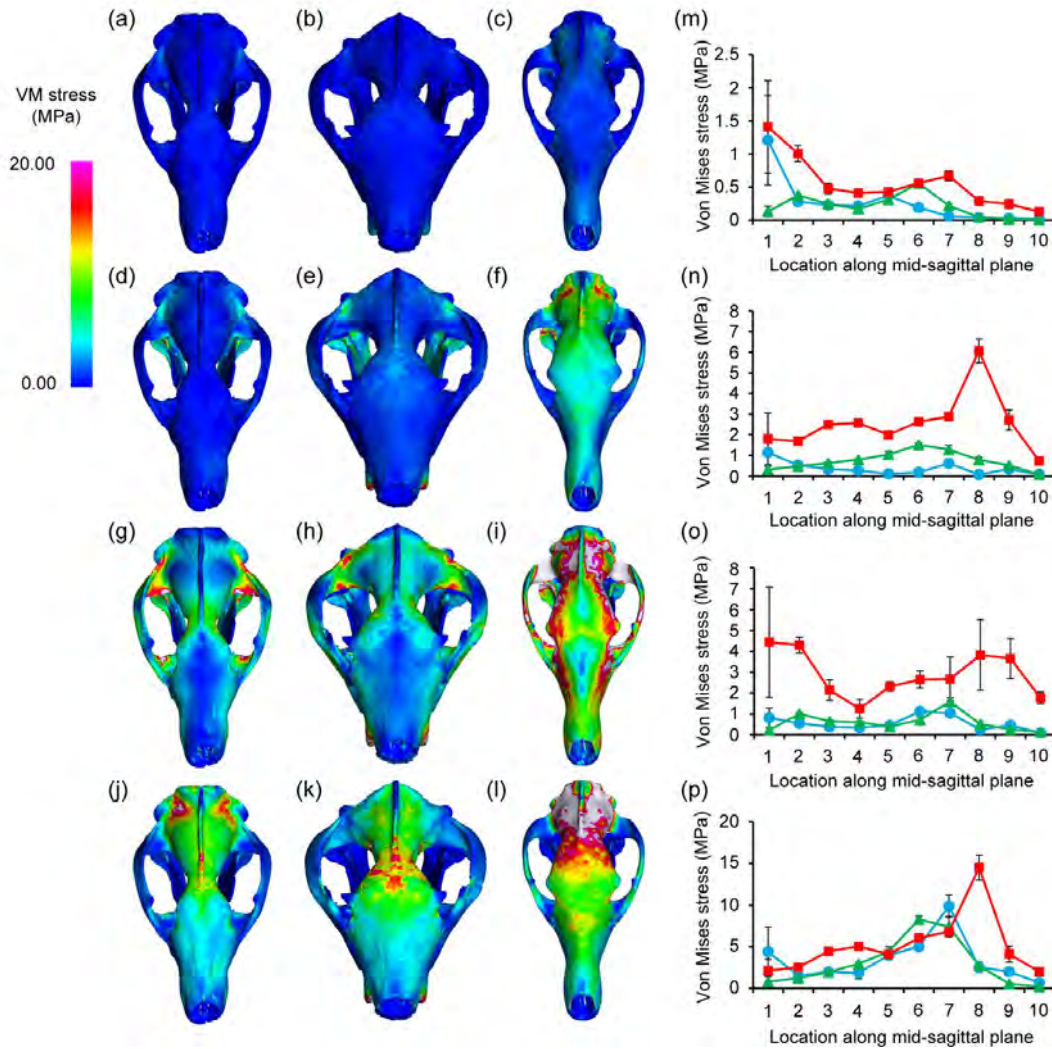


Figure 2.6 Stress distribution for extrinsic loads. Comparison of the distribution of von Mises (VM) stresses in MPa in models scaled to the same surface area during four extrinsic loads: an axial twist (a–c), pullback (d–f), lateral shake (g–i) and dorsoventral (j–l). Species from left to right are the spotted-tailed quoll (a, d, g, j), Tasmanian devil (b, e, h, k) and thylacine (c, f, i, l). VM stress was measured from the anterior to the posterior along the mid-sagittal plane for each species during an axial twist (m), pullback (n), lateral shake (o) and dorsoventral (p). Symbols for each species are: thylacine (square), Tasmanian devil (triangle) and spotted-tailed quoll (circle). White areas indicate VM stress exceeds the scale maximum (2.5 MPa) in those areas.

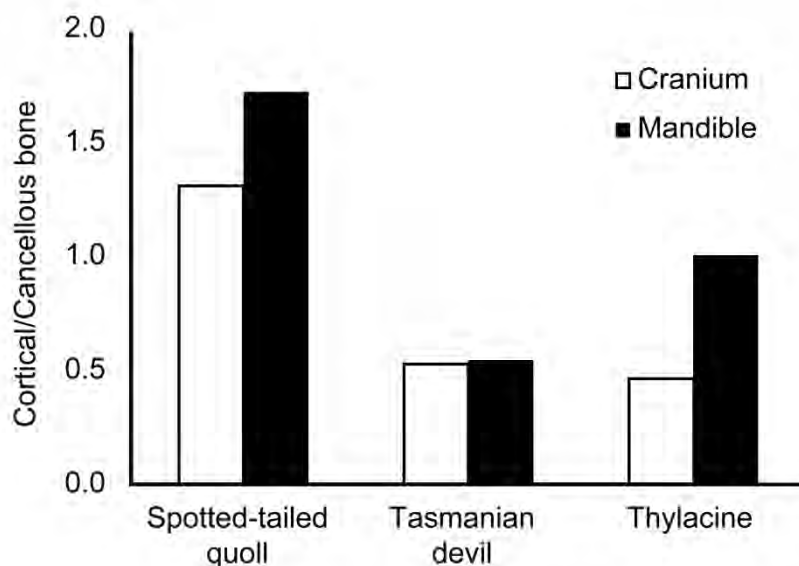


Figure 2.7 Volume ratio of cortical to cancellous bone in the mandible and cranium of the spotted-tailed quoll, Tasmanian devil and thylacine.

2.5 DISCUSSION

Comparatively high levels of stress in the thylacine may be related to its relatively long rostrum (Werdelin 1986). A longer snout may enable higher velocities at the canine, a characteristic of predators specialising in relatively small, more agile prey and use a pounce-pursuit or ambush hunting style. *The spotted-tailed quoll and Tasmanian devil* can catch prey considerably larger than themselves (Jones 1995; Jones and Barmuta 1998). Anecdotal accounts suggest that thylacines may have consumed only soft tissue (Paddle 2000). Certainly, its dentition shows none of the features present in specialised osteophages, such as the Tasmanian devil, which can consume whole carcasses, including teeth, bone and fur (Owen and Pemberton 2005), or species consuming significant proportions of invertebrate food. Given the evidence for a diet restricted with respect to prey size, type and the proportion of the prey item that it could effectively consume, we suggest that the thylacine was particularly sensitive to environmental disturbance. Our results also suggest a potential overlap between the thylacine and the

2.SKULL MECHANICS AND FEEDING BEHAVIOUR OF THE THYLACINE

two dasyurids regarding prey size. Niche overlap has been found between sub-adult Tasmanian devils and male spotted-tailed quolls (Jones and Barmuta 1998), although male spotted-tailed quolls consume a significantly higher proportion of arboreal prey than do sub-adult Tasmanian devils. Current hypotheses suggest that the thylacine preyed on juvenile Tasmanian devils, and Tasmanian devils ate thylacine young and scavenged from kills made by thylacines (Jones 1998). Until late Pleistocene times, this guild also included the still larger marsupial lion (*Thylacoleo carnifex*) (Wroe *et al.* 1999) that likely preyed upon these smaller predators. A shared preference towards mammalian prey may have exposed these species to increased periods of resource scarcity (Wiens 1993).

Our finding that the thylacine was not well adapted to take relatively large prey compared with other marsupials brings into question the putative role of its large gape if not as a means to assist in the capture and killing of large prey. It is possible that thylacines used their wide gape as a threat display, as does the Tasmanian devil (Pemberton and Renouf 1993). Several instances of agonistic gaping behaviour were described in both wild and captive thylacines in response to humans (Paddle 2000). Alternatively, the wide gape angle of thylacines may have been a retained plesiomorphy. The contention that thylacines would have been able to open its jaws to angles of 120° (Mittelbach and Crewdson 2005) is unrealistic and would almost certainly cause dislocation due to the physical constraints of the pivot joint. It is more probable that they were capable of gape angles similar to the Tasmanian devil, which can open their jaws 75–80° (Pemberton and Renouf 1993), which is still a very wide gape for a mammal. Possession of a relatively high bite force in the Tasmanian devil (Wroe *et al.* 2005) was expected and supported by our results. We anticipated that the short, broad skull shape of the Tasmanian devil was primarily responsible for the production of extremely high bite forces in this species by reducing the length of the jaw out-lever, thereby increasing the leverage of the jaw musculature. However, *contra* our expectations, the skull and muscle geometry of the Tasmanian devil did not transmit jaw muscle forces into bite forces more efficiently, with this species producing the

lowest bite forces in models scaled to the same surface area and muscle force. Rather, these high bite forces are the product of a relatively greater muscle force.

As a bone-crunching specialist, we anticipated that the Tasmanian devil would generate less stress in the skull than flesh eating carnivores. In contrast to our expectations, the magnitude and distribution of stress in the mandible of scaled models with uniform bite reaction forces were similar for all species studied here. Differences in the mechanical performance of these dasyuromorphians may be related to their different foraging strategies. Tasmanian devils primarily scavenge whereas spotted-tailed quolls are predominantly active hunters. They are both able to crush the skull of live prey (Ewer 1969; Buchmann and Guiler 1977), although the enlarged cheek-tooth cusps of Tasmanian devils are better adapted for breaking open and consuming bone. Spotted-tailed quolls feed on both fast- and slow-moving prey of various sizes, utilise patchy resources and will take advantage of short term fluctuations in prey abundance (Glen and Dickman 2006; Dawson *et al.* 2007). Distinct differences between the relative proportions and distributions of cancellous and cortical bone in the skull, as revealed in our study, may influence the distribution and magnitude of stress (Thomason 1995). Surprisingly, the Tasmanian devil showed the highest proportion of less stiff cancellous bone. However, it may be that a more compliant skull may enable Tasmanian devils to absorb the shock induced by biting down on relatively hard bone.

Allometric factors may further complicate interpretation here. Recent work on primates suggests that cranial rigidity decreases with increasing size (Strait *et al.* 2010). Our findings are partially consistent with this suggestion insofar as we found that the skull of the smallest species, the spotted-tailed quoll, had the highest proportion of stiff cortical bone.

The Tasmanian devil is currently critically endangered by both facial cancer and the introduction of the red fox (*Vulpes vulpes*) (Hawkins *et al.* 2006). It has been argued that interference competition with Tasmanian devils has been an important factor in thwarting the success of previous attempts to establish red foxes in Tasmania. This may

have been through either aggressive exclusion or predation on denning juveniles. Spotted-tailed quolls also experience competition from feral cats (*Felis catus*) for food and den sites (Jones and Barmuta 1998). Future comparisons of mechanical performance in marsupial carnivores with a wider range of placental carnivores, including potential exotic competitors, may yield useful insights into the likely outcomes of such competition.

2.6 ACKNOWLEDGEMENTS

We thank the University of New South Wales and S. Ingleby at the Australian Museum for providing specimens used in this study. This work was funded by the University of New South Wales Internal Strategic Initiatives Grant to S.Wroe and the Australian Research Council (DP0666374 and DP0987985).

Chapter 3

VIRTUAL RECONSTRUCTION AND PREY SIZE PREFERENCE OF THE MID CENOZOIC THYLACINID, *NIMBACINUS DICKSONI* (THYLACINIDAE, MARSUPIALIA)

In review in PLOS ONE



3.1 ABSTRACT

Thylacinidae is an extinct family of Australian and New Guinean marsupial carnivores, comprising 12 species, the oldest of which are late Oligocene in age. Except for the recently extinct thylacine (*Thylacinus cynocephalus*), most are known from fragmentary craniodental material only. However, a particularly well-preserved skull of the fossil species *Nimbacinus dicksoni* Muirhead and Archer, 1990, has been recovered from middle Miocene deposits in the Riversleigh World Heritage Area, north-western Queensland. Here, to determine whether *N. dicksoni* was more likely a relatively small or large prey specialist, we have digitally reconstructed its skull and applied three-dimensional (3D) Finite Element Analysis (FEA) to compare its mechanical performance with that of three extant marsupial carnivores and the thylacine. Under loadings adjusted for differences in size that simulated forces generated by both jaw closing musculature and struggling prey, we found that stress distributions and magnitudes in the skull of *N. dicksoni* were more similar to those of the living spotted-tailed quoll (*Dasyurus maculatus*) than to its recently extinct relative. Considering FEA results and dental morphology, we predict that *N. dicksoni* likely occupied a broadly similar ecological niche to that of the spotted-tailed quoll, and were likely capable of hunting vertebrate prey that may have exceeded its own body mass.

3.2 INTRODUCTION

Thylacinids first appear in the Australian fossil record during the late Oligocene and include the largest representatives of the Dasyuromorphia, i.e., families Thylacinidae, Dasyuridae and Myrmecobiidae (Lowenstein *et al.* 1981; Sarich *et al.* 1982; Szalay 1982; Thomas *et al.* 1989; Krajewski *et al.* 1992; Krajewski *et al.* 1997; Muirhead and Wroe 1998; Wroe and Archer 2006). While these marsupial predators are generally considered morphologically conservative, variation in their dentition, skull shape and body size (~1-60 kg), suggests considerable trophic diversity within the family (Wroe 2001; Wroe and Milne 2007). In addition to the recently extinct thylacine or Tasmanian 'tiger' (*Thylacinus cynocephalus*), eleven fossil species of thylacinid have been described (Muirhead and Archer 1990; Wroe 1996; Muirhead 1997; Murray 1997; Muirhead and Wroe 1998; Murray and Megirian 2000; Wroe and Musser 2001; Archer *et al.* 2006). Up to five species may have co-existed in the Riversleigh World Heritage Area, north-western Queensland between the late Oligocene to middle Miocene (Murray and Megirian 2000; Archer *et al.* 2006).

The Riversleigh thylacinids inhabited forests (Archer *et al.* 1994; Travouillon *et al.* 2009). These regions were co-habited by an assortment of other carnivorous/omnivorous taxa, including 'giant' carnivorous rat-kangaroos (*Ekaltadeta* spp.), crocodiles (Mekosuchinae, e.g. *Baru darrowi* and *Trilophosuchus rackhami*), flightless dromornithid birds, marsupial lions (Thylacoleonidae), large bandicoots (Peramelemorphia), dasyurids (Dasyuridae), pythons (Pythonidae) and the world's oldest known venomous snakes (Wroe *et al.* 1999; Archer *et al.* 2002; Wroe 2002). Subsequent drying of the Australian continent in the late Miocene led to the gradual replacement of forest environments with open woodlands, shrublands and grasslands (Archer *et al.* 1994; Archer *et al.* 2002; Travouillon *et al.* 2009). These changes appear to broadly correlate with declining thylacinid diversity (Wroe 2003).

To date, interpretations of ecology and feeding behaviour of fossil thylacinids have been largely qualitative. This is, at least in part, because most extinct species are known only from jaw fragments and teeth. The near-complete skull of *Nimbacinus dicksoni* (Muirhead and Archer 1990; Muirhead and Wroe 1998; Wroe and Musser 2001), a medium-sized thylacinid, provides a unique opportunity to investigate feeding ecology in a fossil thylacinid.

Nimbacinus dicksoni was approximately 5 kg in body mass (Wroe 2001). Fossils of *N. dicksoni* have been recovered from Oligocene-Miocene deposits in the Riversleigh World Heritage Area, north-western Queensland and Bullock Creek, Northern Territory (Muirhead and Archer 1990; Murray and Megirian 2000; Wroe and Musser 2001; Archer *et al.* 2006). Its dentition is less specialised than that of the species of *Thylacinus*, but broadly comparable to the living dasyurid, the spotted-tailed quoll (*Dasyurus maculatus*) in the arrangement and geometry of molar shearing crests typically associated with carnivory (Wroe *et al.* 1998; Wroe and Musser 2001).

To date conflicting evidence has been presented regarding the body size of prey *N. dicksoni* may have hunted. Predictions of bite force adjusted for body mass, based on application of 2D beam theory, have suggested that *N. dicksoni* may have taken relatively large prey, as does the slightly smaller spotted-tailed quoll (Wroe *et al.* 2005). However, shape analysis of the cranium has suggested that the species may have been restricted to smaller prey and/or included a higher proportion of invertebrate food in its diet (Wroe and Milne 2007).

Vertebrate skulls are adapted to support a specific suite of functions, including food procurement and processing, protection of the brain and sensory organs (by absorbing the shock of impact) and support the face and neck muscles (Preuschoft and Witzel 2005). According to Wolff's law, bones are formed of mechanically optimised structures of maximal strength and minimal weight that accurately reflect its function (Wolff 1892). The skull shape, function and optimal use can therefore be predicted by its mechanics.

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

The loads imposed on an animal during prey acquisition and feeding play an important role in the evolution of its skull morphology (Dumont *et al.*, 2005). Testing hypotheses regarding the relationship between the form and function of skulls from extinct species requires an understanding of this relationship in living animals (Ross, 2005). A comparative biomechanics approach involving living analogues has increasingly been applied to predict the feeding behaviour of extinct species (McHenry *et al.*, 2007; Wroe *et al.*, 2007; Tseng 2008; Bell *et al.*, 2009; Wroe *et al.*, 2010; Young *et al.*, 2012). Here, to gain further insight in the feeding ecology of *N. dicksoni*, we perform a biomechanical analysis of the skull of *N. dicksoni* to predict its mechanical behaviour. We apply a powerful engineering tool called Finite Element Analysis (FEA) to determine relative stress magnitudes and distributions in the skull of *N. dicksoni* in response to loads simulating the capture and processing of prey. An analysis of the mechanical function of the *N. dicksoni* skull will benefit from comparisons with members of the extinct lineage as well as extant relatives with extensively studied diets. Skull fossils of Thylacinidae are exceedingly rare, with only three species known from fossil crania. Results are compared to those from a range of extant marsupial ‘carnivore’ species including the Tasmanian devil (*Sarcophilus harrisii*), spotted-tailed quoll and northern quoll (*Dasyurus hallucatus*). We also include the thylacine to establish whether the biomechanical performance of *N. dicksoni* more closely resembles that of this larger, more derived thylacinid than dasyurids.

Finite Element Analysis (FEA) is a three-dimensional (3D) computer modelling approach more commonly used by biologists and palaeontologists to examine and compare mechanical performance in biological structures in comparative contexts (Rayfield *et al.* 2001; Moazen *et al.* 2008; Strait *et al.* 2010; Attard *et al.* 2011; Chamoli and Wroe 2011; Oldfield *et al.* 2012). In FEA, continuous structures, such as the skull, are divided into a discrete, finite numbers of elements, allowing the prediction of mechanical behaviour for complex geometric shapes. The structure is analysed in the form of a matrix algebra problem that is solved with the aid of a computer (Thresher and Saito 1973).

Studies of feeding ecology for thylacinids have primarily focused on the most recently extinct member of the family, the thylacine, which survived in Tasmania until 1936 (Paddle 2000). Our understanding of the ecology of the thylacine is chiefly based on morphological comparison and 2D beam theory (Wroe *et al.* 2005; Wroe and Milne 2007), as well as anecdotal accounts of their behaviour in the wild (Guiler 1985; Paddle 2000; Bailey 2001). Elbow joint morphology of thylacines evidently most closely resembles that of extant ambush predators, a compromise between efficient distance locomotion and the ability to manipulate and grapple with prey (Figueirido and Janis 2011). Three-dimensional biomechanical modelling of the skull of the thylacine, extant dasyurids and an introduced Australian predator (*Canis lupus dingo*) have suggested potential limitations on prey body size (Wroe *et al.* 2007; Attard *et al.* 2011).

Morphological and biomechanical comparisons including sympatric native predators in Tasmania indicate that diet of thylacines may have overlapped considerably with the two largest extant marsupial carnivores, the Tasmanian devil and spotted-tailed quoll (Jones and Barmuta 1998; Jones and Stoddart 1998; Attard *et al.* 2011). These species represented the three largest marsupial carnivores in Tasmania at the time of European settlement.

The Tasmanian devil is today the largest living marsupial carnivore (mean adult male weight 8.7 kg, female 6.1 kg) (Bradshaw and Brook 2005). Relative to its body size, its predicted bite force is greater than that of any other extant mammal studied to date (Wroe *et al.* 2005). The robust jaw-closing muscles of the Tasmanian devil generate powerful bites capable of crushing bone and teeth (Werdelin 1986; Jones 2003; Attard *et al.* 2011). They are able to consume up to 40% of their body mass in a single meal (Pemberton and Renouf 1993) and are the only specialised scavengers among living marsupials, filling a broadly similar ecological niche to that of osteophagous hyenas (Jones and Stoddart 1998). However, they are also opportunistic hunters known to prey on a range of species, including small- to medium-sized mammals and birds that may exceed their body mass, as well as occasionally consuming plant material (Guiler 1970; Taylor 1986).

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

Quolls are represented by four extant species in Australia and two in New Guinea (Groves *et al.* 2005). The largest quoll, the spotted-tailed quoll (maximum weight 7 kg) has a broad diet consisting of a variety of taxa, mainly consisting of mammals and insects, but will occasionally feed on birds and reptiles (Edgar and Belcher 1995; Glen and Dickman 2006). They are opportunistic hunters, varying their diet in response to environmental disturbances and short-term fluctuations in prey abundance (Glen and Dickman 2006; Dawson *et al.* 2007). They are capable of catching mammals approaching or exceeding their body mass, including the common brushtail possum (*Trichosurus vulpecula*), common ringtail possum (*Pseudocheirus peregrinus*), red-necked pademelon (*Thylogale thetis*) and long-nosed bandicoot (*Perameles nasuta*) (Belcher 1995; Glen and Dickman 2006; Belcher *et al.* 2007). These larger prey species constitute a higher proportion of the diet of adult male spotted-tailed quolls, while females and immature spotted-tailed quolls more frequently feed on smaller bodied mammals (Jones 1997; Dawson *et al.* 2007). The northern quoll is the smallest and most arboreal of the four Australian quolls, weighing up to 1.2 kg (Braithwaite and Begg 1995; Strahan 1995). Although primarily insectivorous, this active hunter can feed on a variety of foods: fruits, small mammals, birds, reptiles, frogs and carrion (Belcher 1995; Oakwood 1997; Pollock 1999).

In this study we determine whether *N. dicksoni* was capable of killing large prey relative to their body size, or was restricted to catching small bodied species. By digital reconstruction of its skull and applied 3D FEA, we compare its mechanical performance with that of three extant marsupial carnivores and the thylacine. We use previously applied scaling procedures (McHenry *et al.* 2007) to account for differences in body mass, allowing for comparison of results between species. We predict that *N. dicksoni* will have similar distributions and magnitudes of craniomandibular stress to that of the similar-sized *D. maculatus*, which would suggest a dietary niche including relatively large-bodied prey. We further hypothesise that the relatively long rostrum of *T. cynocephalus* will result in higher stresses in the skull during biting and prey procurement than other dasyuromorphians.

3.3 MATERIALS AND METHODS

3.3.1 Specimens

Biomechanical performance of the skull of *N. dicksoni* (QMF36357) was compared with that of four dasyuromorphian species covering a range of craniodental morphologies and feeding ecologies. These comprised three extant dasyurids (northern quoll, TMM M-6921; spotted-tailed quoll, UNSW Z20; Tasmanian devil, AM10756), and one thylacinid (thylacine, AM1821). Institutional abbreviations are QMF (Queensland Museum Fossil), TMM (Texas Memorial Museum), UNSW (University of New South Wales) and AMNH (American Museum of Natural History).

We generated 3D FEMs of each skull on the basis of computed tomography X-ray (CT) scan data. Digimorph (University of Texas; <http://www.digimorph.org>) was the source of CT data of a northern quoll skull (0.0784 mm slice thickness, 0.0784 mm inter-slice distance). Other skulls were scanned in a Toshiba Aquillon 16 scanner (Toshiba Medical Systems Corporation, Otawara, Tachigi, Japan) at the Mater Hospital, Newcastle, NSW (1 mm slice thickness, 0.8 mm inter-slice distance, 240 mm field of view). Surface meshes were generated from the CT data using Mimics 13.2 and converted to FEMs in STRAND7 (ver. 2.4) following previously established protocols (McHenry *et al.* 2007; Wroe 2008; Attard *et al.* 2011).

3.3.2 Digital reconstruction of *Nimbacinus dicksoni*

For detailed descriptions of *N. dicksoni* see Muirhead and Archer (1990) and Wroe and Musser (2001). The skull is very well preserved, although some regions are absent or damaged. Specifically, some damage/deformation is present at the postorbital processes, frontal, maxillary and nasal bones, which are compressed dorsoventrally. These damaged regions were reconstructed according to the morphology of surrounding bone regions once the damaged areas had been isolated and deleted (Benazzi *et al.* 2011). Regions of bone that showed only minor damage were smoothed to create a coherent surface mesh for later solid meshing.

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

The right and left dentaries were largely intact but missing the superior regions of the coronoid processes, the temporomandibular joints (TMJ), condyles and angular processes. The anterior of the mandible is broken, separating both dentaries. We used the right dentary as a basis for reconstruction because its dentition was more far more complete, with only the incisors missing. We used a surface mesh of the right dentary of the spotted-tailed quoll to reconstruct posterior regions of the right dentary of *N. dicksoni*. The spotted-tailed quoll was chosen as its mandible was most similar in shape to that of *N. dicksoni*, thereby minimising the extent of warping needed (and see below).

Reconstruction involved scaling the dentary of the spotted-tailed quoll to the same size as that of *N. dicksoni*, the missing posterior region of the *N. dicksoni* specimen was then isolated on the spotted-tailed quoll specimen and the mesh fitted to the existing structure in the mesh of *N. dicksoni* using Iterative Closest Point (ICP) registration. ICP is an algorithm that revises the transformation needed to minimize the distance between the points of two partially overlapping meshes. This process re-oriented the spotted-tailed quoll dentary in accordance with the morphology of the *N. dicksoni* dentary (Besl and McKay 1992). The anterior region of the spotted-tailed quoll dentary was deleted and the posterior region 'warped' so that overlapping regions of the coronoid process and angular process from the spotted-tailed quoll mesh matched the existing morphology of *N. dicksoni* (method followed Oldfield *et al.* 2012; Parr *et al.* 2012). Similarly, the TMJ was warped so that the condyle articulated with and fitted the cotyle of the cranium. The left dentary was created by mirroring the reconstructed right dentary. These were positioned so that the condyles articulated with the cranium, the outer surfaces of the lower molars made contact with the inner surface of the upper molars, and the tips of the lower canines aligned with their 'sockets' in the cranium (see Figure 3.1).

It is important to note that the shape of the warp was determined by the existing regions of the *N. dicksoni* dentary, the need for the condyle to articulate with the cotyle at the TMJ and for the coronoid process to fit between the cranium and the zygomatic arch. These requirements act as restraints on the warp such that the shape of the starting mesh (spotted-tailed quoll in this case) is not important in the sense that the warping process

would always end with a similarly shaped posterior region of the mandible regardless of which taxon was used. We reiterate that spotted-tailed quoll was used because it was the most similar in shape and therefore required less ‘warping’.

The *N. dicksoni* cranium was missing the following teeth: left I1-4, right I1, 3-4, both right and left C1 and right LDP2. The existing I2 and LDP2 on *N. dicksoni* were mirrored. All incisors were missing from the mandible. Incisors from the spotted-tailed quoll were isolated, scaled and fitted into the empty tooth sockets on *N. dicksoni*. Figure 3.1 displays the completed reconstruction of *N. dicksoni*.

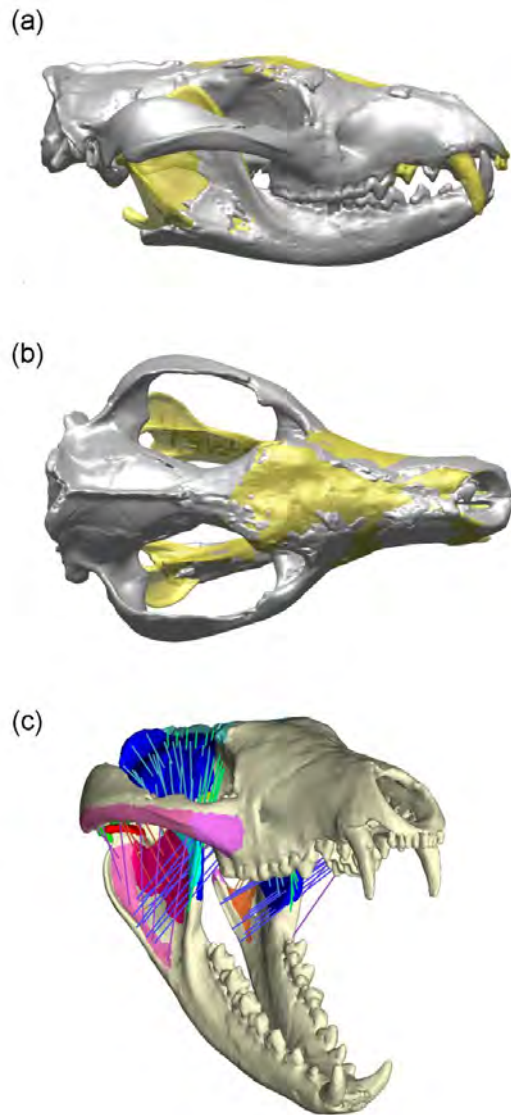


Figure 3.1 Digital reconstruction of *Nimbacinus dicksoni*. Original (grey) and reconstructed 3D (yellow) in (a), lateral view; (b), dorsal view; (c), Pre-processed FE model of *N. dicksoni*, showing jaw musculature represented by trusses.

3.3.3 Finite element models

The assembly of FEMs largely follows previously published procedures (McHenry *et al.* 2007; Wroe *et al.* 2010; Attard *et al.* 2011). As the skull of *N. dicksoni* was not fully preserved, we were unable to assign multiple material properties to the digital reconstruction without introducing additional assumptions. Consequently, as in most FEA incorporating fossil material (Rayfield *et al.* 2001; Rayfield 2007; Wroe *et al.* 2010), all FEMs were homogeneous and assigned a single material property for cortical bone ($E = 13.7$ GPa, $\nu = 0.3$, where E is Young's modulus of elasticity and ν is Poisson's ratio) (Cook *et al.* 1982) in order to enable direct comparisons between species. Young's modulus is a measure of stiffness in the material, whereas Poisson's ratio is used for stress and deflection analysis of structures. Each homogenous model was comprised of four-noded tetrahedral elements or 'bricks'.

3.3.4 Modeling masticatory muscle forces

Jaw elevators were modeled as seven muscle subdivisions: *temporalis superficialis*, *temporalis profundus*, *masseter superficialis*, *masseter profundus*, *zygomaticomandibularis*, *pterygoideus internus* and *pterygoideus externus* (Wroe *et al.* 2007). Proportions used for each jaw muscle division were based on muscle mass proportions from a dissected Virginia opossum (*Didelphis virginiana*) (Turnbull 1970). Muscle forces were predicted on the basis of maximum cross-sectional areas (CSA) using the 'dry skull' method (Thomason 1991). To improve the accuracy of our CSA measurements, we used our FEMs to record the co-ordinates of ~100 nodes at the perimeter of each muscle cross sectional area (Chamoli and Wroe 2011). The FEM was moved to the correct orientation described by Thomason (1991) to select nodes outlining the CSA. The node co-ordinates were then plotted in plane geometry software, GEUP 5 (version 5.0.3) and connected to form a multi-sided polygon. The area of the polygon was measured to estimate the CSA of each major jaw closing muscle. Data for muscle forces are in Table 3.1. To minimise the incidence of artifacts at bite points and muscle origin and insertion areas, surface regions at these sites were tessellated using a network of stiff beam elements (Clausen *et al.* 2008).

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

Table 3.1 Muscle forces used for each jaw muscle division in unscaled intrinsic models of the northern quoll, spotted-tailed quoll, Tasmanian devil, *Nimbacinus dicksoni* and thylacine. These were calculated using muscle mass proportions from a dissected Virginia opossum (Turnbull 1970). Muscle forces were scaled for a bilateral canine bite by multiplying the muscle force by the ratio between bite force estimated using body mass regressions and maximum bite force estimated from the unscaled FEM.

	Muscle force distribution (in Newtons) of jaw muscle groups				
	Northern quoll	Spotted-tailed quoll	<i>Nimbacinus dicksoni</i>	Tasmanian devil	Thylacine
<i>Temporalis superficialis</i>	2.13	6.73	11.24	11.84	26.78
<i>Temporalis profundus</i>	2.13	6.71	11.21	11.80	26.70
<i>Masseter superficialis</i>	2.06	6.51	10.87	11.45	25.90
<i>Masseter profundus</i>	2.01	6.34	10.58	11.14	25.20
<i>Zygomaticomandibularis</i>	2.31	7.29	12.17	12.81	28.98
<i>Pterygoideus internus</i>	2.26	7.13	11.90	12.53	28.35
<i>Pterygoideus externus</i>	2.01	6.34	10.58	11.14	25.20
TOTAL	14.91	47.04	78.56	82.72	187.11
Scaled muscle force	0.67	0.52	0.59	0.68	1.02

3.3.5 Restraints, loading conditions and scaling

Dasyurids frequently use a penetrating canine bite to kill prey (Fleay, 1932; Jones, 1995; Jones and Stoddart, 1998; Pellis and Nelson, 1984; Pellis and Officer, 1987) which involves the application of a bending load (Dumont and Herrel, 2003). We simulated bilateral canine biting (intrinsic load) and four extrinsic loads to simulate loads generated by struggling prey (axial twist, lateral shake, pullback and dorsoventral) for all models using protocols described by Attard *et al.* (2011) and following McHenry *et al.* (2007). Extrinsic loads were modelled without applying bite forces so as to clearly reveal the different influences of each separate loading (McHenry *et al.* 2009). A gape angle of 35° was applied in all linear static load cases.

A considerable size range exists between specimens considered in the present study. The relationship between bite force and body mass is negatively allometric (Wroe *et al.* 2005; Christiansen and Wroe 2007). To account for differences in body mass, load cases were solved following the scaling procedures of McHenry *et al.* (2007). Here, for each model, an estimate of bite force was made based on regression of body mass to bite force for dasyuromorphians [$z = 0.6998 (\log y) + 1.8735$, where $y = \text{mass (g)}$ and $z = \text{bite force at canines (N)}$] (Wroe *et al.* 2005), with body mass for each specimen predicted using the equation based on lower molar row length [$\log y = -1.075 + 3.209(\log x)$, where $x = \text{lower molar length (mm)}$, and $y = \text{mass (g)}$] as presented by Myers (2001). Muscle forces were then scaled for each specimen to achieve bite forces predicted on the basis of body mass. FEMs were solved using these scaled muscle forces. The maximum bite force measured in Newtons (N) was estimated for a bilateral canine bite using FEMs with unscaled, specimen-specific estimated muscle forces.

An H-frame connecting the canines of the upper and lower jaws was used to apply extrinsic forces, with forces applied at the centre of the frame (Wroe *et al.* 2007; Attard *et al.* 2011). The force (N) applied to extrinsic loads was equivalent to 100 times the animal's estimated body mass for an axial twist, and 10 times the animal's estimated body mass for a lateral shake, pullback and dorsoventral (Myers 2001).

Von Mises (VM) stress is a good predictor of failure in ductile materials such as bone (Nalla *et al.* 2003; Tsafnat and Wroe 2010) and VM stress is used here as a metric for comparison between models following Attard *et al.* (2011). Nodes were selected at equidistant points along the mid-sagittal plane, zygomatic arch and mandible (Figure 3.2) and at each node values were calculated by averaging VM stress recorded in the surrounding elements to assess changes in stress magnitudes and distributions under different loadings.

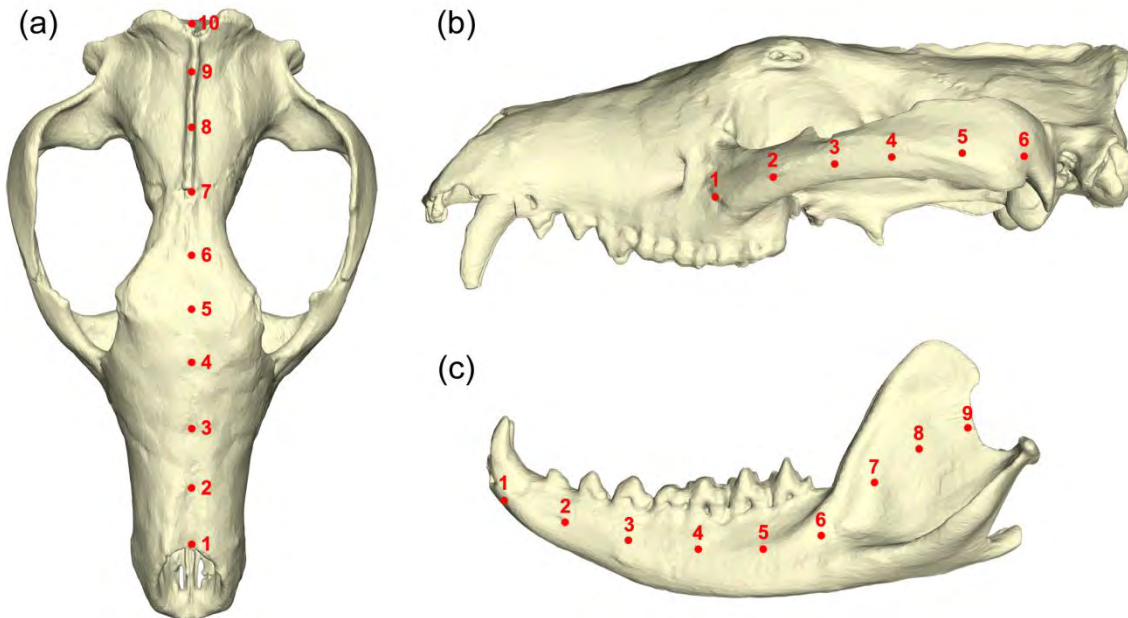


Figure 3.2 Position of nodes selected at equidistant points along the (a) mid-sagittal plane, (b) zygomatic arch, and (c) mandible to measure distribution of VM stress for each loading case.

3.4 RESULTS

The predicted body mass (kg) of each species was generally within the expected range for each of the extant species (Table 3.2). Body mass estimates ranged from 0.78 kg for the northern quoll, up to 32.49 kg for the thylacine. However, the body mass estimated for the Tasmanian devil of 14.20 kg was slightly above the upper limit observed for males (13 kg) (Owen and Pemberton 2005), possibly because the teeth and skull are relatively large in this species. Predicted maximum muscle forces for *N. dicksoni* (407 N) were relatively high, being intermediate between the spotted-tailed quoll (300 N) and Tasmanian devil (706 N) (Table 3.3).

Table 3.2 Predicted body mass (kg) calculated using the regression equation for dasyuromorphians provided by Myers (2001) based on lower molar row length. Temporalis and masseteric muscle forces (N) were calculated based on cross-sectional area (Thomason 1991).

Species	Predicted body mass (kg)	Temporalis muscle force (N)	Masseteric muscle force (N)	Total muscle force (N)
Northern quoll	0.78	67.60	55.89	123.49
Spotted-tailed quoll	2.88	211.00	178.67	389.67
<i>Nimbacinus dicksoni</i>	5.25	282.38	368.33	650.71
Tasmanian devil	14.20	300.46	384.73	685.19
Thylacine	32.49	706.64	843.21	1, 549.86

Table 3.3 Maximum bite forces (N) for unscaled homogeneous models during a bilateral canine for the northern quoll, spotted-tailed quoll, Tasmanian devil, *Nimbacinus dicksoni* and the thylacine.

	Maximum bite force (N) for a bilateral canine
Northern quoll	93.70
Spotted-tailed quoll	299.81
<i>Nimbacinus dicksoni</i>	406.81
Tasmanian devil	705.84
Thylacine	835.94

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

The thylacine displayed comparatively high levels of VM stress in the cranium and mandible for most simulations (Figure 3.3-3.6). This is consistent with results of Attard *et.al.* (2011), who applied 3D FEA to compare mechanical performance in the skull of the thylacine with extant Tasmanian marsupial carnivores. The northern quoll showed relatively high levels of stress in the posterior of the mandible for a canine bite (Figure 3.3a), and along the ventral surface of the ramus for most extrinsic loads (Figure 3.5a-c).

The regions of highest stress along the dentary of *N. dicksoni* were located at the coronoid fossa and condylar process (Figure 3.3e). These regions of peak stress may be in part an artifact of reconstruction. Otherwise the dentary of *N. dicksoni* revealed similar stress patterns for a bilateral bite to the spotted-tailed quoll (Figure 3.3). The distribution of stress for *N. dicksoni* in the cranium in response to a bilateral bite was intermediate between the Tasmanian devil and spotted-tailed quoll (Figure 3.4). The magnitudes of stress along the mid-sagittal plane of *N. dicksoni* were slightly higher than for the Tasmanian devil and lower than for the spotted-tailed quoll (Figure 3.3g).

The highest stress in the cranium occurred within the posterior regions of the zygomatic arches for all species in response to a bilateral canine bite (Figure 3.4). The thylacine was the only species to show two distinct peaks in stress for a bilateral bite along the mid-sagittal crest (Figure 3.4f). These stress points occurred at the temporal ridge and at the most narrowed region of the nasal (Figure 3.4e). Von Mises stress measured along the mid-sagittal crest for a bilateral bite revealed one point of peak stress halfway along the frontal of the spotted-tailed quoll, Tasmanian devil and *N. dicksoni* and at the temporal ridge for the northern quoll (Figure 3.4).

Stress was quite evenly distributed along the dentaries for all species in response to lateral shaking and axial twisting, with the exception of the northern quoll, wherein stresses peaked anteriorly (Figure 3.5u, v). An axial twist resulted in much higher levels of stress along the mid-sagittal crest for the northern quoll compared to all other

species, and peaked at the anterior of the nasal and at the frontal (Figure 3.6b). The northern quoll also had notably higher levels of stress along the mandible than the other species for a lateral shake and axial twist (Figure 3.5u, v), while the Tasmanian devil and thylacine showed higher levels of stress along the mandible for a pullback and dorsoventral shake (Figure 3.5w, x). Two points of peak stress were apparent along the dentary for the thylacine in these two simulations; one at the most anterior point, and the second at the coronoid fossa. Stress distribution along the dentary of the Tasmanian devil followed a similar trend for a pullback and dorsoventral shake; peaking at the ramus inferior to M1 then gradually decreasing posteriorly.

3. PREY SIZE PREFERENCE OF *NIMBACINUS DICKSONI*

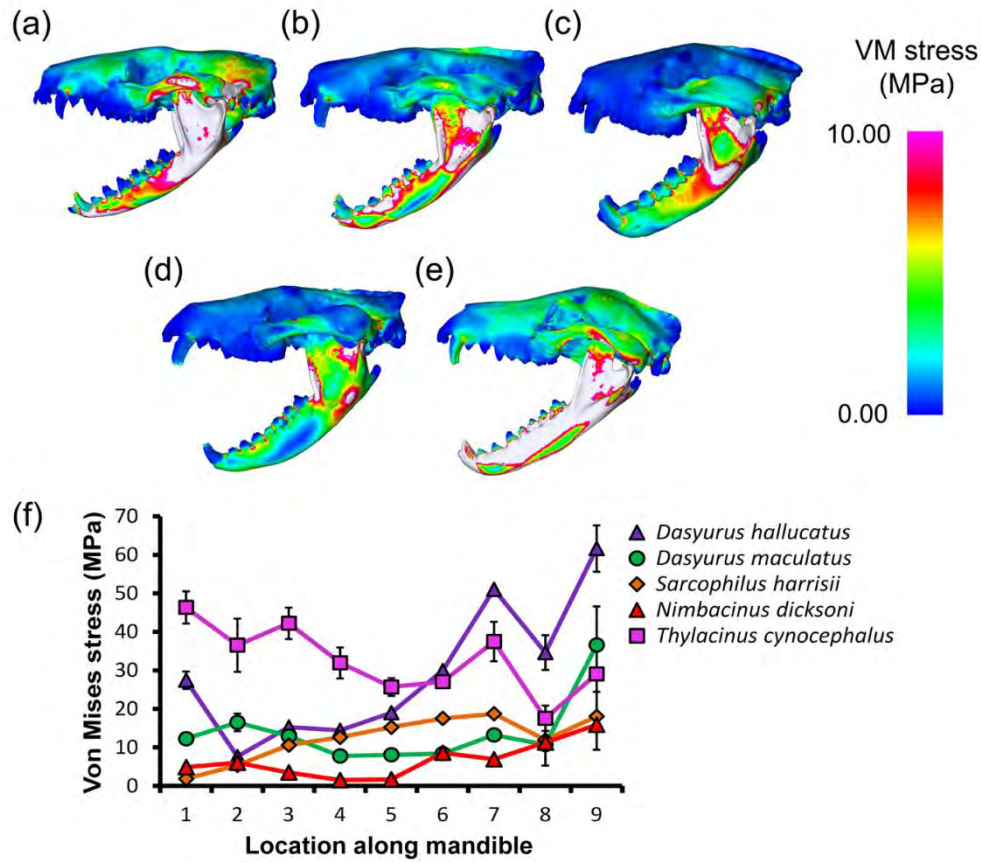


Figure 3.3 Stress distribution along mandible during a bilateral canine bite. Stress distribution in scaled homogeneous models with bite force scaled based on theoretical body mass during a bilateral canine bite for (a) northern quoll, (b) spotted-tailed quoll, (c) Tasmanian devil, (d) *Nimbacinus dicksonim*, and (e) thylacine. (f) Distribution of von Mises (VM) stress was measured from anterior to posterior along the mandible.

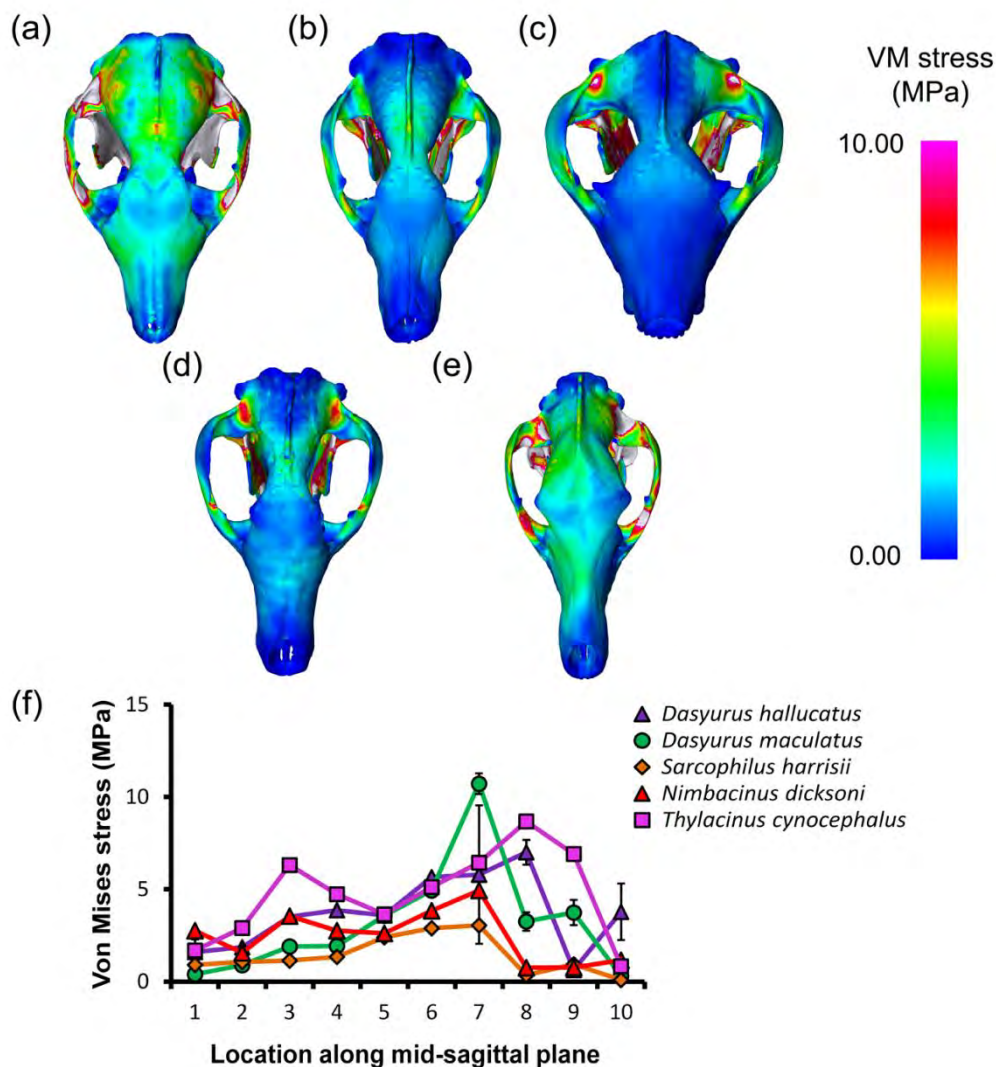


Figure 3.4 Stress distribution along mid-sagittal crest during a bilateral canine bite. Stress distribution in homogeneous models with bite forces scaled based on predicted body mass during a bilateral canine bite for (a) northern quoll, (b) spotted-tailed quoll, (c) Tasmanian devil, (d) *Nimbacinus dicksoni*, and (e) thylacine. (f) Distribution of von Mises (VM) stress was measured from anterior to posterior along the mid-sagittal plane.

3. PREDICTION OF DIET OF *NIMBACINUS DICKSONI*

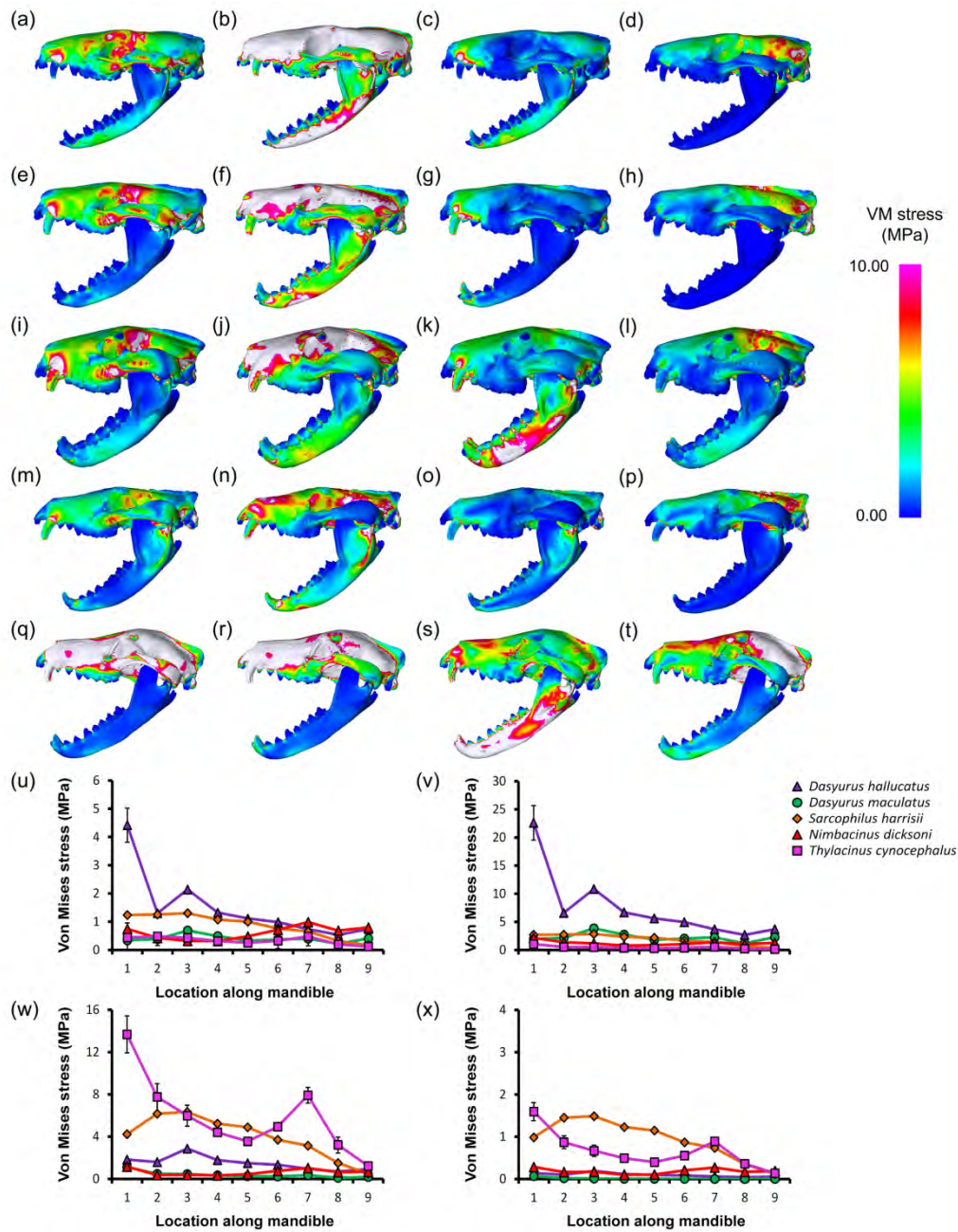


Figure 3.5 Stress distribution along mandible for extrinsic loads. Stress distribution in scaled homogeneous models with bite force scaled based on predicted body mass during a (a), (e), (i), (m), (q) lateral shake, (b), (f), (j), (n), (r) axial twist, (c), (g), (k), (o), (s) pullback, and (d), (h), (l), (p), (t) dorsoventral. Species compared were (a-d) northern quoll, (e-h) spotted-tailed quoll, (i-l) Tasmanian devil, (m-p) *Nimbacinus dicksoni*, and (q-t) thylacine. Distribution of von Mises (VM) stress was measured from anterior to posterior along the mandible for a (u) lateral shake, (v) axial twist, (w) pullback, and (x) dorsoventral.

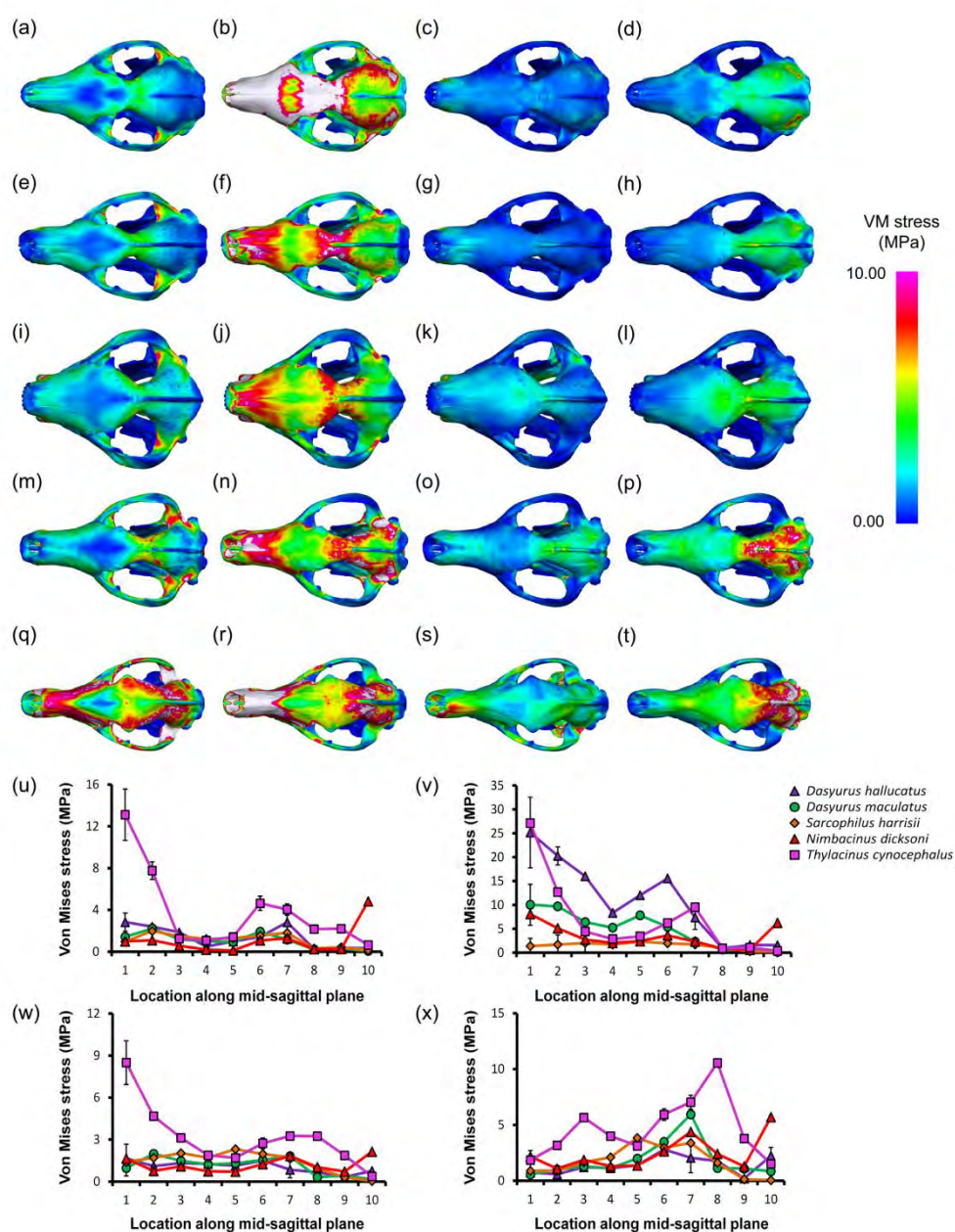


Figure 3.6 Stress distribution along mid-sagittal crest for extrinsic loads. Stress distribution in scaled homogeneous models with bite force scaled based on predicted body mass during a (a), (e), (i), (m), (q) lateral shake, (b), (f), (j), (n), (r) axial twist, (c), (g), (k), (o), (s) pullback, and (d), (h), (l), (p), (t) dorsoventral. Species compared were (a-d) northern quoll, (e-h) spotted-tailed quoll, (i-l) Tasmanian devil, (m-p) *Nimbacinus dicksoni*, and (q-t) thylacine. Distribution of von Mises (VM) stress was measured from anterior to posterior along the jaw for a (u) lateral shake, (v) axial twist, (w) pullback, and (x) dorsoventral.

3.5 DISCUSSION

Differences in biomechanical performance between the three extant dasyurids included in this study appear consistent with their respective known feeding behaviours. The northern quoll showed comparatively higher levels of stress in most simulations than the Tasmanian devil and spotted-tailed quoll. The northern quoll eats invertebrates and other relatively small prey (Belcher 1995; Oakwood 1997; Pollock 1999), which may not require adaptation to sustain the full range of extrinsic loads simulated here. This species shows particularly high VM stress in axial twisting. However, it performs relatively well under pull-back loading, may be linked to a capacity for pulling invertebrates from the ground. Observational studies on wild northern quolls will be required to confirm the functional role of their skull in prey acquisition. Overall consistencies found between known prey size and biomechanical performance for extant dasyuromorphians underscore the potential value of projections based on comparative FEA for extinct taxa.

Our comparative biomechanical modelling of dasyuromorphian skulls suggests considerable differences in the predatory behaviour between the two thylacinids considered here. Our 3D based results indicate that the Oligo-Miocene *N. dicksoni* had a high bite force for its size, comparable to that of extant dasyurids known to specialise on relatively large prey, the spotted-tailed quoll and Tasmanian devil (Belcher 1995; Jones and Barmuta 1998). In light of comparable levels of carnassialisation in the cheektooth dentition with the spotted-tailed quoll, and a lack of obvious dental specialisation consistent with regular bone-cracking, our results suggest a predominantly carnivorous diet for *N. dicksoni* that may have included relatively large prey.

Although our FEA results for *N. dicksoni* show a capacity to kill prey approaching or exceeding its own body mass, its prey range may have been limited by competition with sympatric carnivores. The extent of niche overlap and competition within this ancient, large carnivore community may have been partially alleviated by occupying different

habitats and specialising in different hunting strategies. Variation in the mode of killing and habitat use of each carnivore may have made them suited to particular types of prey, and allow partitioning of food resources. The recovery of a near complete skeleton of *N. dicksoni* (Muirhead and Archer 1990) will provide further information on the locomotion and predatory behaviour based on postcranial material; for example, was *N. dicksoni* as arboreal as the extant spotted-tailed quoll?

Differences in mechanical performance suggest that the thylacine is unusual relative to other dasyuromorphians, including, *N. dicksoni*, as indicated by distinctly higher VM stresses than all other species in response to each loading case. These results further support the contention by Attard *et al.* (2011) that niche breadth of thylacines may have been more limited and that it likely preyed on small- to medium-sized vertebrates such as wallabies, possums and bandicoots.

Although measures of skull performance in response to forces imposed by struggling prey revealed closer similarity between the fossil thylacinid *N. dicksoni* and large extant carnivorous dasyurids, than with the thylacine, there were differences. Our reconstruction suggests that the TMJ was more elevated in *N. dicksoni* than in the spotted-tailed quoll, and higher relative to the height of the cheektooth row. The precise positioning of the TMJ could influence masticatory function (Greaves 1980). The TMJ is a complex joint and is important for occlusion and mastication (Breul *et al.*, 1999; Hylander, 1979). The position of the TMJ can influence bite strength and muscle activation (Hickman and Cramer, 1998). Trends in the position of the TMJ along the anterior-posterior axis have been observed in terrestrial mammalian carnivores (Wroe and Milne, 2007). Conclusive determination must await the discovery of more complete cranial material.

Morphological evidence from past studies further demonstrates diversity within this family. The smallest thylacinid, *Muribacinus gadiyuli*, is believed to have fed on relatively small vertebrates because it lacks dental features present in large prey

specialists (e.g. robust protoconids and brachycephalisation) such as similarly sized spotted-tailed quoll (Wroe 1996). The only dasyurid cranial specimen dating from pre-Pliocene deposits was recovered in Riversleigh. The archaic dasyurids were no longer present by the Pliocene, and modern genera such as *Planigale* and *Sminthopsis* begin to appear (Archer *et al.*, 2000). The variety of feeding behaviours among thylacinids may have helped facilitate their co-existence within different ecological niches that were later filled or displaced by diversifying carnivorous dasyurids.

3.6 ACKNOWLEDGEMENTS

This research would not have been possible without Australian Research Council support to M. Archer, and S. J. Hand, resulting in recovery and preparation of the Riversleigh *Nimbacinus* specimen used in this study. We thank S. Ingleby from the Australian museum for providing several comparative specimens and the makers of Digimorph for access to CT scan data. This research was funded by Australian Research Council (DP0666374 and DP0987985) and University of New South Wales Internal Strategic Initiatives Grant to S. Wroe.

Chapter 4

NEW INSIGHT FROM THE OLD: USING STABLE
ISOTOPES TO ASSESS MARSUPIAL CARNIVORE
VULNERABILITY TO ANTHROPOGENIC IMPACTS



4.1 ABSTRACT

Maximum prey size and competition are important indicators of species extinction vulnerability, particularly among large mammalian carnivores. The thylacine (*Thylacinus cynocephalus*) was the largest marsupial carnivore to survive up to European settlement in Tasmania and became extinct in 1936. Factors contributing to their extinction have been debated and will benefit from an improved understanding of their feeding ecology. The dietary composition of Tasmanian thylacines was explored using carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) stable isotope ratios of preserved tissues from thylacines and potential prey species. Tissues from sympatric marsupial carnivores were also collected to test for niche overlap with the thylacine. Furthermore, we integrate historical and recent stable isotope data of marsupial carnivore tissues to assess long-term shifts in their diet and habitat use following European arrival in Tasmania. Using a multiple-source mixing model (MixSIR), we speculate that hunting of medium-sized (13-30 kg) mammals was likely to be a major element of thylacine subsistence in Tasmania. This may have included the common wombat (*Vombatus ursinus*), red-necked wallaby (*Macropus rufogriseus*) and Tasmanian pademelon (*Thylogale billardierii*). Severe overexploitation of predominant prey species by humans since the early 19th century may have had bottom-up effects on thylacine abundance.

There was a high degree of trophic niche overlap both between thylacine sexes and across other members of the marsupial carnivore guild, which may have resulted in high competitive pressure. Alternatively, there may have been little intraguild competition, as stable isotopes give an averaged indication of the prey used, rather than identifying specific prey types. Differences in $\delta^{13}\text{C}$ values between the thylacine and the two largest remaining marsupial carnivores, the Tasmanian devil (*Sarcophilus harrisii*) and spotted-tailed quoll (*Dasyurus maculatus*), suggest differences in habitats utilised by these predators, which may have helped alleviate the pressure of intraguild competition. The

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

Tasmanian devil and spotted-tailed quoll showed shifts in their $\delta^{15}\text{N}$ values following European settlement. This may be attributed to changing prey availability or changes in the source nitrogen at the base of the food as a result of landscape modification.

Lowering of Tasmanian devil and spotted-tailed quoll keratin $\delta^{13}\text{C}$ values over ~180 years is suggestive of a transition in habitats used as animals moved from predominately mosaic grassland landscapes towards denser vegetation. This increased understanding of top predator ecology and their response to disturbances in the ecosystem are relevant for the conservation and preservation of Australia's last few remaining large marsupial carnivores that face similar challenges as the thylacine for their survival.

4.2 INTRODUCTION

Large mammalian carnivores are being lost from the planet at an astonishing rate (Morrison *et al.* 2007). This is primarily due to humans activities such as exploitation (Brashares *et al.* 2001) and the removal and fragmentation of habitats (Woodroffe 2000; Ceballos and Ehrlich 2002; Sechrest 2003; Munday 2004). Specialised predators are more vulnerable to environmental fluctuations and so are at a higher risk of extinction than generalised predators (Muñoz-Durán 2002). Niche overlap among predators can lead to an increase in competition and a reduction in available resources in the ecosystem (Charnov *et al.* 1976). The loss of high trophic level species can cause cascading trophic effects and alter trophic interactions (Crooks and Soulé 1999; Berger *et al.* 2001b; Côté *et al.* 2004; Estes *et al.* 2011). Understanding how biological species traits have contributed to past and present population declines and extinctions will provide us with a long-term perspective, which is essential to predict future changes in species and food web structure.

The island of Tasmania contains the largest marsupial carnivores in the world (Jones 1997) and provides a unique system to study resource specialisation, competition and extinction risk among large predators. At the time of European settlement, this guild comprised of the thylacine (*Thylacinus cynocephalus*), Tasmanian devil (*Sarcophilus harrisii*), spotted-tailed quoll (*Dasyurus maculatus*) and eastern quoll (*Dasyurus hallucatus*). The thylacine became extinct in 1936 and all other species are now in danger of extinction (IUCN 2012). The likelihood of endemic mammal extinctions on islands are associated with a combination of factors, including island area and location, species ecology and body size (Alcover *et al.* 2002).

The status of the thylacine as a meat specialist has been universally agreed upon, but whether they were a predator of relatively large mammalian prey remains a long-running subject of debate (Case 1985; Jones and Stoddart 1998; Paddle 2000; Johnson and Wroe 2003; Wroe *et al.* 2007; Attard *et al.* 2011). Thylacines were by far the largest

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

marsupial carnivore to survive up to European settlement in Australia; with an average head to tail length of 1.2 metres and average body weight of 30 kg (Watts 1993; Paddle 2000). As hunting is an energy consuming activity, large predators may be able to optimise their net energy gain by targeting larger prey items (Caraco and Wolf 1975). A striking transition in prey selection occurs at a predator mass of about 21.5 kg, with those species below the threshold taking prey of <45% their mass and those above taking prey of >45% their mass (Carbone *et al.* 1999). As a large-bodied predator, relying on prey <13 kg in mass would have been energetically constraining for thylacines: their food may have been inadequate to support them unless smaller prey species were abundant (Attard *et al.* 2011). However, pervasive selection for larger prey size in large carnivores can lead to dietary specialisation and increased vulnerability to extinction (Van Valkenburgh 2004).

The stable isotopes ratio of carbon ($^{13}\text{C}/^{12}\text{C}$; $\delta^{13}\text{C}$) and nitrogen ($^{15}\text{N}/^{14}\text{N}$; $\delta^{15}\text{N}$) in animal tissues have been increasingly used to provide a direct indication of the foraging ecology and habitat of extinct species (Vogel and Van der Merwe 1977; Lee-Thorp *et al.* 1989; Hobson and Montevecchi 1991; Hilderbrand *et al.* 1996; Richards *et al.* 2000; Fernández-Mosquera *et al.* 2001; Clementz *et al.* 2003; Feranec 2003; 2004; MacFadden *et al.* 2004; Kohn *et al.* 2005; Koch *et al.* 2007; Crowley *et al.* 2011). By measuring stable isotope ratios of various fauna in a paleo-ecosystem, it is possible to examine aspects of food web structure (Peterson and Fry 1987). The step-wise enrichment of ^{15}N in a consumer's tissue from their diet (3-5‰) provides a robust measure of an animal's trophic position (DeNiro and Epstein 1981; Minagawa and Wada 1984; Vander Zanden *et al.* 1997; Post 2002). In terrestrial ecosystems, differences in $\delta^{13}\text{C}$ are related to the photosynthetic pathway of plants at the base of the food web and are used to reflect an animal's habitat (DeNiro and Epstein 1978).

Human-caused disturbances are a fact but the ecological consequences are uncertain. Monitoring stable isotopic signatures of high trophic level organisms are preferentially used to indicate overall changes in the food web structure and vegetation, as they more accurately reflect average isotopic fluctuations across the environment than lower trophic level organisms (Bump *et al.* 2007). Multi-decadal variability in predator isotopic composition has been investigated extensively in marine and aquatic systems (Burns *et al.* 1998; Hirons *et al.* 2001; Hilton *et al.* 2006; Newsome *et al.* 2007; Cerling *et al.* 2008; Christensen and Richardson 2008), yet has been rarely studied in terrestrial systems (Féret *et al.* 2003; Bump *et al.* 2007). Due to the lack of empirical evidence, the historical vegetation structure of the Tasmanian landscape and its impact on the ecology of large predatory marsupials remains widely unknown (Seamster 2010).

The aim of this study is to (i) identify prey body size range and proportional composition of prey items in the thylacine diet, (ii) assess niche partitioning within and among sympatric marsupial carnivores, and (iii) assess long-term shifts in marsupial carnivore diet and habitat use in Tasmania. To infer the diet of the thylacine, we analysed stable isotope values of preserved tissues from thylacines and potential prey species present in Tasmania. The diet of thylacines was compared with sympatric marsupial carnivores by measuring the breath of each species' diet (i.e. niche width, Schoener 1971) to assess their potential for competition. Due to their high degree of scavenging, we predict that the Tasmanian devil would show higher variability in their trophic niche compared to other marsupial carnivores. As a sexually dimorphic species, we hypothesise that the thylacine would have intersexual differences in their feeding and habitat preferences. The isotopic ecology of museum preserved tissues from marsupial carnivores will provide important baseline data about Tasmanian native vegetation and provide concurrent information about the long-term shifts in the ecology of Tasmania's marsupial carnivore guild.

4.3 MATERIALS AND METHODS

4.3.1 Specimens and tissues

4.3.1.1 *Marsupial carnivores*

Tissues were collected from thylacine ($n=34$), Tasmanian devil ($n=59$), spotted-tailed quoll ($n=57$) and eastern quoll ($n=49$) specimens for stable isotope analysis. All specimens were from wild stock (i.e. died/killed in the wild) and were derived from Tasmania. Only tissues from adult specimens were included in this study as juveniles may have enriched $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values from weaning (Fogel *et al.* 1989; Millard 2000; Fuller *et al.* 2006). Due to the limited number of available wild thylacine and sympatric carnivore museum-archived specimens, multiple different tissues were acquired for this study. Bone was preferentially used, however, if bone was unavailable or too invasive to sample, we instead collected hair or a vibrissa from the specimen. Stable isotope results were pooled together for some analysis (see section 4.3.7 and 4.3.9) where there was an insufficient sample size to analyse each tissue type (collagen and keratin) separately. Differences in stable isotope values between tissues were accounted for prior to statistical analysis for pooled tissue types (see section 4.3.6). A list of all marsupial carnivore specimens, including museum records and the tissue type used in this study are displayed in Appendix A1.

4.3.1.2 *Potential prey species*

We collected skin tissue from 12 potential prey species of the thylacine mentioned in anecdotal accounts (Guiler 1985; Paddle 2000). Prey species were from a range of feeding niches (herbivore-insectivore-carnivore) and body sizes ($<1\text{-}66\text{ kg}$) (Appendix A3). Sample size and mean ($\pm\text{SD}$) $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for the thylacine and potential prey species are shown in Figure 4.1. We limited our analysis to prey specimens collected between 1905 and 1964 in Tasmania to control for potential sources of variation in prey isotopic values over time (e.g. primary production shifts, nutrient

inputs or changes in the prey dietary preferences) (Yoshioka *et al.* 1994). Museum records of all prey specimens included in this study are listed in Appendix A2. Due to the rarity of preserved prey specimens from this era and location, only a relatively small number of samples (1-6 specimens per species) were available for this study.

Prey muscle tissue is preferentially used in stable isotope food web studies as its $\delta^{15}\text{N}$ values more closely reflects that of the whole body than other tissues (Kelly 2000; Sotiropoulos *et al.* 2004; Becker *et al.* 2007). However, sampling muscle is not possible in the reconstruction of most historic food webs, where only collagen or keratin tissues have been preserved (Hilderbrand *et al.* 1996). Studies on laboratory mice (*Mus musculus*), domestic rabbits (*Oryctolagus cuniculus*), and American black bears (*Ursus americanus*) fed on a fixed diet, as well as humpback whales (*Megaptera novaeangliae*) from strandings showed no difference in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ between skin collagen and lipid-extracted muscle (Todd *et al.* 1997). Muscle and skin collagen also have similar turnover rates, and are expected to reflect the integration of diet over the most recent few months of the animal's life prior to collection (Tieszen *et al.* 1983; Rucklidge *et al.* 1992; Hobson 1999). For this reason, we decided to analyse skin tissue for all prey specimens.

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

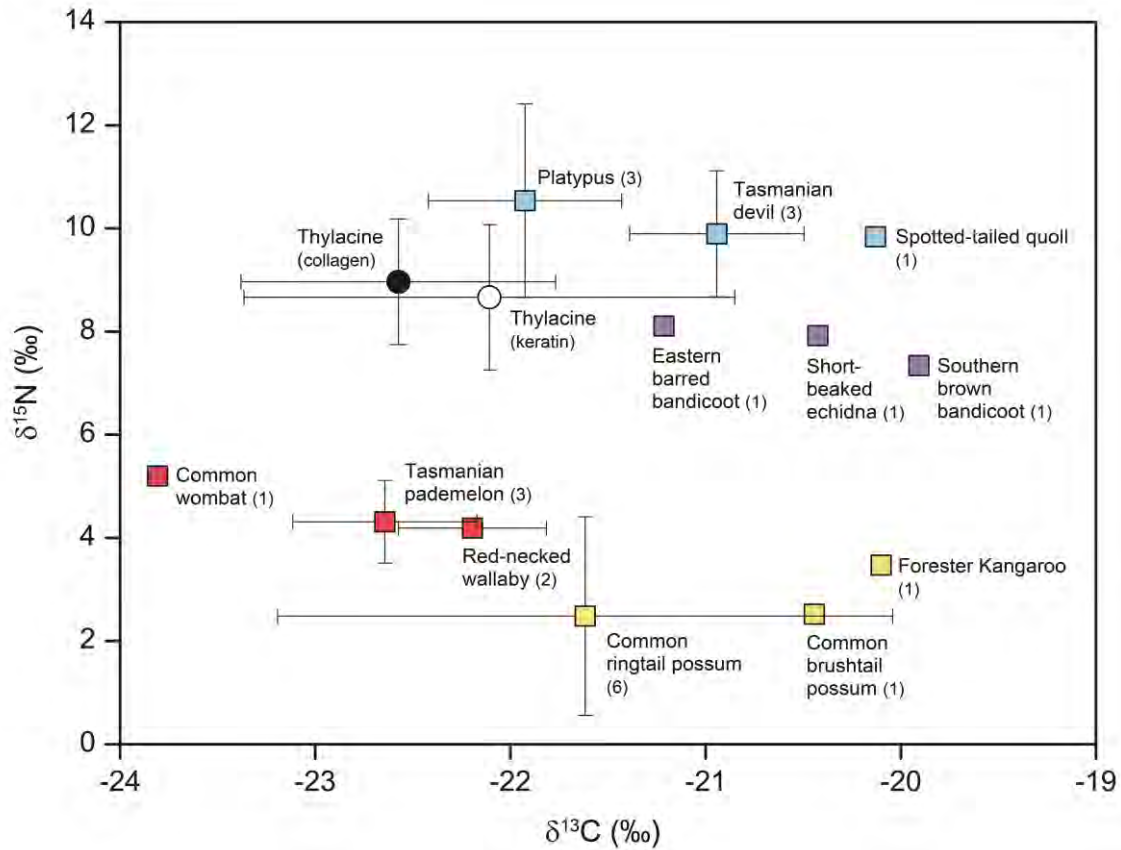


Figure 4.1 Relationship between stable isotope signatures of thylacines (circle symbol; black = bone collagen, white = keratin) and potential prey species (square symbol). Mean ($\pm$ SD) table isotope values are present for each species sampled between 1835 and 1964 (sample size in brackets). The stable isotope values have not been adjusted for trophic enrichment. Refer to Appendix A4 for average C/N ratios of each potential prey species.

4.3.2 Sample acquisition

Bone was sampled from skull and skeletons, and vibrissae were preferentially collected from skins and taxidermies. Where all vibrissae were absent or were too invasive to sample, we instead collected skin and/or hair from the specimen. Bone fragment(s) that had previously fallen from the specimen were preferentially used. If loose material was not present, a small quantity of bone (approximately 0.2 g) was removed from the nasal turbinates using tweezers to minimise external damage to the skull. A single vibrissa was removed from each specimen by plucking it with tweezers, or cutting the vibrissa close to the skin surface. A small sample of skin covering $\sim 0.5 \text{ cm}^2$ surface was cut from underside of the rear right limb of each specimen with a scalpel. As particular preservation methods can alter stable isotope signatures (Bugoni *et al.* 2008), we did not collect any tissues preserved in solution.

4.3.3 Sample preparation

4.3.3.1 Bone

Collagen was extracted from bone using a method modified from Birchall *et al.* (2005) and Longin (1971). Dirt and any adhering soft tissue was removed from the exterior bone surface using a scalpel blade. Bone fragment(s) were weighed (0.2-0.5 g) and placed into pre-weighed 12 ml glass test tubes. The test tubes were covered with aluminium foil and the samples demineralised by one rinse in cold (-2°C) 8 ml 0.5 M HCl at -2°C for at least 2 days. Samples were shaken twice daily, and the solvent was changed every 4 days. Demineralisation was complete once the bone softened and no more bubbles were released. The solvent was decanted using 8 μm EZfilters (Elkay Laboratory Products (UK) Ltd., Basingstoke) and discarded. Remaining traces of the solvent were removed by three rinses in distilled water. The remaining collagen pellet was gelatinised by heating in 8 ml pH 3.0 water at 75°C for 48 h. Plastic lids were placed on each tube and aluminium foil was used to cover the top of the tubes to keep the lids securely in place. After 48 h, all the collagen should have dissolved, leaving behind all acid

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

insoluble material. The supernatant liquor was filtered off into labelled and pre-weighed plastic test tubes using 8 µm EZfilters. The samples were left overnight at -20°C, at a sharp angle to increase surface area of the solution, and then transferred to -80°C for a minimum of two days. The samples were freeze-dried for at least 6 days until dry. The lyophilised material contained collagen. The plastic tubes containing collagen were weighed to estimate the yield of collagen. Between 0.8-1.2 mg collagen was placed into pre-weighed tin capsules.

4.3.3.2 *Hair and vibrissae*

Vibrissa and hair samples were washed in 12 ml glass test tubes by two rinses (30 min each) in distilled water, one rinse (30 min) in a 2:1 (v/v) mixture of methanol:chloroform followed by one rinse (30 min) in a 2:1 (v/v) mixture of chloroform: methanol. Samples were then rinsed twice (30 min) in distilled water to remove any solvent. Samples were ultrasonicated at room temperature for all washes and were left to air dry between washes. The total length and weight of each vibrissa was measured after the final wash. The vibrissa tip was cut using a scalpel (0.3-0.5 mg) and analysed for each specimen. The length of each vibrissa segment was recorded before placing it into pre-weighed tin foil capsules. Hairs were weighed (0.3-0.5 mg per sample; equivalent to 10-20 hairs) and placed into pre-weighed tin foil capsules. Each tin capsule was rolled into a small ball and stored in a 96-well plate for stable isotope analysis.

4.3.3.3 *Skin*

Collagen was extracted from each skin sample using modified protocols from Finucane (2007). The skin samples were cleaned physically by plucking and scraping hair from the skin surface. Samples were then defatted by three rinses (1 h each) in a 2:1 (v/v) mixture of chloroform-methanol in an ultrasonic bath at room temperature. All traces of the solvent were removed from the soft tissue by two rinses (30 min each) in distilled ionised water in an ultrasonic bath at room temperature. Skin samples were dissolved by

placing them in ~8 ml of pH 3.0 water at 75°C for 48 hours. The dissolved protein was then filtered using 8 µm EZfilters, and the insoluble residue discarded. The gelatinised protein was frozen overnight at -20°C, then was moved to -80°C for a minimum of two days. The samples were freeze dried for 6 days to remove all moisture. Lyophilised protein (0.8-1.2 mg) from skin tissue was measured into pre-weighed tin capsules. Each tin capsule was rolled into a small ball and stored in a 96-well plate for stable isotope analysis.

4.3.4 Sample analysis

Tissue $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were determined by flash combustion to produce CO_2 and N_2 and measured in a Finnegan Mat 252 isotope-ratio mass spectrometer coupled with a Finnegan control interface and Europa preparation element analyser (Environmental Isotopes Inc., North Ryde, Sydney). Stable isotope abundances were measured as the ratio between the heavy to light isotope in the sample compared to the same ratio in an international standard. Isotope ratios were expressed as delta (δ) values in parts per thousand (‰ or per mil) relative to Pee Dee Belemnite (C) and AIR (N), respectively. We also obtained the ratio between carbon and nitrogen, which was used as a measure of sample quality. Median values were used in analysis for each specimen when more than one sample was analysed from the same tissue type. The $\delta^{15}\text{N}$ value of thylacine specimen GMN782 (29.5‰) and the $\delta^{13}\text{C}$ values of eastern quoll specimens A16 (1.7‰) were unusually high due to possible contamination. These values were excluded from the study.

4.3.5 Controlling animal $\delta^{13}\text{C}$ values for the Suess effect

Depletion in the $\delta^{13}\text{C}$ of atmospheric carbon dioxide (CO_2) over the past century due to fossil fuel combustion can influence the $\delta^{13}\text{C}$ values of our specimens as they were collected over a long period (Lee *et al.* 2005; Long *et al.* 2005; Roth *et al.* 2007). To control for this source of variation, known as the Suess effect (Suess 1955), we subtracted the difference in atmospheric concentrations of $\delta^{13}\text{C}$ in the year the specimen

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

died from the raw $\delta^{13}\text{C}$ value measured in the specimen. The atmospheric concentrations of $\delta^{13}\text{C}$ was approximated by $\delta^{13}\text{C}_{\text{atm}} = k - eat^2$, where k and a were estimated parameters and t was an index to year in which 1 represented 1880 (Long *et al.* 2005). We used a k value of -5.5656 and a value of 6.0932×10^{-5} , obtained from Long *et al.* (2005). Thylacine specimens with an unknown year of death or acquisition were adjusted for $\delta^{13}\text{C}_{\text{atm}}$ levels from the year 1895, which is estimated to be the peak year that thylacines were killed as part of the government bounty (Bulte *et al.* 2003).

4.3.6 Inter-tissue isotopic spacing

A pilot study on Tasmanian devil tissues were used to correct for potential enrichment in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between bone collagen, skin collagen, hair and vibrissae (see Appendix B for methodology). The average enrichment in $\delta^{13}\text{C}$ between bone collagen and vibrissa was found to be significantly different (Wilcoxon matched pairs signed rank test; $P=0.01$, Appendix B3). For analysis involving pooled tissue types, we applied an average enrichment value of 1.69‰ to all vibrissa samples to more closely reflect the stable isotope signature of bone collagen. No other differences were detected between average enrichment between bone collagen and other tissues, thereby allowing these tissues to be used interchangeably in this study without applying a correction factor.

4.3.7 Niche overlap and sex-biased variation

The data did not fit the assumptions of a one-way ANOVA, so non-parametric methods were used for species and sex-based comparisons. A bivariate plot was used to qualitatively describe the spread of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for each marsupial carnivore dating between 1803 and 1936 (thylacine $n=34$, Tasmanian devil $n=12$, spotted-tailed quoll $n=10$ and eastern quoll $n=8$), regardless of sex. A Kruskal-Wallis test and Mann-Whitney U -test was used to assess differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between species and test for niche overlap. Results were judged significant when $P<0.05$.

The Mann-Whitney U -test was used to test for differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between sexes for the thylacine (female $n=5$; male $n=8$), Tasmanian devil (female $n=24$; male $n=19$), spotted-tailed quoll (female $n=7$; male $n=29$) and eastern quoll (female $n=10$; male $n=20$), regardless of their year of collection. Intersexual differences were evaluated for collagen and keratin tissues separately where there was a sufficient sample size in addition to pooled tissues after accounting for inter-tissue isotopic spacing (section 4.3.6). Data was analysed in SPSS (version 20 SPSS Inc., Chicago, USA).

4.3.8 Diet reconstruction and prey size

There were very few large (average body mass >30 kg) native mammals in Tasmania for thylacines to potentially hunt; the forester kangaroo (*Macropus giganteus*; female, 37 kg; male, 66 kg; Strahan 1995) and Tasmanian emu (*Dromaius novaehollandiae diemenensis*; ~ 40 kg; Heupink *et al.* 2011). Tasmanian devils (mean weight females 6.1 kg, males 8.7 kg; Jones 1995) primarily consume medium-sized mammals (Jones 1997), whereas spotted-tailed quolls (mean weight female 1.8 kg, males 3.5 kg; Jones 1995) consume mostly small- to medium-sized prey (Glen and Dickman 2006). Eastern quolls (mean weight males 1.1 kg, females 0.7 kg; Jones and Barmuta 1998) commonly hunt prey less than 1 kg in body mass (Jones 1997). Understanding prey size patterns in the thylacine can be used to identify trophic interactions in this historical food web.

To determine the relative contributions of potential prey types to the diet of the sampled thylacine population, we used the Bayesian stable isotope mixing model, MixSIR. MixSIR accounts for uncertainty associated with diet-tissue fractionation and stable isotope values of the consumer (mixture) and its food (sources) by applying standard deviations to predator, prey and diet-tissue fractionation data (Moore and Semmens 2008). The contribution of each source to the mixture can thus be accurately obtained in the form of probability distributions while accounting for uncertainty in stable isotope values for prey groups represented by a small sample size.

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

Bone collagen $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of 33 thylacines were included in MixSIR. As stable isotope mixing models require that the isotopic values of all prey categories are significantly different from each other, prey species with similar $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were combined to form four isotopically distinguishable prey groups. Each group belonged to a specific feeding niche. These were (1) Carnivores (Tasmanian devil, spotted-tailed quoll and platypus (*Ornithorhynchus anatinus*)), (2) Insectivores (eastern barred bandicoot (*Perameles gunnii*), southern brown bandicoot (*Isodon obesulus*) and short-beaked echidna (*Tachyglossus aculeatus*)), (3) Herbivores A (common wombat (*Vombatus ursinus*), Tasmanian pademelon (*Thylogale billardierii*) and red-necked wallaby (*Macropus rufogriseus*)) and (4) Herbivores B (common brushtail possum (*Trichosurus vulpecula*), common ringtail possum (*Pseudocheirus peregrinus*) and forester kangaroo). Both herbivorous groups contained grazers and browsers (Appendix A3) and were unable to distinguished based on food preferences. MixSIR assumes that each individual predator consumes all possible sources incorporated in the model. For each prey group, we applied a mean and standard deviation for $\delta^{13}\text{C}$ (Carnivores, mean=-21.5 $\pm$ 0.7; Insectivores, mean=-20.9 $\pm$ 0.7; Herbivores A, mean=-22.7 $\pm$ 0.7 and Herbivores B, mean=-21.3 $\pm$ 1.5) and $\delta^{15}\text{N}$ (Carnivores, mean=10.2 $\pm$ 1.3; Insectivores, mean=9.4 $\pm$ 1.8; Herbivores A, mean=4.4 $\pm$ 0.6 and Herbivores B, mean=2.5 $\pm$ 1.7).

Potential prey species were divided into three size classes based on their average body mass: small (<12kg), medium (13-30 kg) and large (>30 kg; exceeding thylacine's average body mass; Paddle 2000). A literature search was used to determine the body mass average and range for each prey species, and is listed in Appendix A3. The group Carnivores and Insectivores consisted only of small-sized species and Herbivores A primarily consisted of medium-sized species. Herbivores B is primarily composed of samples from small prey species (common brushtail possum and common ringtail possum), and has one large prey species, the Forester kangaroo represented by a single sample. Prey species referred to in the literature that were unaccounted for in this study due to unavailable material include two smaller species of macropodids ((long-nosed potoroo (*Potorous tridactylus*) and Tasmanian bettong (*Bettongia gaimardi*)), water

birds, rodents (e.g. water rat (*Hydromys chrysogaster*), black rat (*Rattus rattus*) and brown rat (*Rattus norvegicus*)) and sheep (*Ovis aries*) (Paddle 2000).

4.3.8.1 Diet-tissue fractionation

Diet-tissue fractionation values have been studied in one marsupial species, the long-nosed bandicoot (*Parameles nasuta*) using plasma and blood. Fractionation values of both tissues sampled from the long-nosed bandicoot were compared with fractionation values reported in the same tissue of placental mammals to assess similarities (Appendix C1 and C2). As $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ fractionation values of the long-nosed bandicoot fell within the range of other mammal species, it is reasonable to use fractionation values of placental carnivores to estimate trophic fractionation in marsupial species.

We estimated $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ fractionation between diet and bone for terrestrial carnivores based on previous studies involving individual laboratory trials or field observations on single species and whole ecosystems (Appendix C3). The mean $\Delta\delta^{13}\text{C}$ and $\Delta\delta^{15}\text{N}$ bone collagen-diet for terrestrial carnivores was 1.13‰ and 3.89‰, respectively. These values were higher than average enrichment values generally accepted for mammals: 1.00‰ and 3.40‰ for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, respectively (DeNiro and Epstein 1981; Minagawa and Wada 1984; Peterson and Fry 1987; France and Peters 1997). Tissue and trophic level specific enrichment values are more appropriate to use than these 'rule of thumb' values, where available (Eggers and Hefin 2000). Bocherens *et al.* (2003) noted that most studies that investigate bone collagen to diet fractionation values of terrestrial predators include only one species as the main prey, and thus may not reflect the actual enrichment value, which would include contributions of other possible prey. The standard deviation for $\Delta\delta^{13}\text{C}$ and $\Delta\delta^{15}\text{N}$ bone collagen-diet for terrestrial carnivores obtained from the literature was averaged (0.60‰ for $\delta^{13}\text{C}$ and 0.80‰ for $\delta^{15}\text{N}$, Appendix C3) and incorporated into MixSIR. By fully accounting for uncertainty in fractionation values, we were able to obtain accurate estimates of source contributions. The model was run for 10,000,000 iterations.

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

4.3.9 Long-term trends in marsupial carnivore stable isotope values

All marsupial carnivore specimens with a known year of collection were included to examine temporal trends in $\delta^{13}\text{C}$ or $\delta^{15}\text{N}$ following European settlement in Tasmania. Spearman correlation coefficient was used to examine temporal trends in $\delta^{13}\text{C}$ or $\delta^{15}\text{N}$ values for thylacines (1852-1930), Tasmanian devil (1921-2012), spotted-tailed quoll (1914-2008) and eastern quoll (1921-2012). The Spearman correlation coefficient is a non-parametric test that measures whether a monotonic relationship exists between two variables, whereby (1) as the value of one variable increases, so does the value of the other variable; or (2) as the value of one variable increases, the other variable value decreases.

To more accurately determine when any change in stable isotope values occurred for a given species, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were divided into four categories based on the year of acquisition; 1900-1959, 1960-1979, 1980-1999, 2000-2012. These periods were chosen somewhat arbitrarily but represent convenient 20-year periods for comparative purposes, with the exception of 1900-1959 and 2000-2012. As few specimens were available, category 1900-1959 includes all specimens collected from this longer time frame. A General Linear Model with a post-hoc Tukey test was used to test for differences between $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between the four time categories for all extant species.

Stable isotope values of the thylacine were compared between specimens collected before and after the initiation of the government bounty scheme in 1888 (<1888 ; $n=16$ and ≥ 1888 ; $n=13$, respectively). The Shapiro-Wilk test was used to test for data departure from normality and is appropriate to use for small sample sizes. As the data was not distributed normally, and was not able to be normalised by transformation methods, this data was analysed using the Mann-Whitney U -test. Temporal data analysis was performed using SPSS (version 20 SPSS Inc., Chicago, USA).

4.4 RESULTS

The thylacine population has a large range of $\delta^{15}\text{N}$ (4.3‰) and $\delta^{13}\text{C}$ (4.0‰) values. A significant difference in $\delta^{13}\text{C}$ (Kruskal-Wallis, $X^2=23.51$, d.f.=3, $P<0.001$) and non-significant difference in $\delta^{15}\text{N}$ values (Kruskal-Wallis, $X^2=17.18$, d.f.=3, $P<0.01$) was found between marsupial carnivores collected in 1936 or earlier. A Mann-Whitney U -test was used to identify which species varied in their median stable isotope values for pooled tissues. The $\delta^{13}\text{C}$ values of the thylacine were significantly (Mann-Whitney U -test, $Z=-3.81$, $P<0.001$, $n=34,12$ for the Tasmanian devil; $Z=-3.16$, $P<0.01$, $n=34,10$ for the spotted-tailed quoll; $Z=-3.05$, $P<0.01$, $n=34,8$ for the eastern quoll) lower (Figure 4.2) than all marsupial carnivores. The thylacine $\delta^{15}\text{N}$ values was significantly (Mann-Whitney U -test, $Z=-3.08$, $P<0.01$, $n=34, 12$) lower (Figure 4.2) than the Tasmanian devil. No other significant differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were detected between species. There were no significant intersexual differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for keratin, collagen or pooled tissues for all species (Table 4.1), though this result should be taken with caution due to the small sample size available for this analysis.

The bivariate plot of isotopic signatures shows the mixing space of potential prey groups (Figure 4.3), within which proximity to the thylacine indicates the greatest contribution to diet (Moore and Semmens 2008). Enrichment values (1.13‰ and 3.89‰ for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, respectively) were added to all prey groups in the plot. The average stable isotope value of thylacines was shifted to the left of the prey mixing space, represented by the polygon. Thylacine stable isotope values were closest to medium-sized herbivores (Herbivores A), which included the common wombat, Tasmanian pademelon and red-necked wallaby. Estimates for the group Herbivores A ranged from 81 to 100% of total diet for the thylacine population. Estimates for the group Carnivores, Insectivores and Herbivores B were 0 to 17%, 0 to 16% and 0 to 8% of total diet for the thylacine, respectively. The $\delta^{13}\text{C}$ standard deviation of the thylacine population falls slightly outside of the prey mixing space. This may be caused by prey

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

species unaccounted for in the model or the variance of prey group stable isotope values may be greater than that estimated. MixSIR had 988 posterior draws, with no duplicate draws in the posterior chain. The ratio between the posterior at the best draw and the total posterior density was less than 0.01.

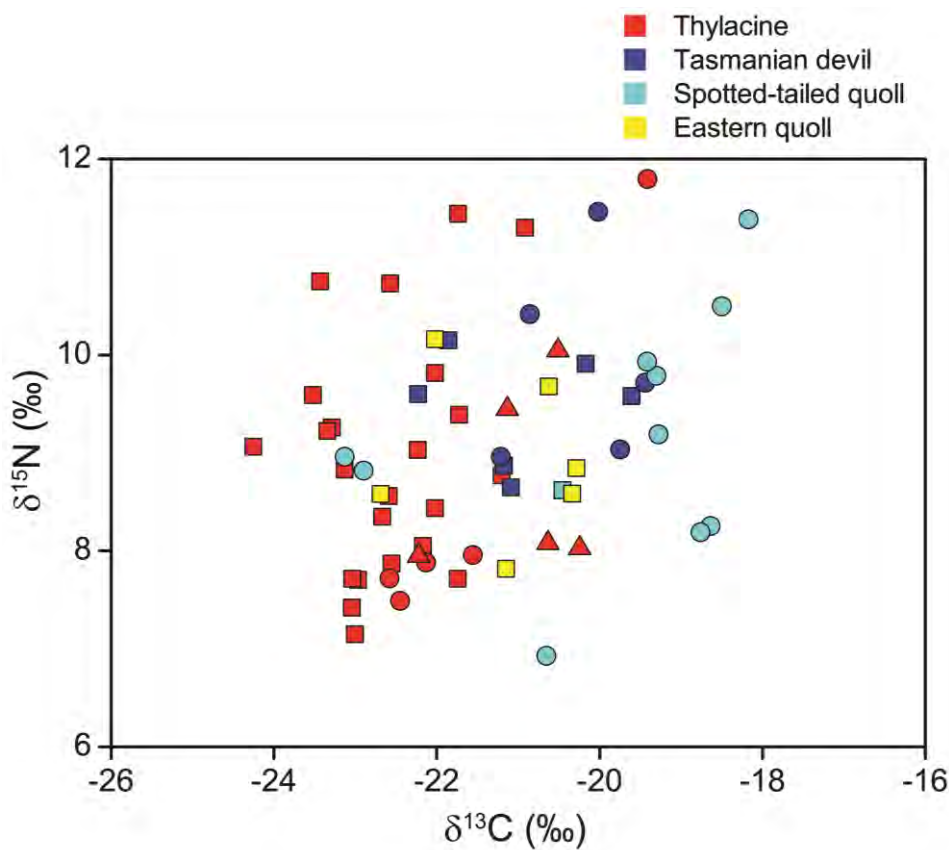


Figure 4.2 Bivariate plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of the thylacine ($n=34$; red), Tasmanian devil ($n=12$; dark blue), spotted-tailed quoll ($n=10$; light blue) and eastern quoll ($n=8$; yellow) dating from 1803 to 1936 in Tasmania. Symbols represent tissue analysed; bone collagen (square), hair (circle) and vibrissa (triangle) samples.

Table 4.1 Mann-Whitney *U*-test for comparisons in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between sexes (M = male, F = female) for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll. All specimens were wild adults collected from Tasmania at any time following European settlement. Mean ($\pm$ SD) $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for each sex are provided. Comparisons between sexes were completed for bone collagen, keratin (hair and vibrissae) and both tissue types combined (corrected for inter-tissue differences between vibrissae and bone collagen) for species where there was a sufficient sample size.

Species	Sex	$\delta^{13}\text{C}$ (‰)				$\delta^{15}\text{N}$ (‰)			
		Mean $\pm$ SD	Z	P-value	n	Mean $\pm$ SD	Z	P-value	n
Bone collagen									
Tasmanian devil	M	-22.0 $\pm$ 0.7	-1.29	-0.20	14	9.6 $\pm$ 1.7	-1.37	0.17	14
	F	-21.4 $\pm$ 1.1			12	9.0 $\pm$ 1.8			12
Keratin									
Eastern quoll	M	-21.3 $\pm$ 1.0	-1.15	0.25	12	9.6 $\pm$ 1.5	-0.93	0.35	12
	F	-20.3 $\pm$ 1.8			7	8.8 $\pm$ 1.6			7
Keratin and bone collagen									
Thylacine	M	-21.8 $\pm$ 1.2	-0.22	0.83	8	9.2 $\pm$ 1.6	-1.10	0.27	8
	F	-22.2 $\pm$ 0.3			5	10.8 $\pm$ 2.8			5
Tasmanian devil	M	-21.9 $\pm$ 0.9	-0.57	0.57	19	10.1 $\pm$ 2.1	-1.09	0.28	19
	F	-22.1 $\pm$ 2.2			24	9.6 $\pm$ 2.1			24
Spotted-tailed quoll	M	-21.0 $\pm$ 1.4	-0.34	0.73	29	9.2 $\pm$ 2.4	-0.42	0.67	29
	F	-21.1 $\pm$ 1.8			7	9.7 $\pm$ 1.9			7
Eastern quoll	M	-21.4 $\pm$ 0.9	-1.66	0.09	20	8.6 $\pm$ 2.1	-0.46	0.64	20
	F	-20.3 $\pm$ 1.7			10	9.0 $\pm$ 1.4			10

*Denotes significant trend (where *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$).

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

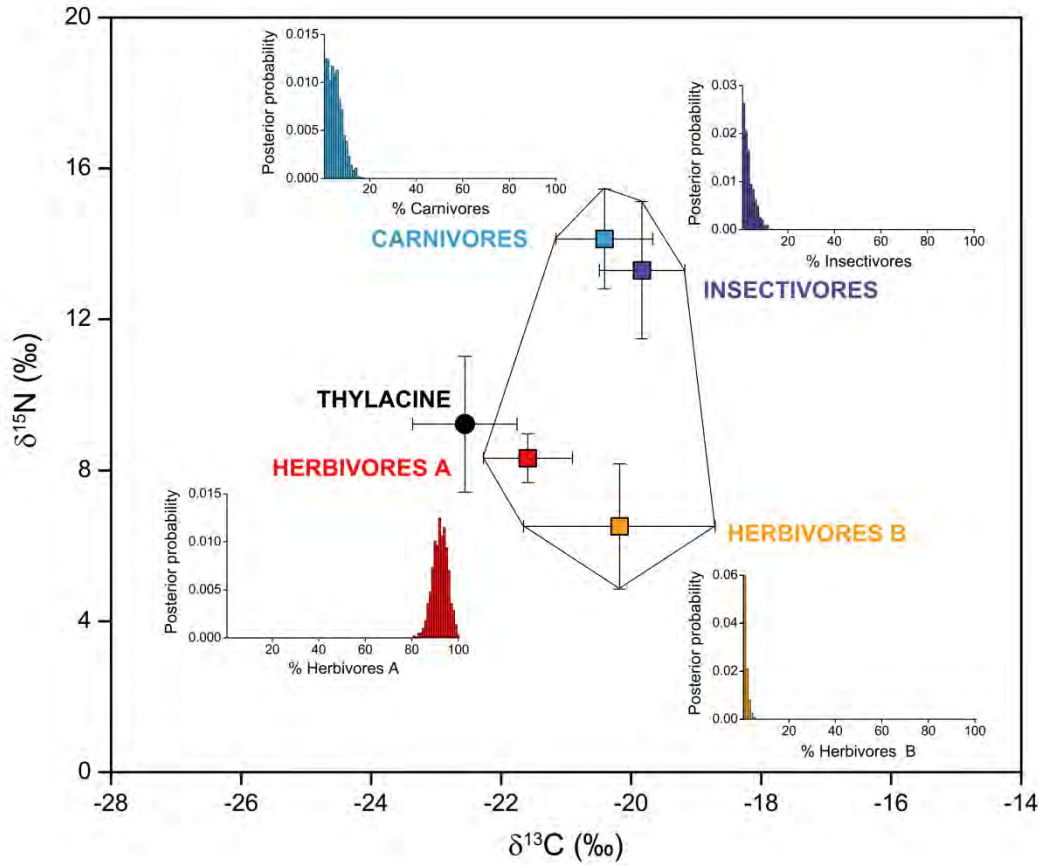


Figure 4.3 Bivariate plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ($\pm\text{SD}$) of thylacine bone collagen ($n=33$; black circle symbol) and skin collagen of potential prey species (square symbol) grouped into four isotopically distinguished categories; Carnivores (blue), Insectivores (purple), Herbivores A (red square) and Herbivores B (yellow square). The polygon describes the mixing space of potential prey groups. Prey stable isotope values have been enriched by $1.13 \pm 0.60\text{‰}$ for $\delta^{13}\text{C}$ and $3.89 \pm 0.80\text{‰}$ for $\delta^{15}\text{N}$. The proximity of the mixture (thylacine) to the source (prey groups) indicates the greatest contribution to the diet. Posterior contributions are measured for each prey group from the Bayesian mixing model, MixSIR.

The $\delta^{13}\text{C}$ values for collagen and keratin samples from these four marsupial carnivores, for which the year of collection was known, were compared with the $\delta^{13}\text{C}$ values of the same samples corrected with an atmospheric model (Figure 4.4). The thylacine showed no monotonic trend in stable isotope ratios from 1852 to 1930 before and after correcting for the Suess effect (Table 4.2 and Table 4.3), nor was there a significant difference in thylacine stable isotope values of pooled tissue types before and after the initiation of the government bounty in 1888 (Mann-Whitney *U*-test; $Z = -0.23$, $P=0.82$, $n=15,13$ for $\delta^{13}\text{C}$ and $Z = -1.32$, $P=0.19$, $n=15,13$ for $\delta^{15}\text{N}$).

Uncorrected and corrected keratin $\delta^{13}\text{C}$ values were negatively correlated with collection year for the Tasmanian devil (Spearman's correlation coefficient; uncorrected, $n=23$, $q = -0.56$, $P<0.01$; corrected, $n=23$, $q = -0.41$, $P=0.05$; Table 4.2) and spotted-tailed quoll (Spearman's correlation coefficient; uncorrected, $n=40$, $q = -0.55$, $P<0.01$; corrected, $n=40$, $q = -0.34$, $P=0.03$; Table 4.2). Uncorrected collagen $\delta^{13}\text{C}$ values were negatively correlated with collection year for the Tasmanian devil (Spearman's correlation coefficient; $n=38$, $q = -0.38$, $P=0.02$; Table 4.2), yet were not significantly correlated for uncorrected collagen $\delta^{13}\text{C}$ values (Spearman's correlation coefficient; $n=38$, $q = 0.12$, $P=0.47$; Table 4.2). Uncorrected keratin $\delta^{13}\text{C}$ values in the eastern quoll revealed a significant negative correlation with the collection year (Spearman's correlation coefficient; $n=34$, $q = -0.46$, $P<0.01$). However, no significant correlation was evident in the eastern quoll keratin $\delta^{13}\text{C}$ values after applying the atmospheric model correction (Spearman's correlation coefficient; $n=34$, $q = -0.12$, $P=0.51$). Both corrected and uncorrected collagen $\delta^{13}\text{C}$ values showed no significant correlation with collection year for both quoll species (see Table 4.2).

Keratin $\delta^{15}\text{N}$ values were positively correlated with collection year for the Tasmanian devil (Spearman's correlation coefficient; $n=23$, $q = 0.45$, $P=0.03$; Table 4.3). No significant correlation was evident for collagen or keratin $\delta^{15}\text{N}$ in other species (Table 4.3).

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

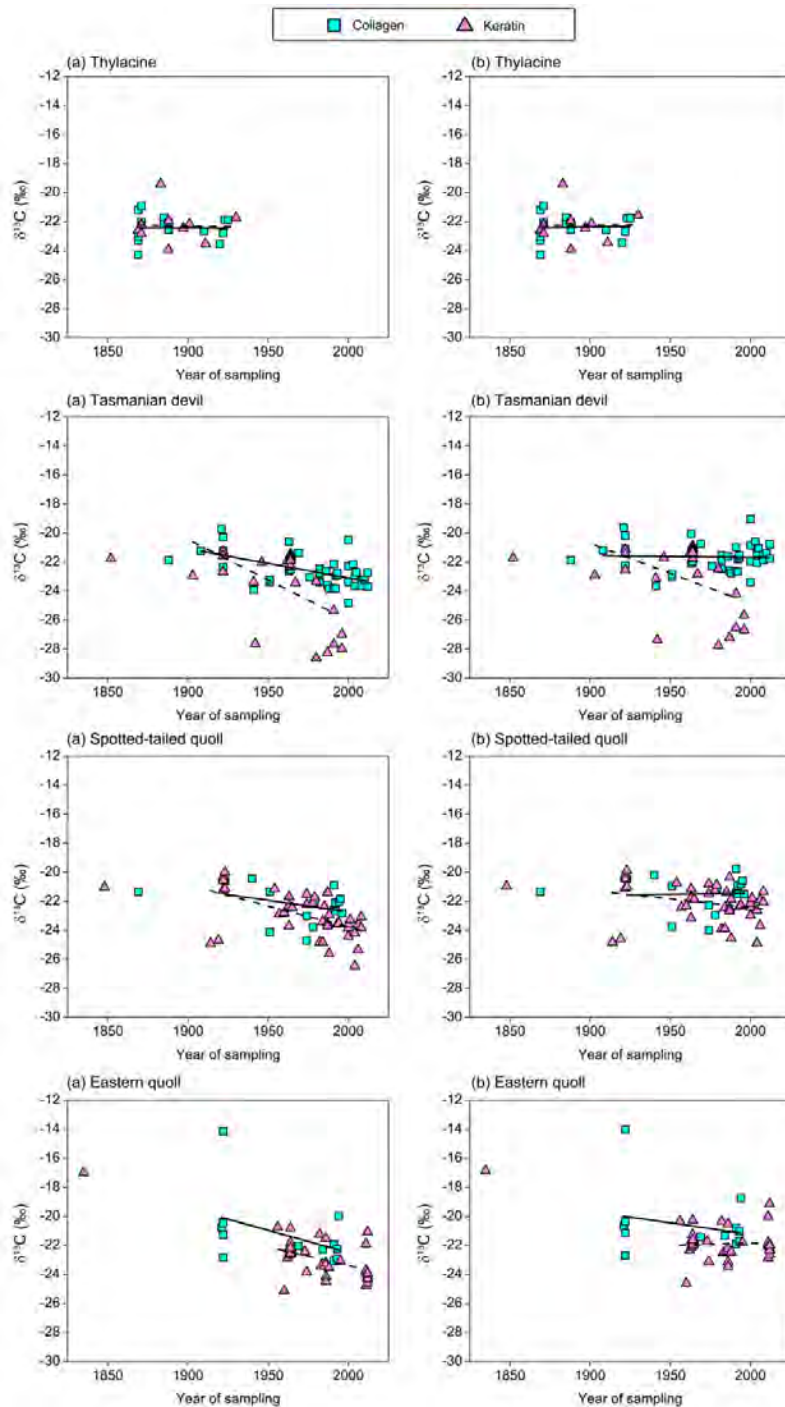


Figure 4.4 Comparison of the $\delta^{13}\text{C}$ values for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll from 1830 to 2012, Tasmania. Values were corrected with an atmospheric model calculation (Long *et al.* 2005). Blue squares with no dashed trendline and pink triangles with dashed trendline indicate values for collagen (bone) and keratin (hair and vibrissa) from specimens, respectively.

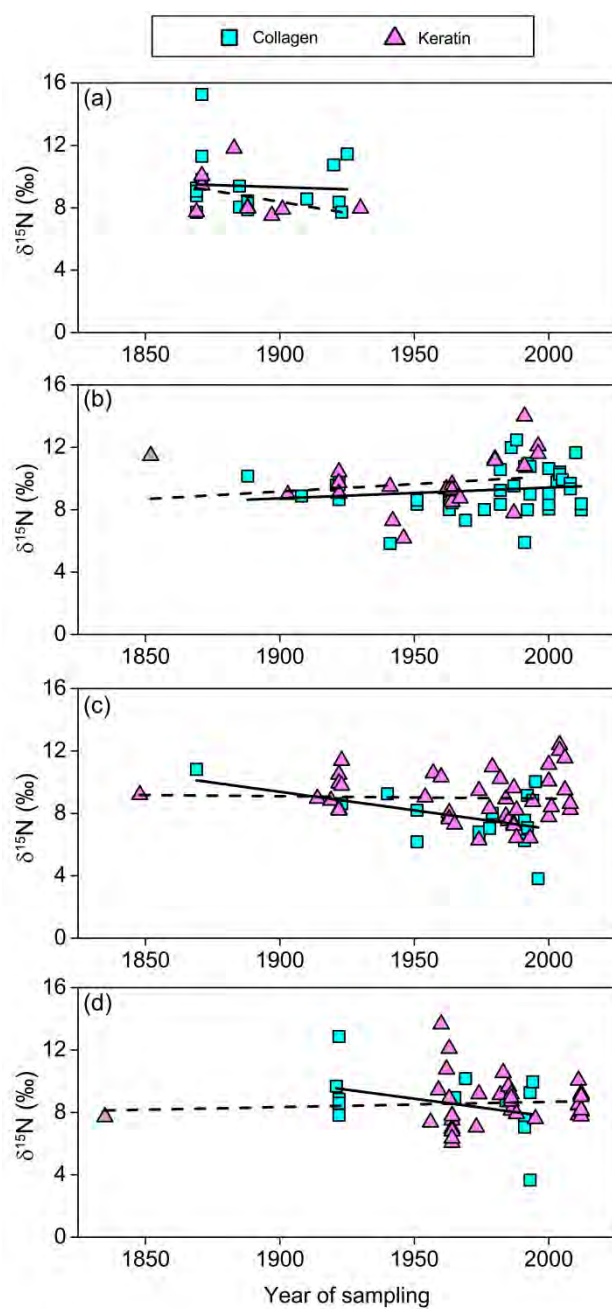


Figure 4.5 Comparison of the $\delta^{15}\text{N}$ values for the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll from 1830 to 2012, Tasmania. Circles and triangles indicate values for collagen (bone) and keratin (hair and vibrissa) from specimens, respectively.

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

Table 4.2 Spearman correlation coefficient for $\delta^{13}\text{C}$ values for thylacines (1852-1930), Tasmanian devil (1921-2012), spotted-tailed quoll (1914-2008) and eastern quoll (1921-2012) in Tasmania that were uncorrected and uncorrected with an atmospheric model (Long et al. 2005).

Species	Collagen $\delta^{13}\text{C}$ (‰)			Keratin $\delta^{13}\text{C}$ (‰)		
	<i>n</i>	<i>q</i>	<i>P</i>	<i>n</i>	<i>q</i>	<i>P</i>
Uncorrected						
Thylacine	17	0.22	0.40	10	0.12	0.73
Tasmanian devil	38	-0.38	0.02*	23	-0.56	<0.01**
Spotted-tailed quoll	15	-0.14	0.62	40	-0.55	<0.001***
Eastern quoll	14	-0.34	0.23	34	-0.46	<0.01**
Corrected						
Thylacine	17	0.24	0.36	10	0.15	0.67
Tasmanian devil	38	0.12	0.47	23	-0.41	0.05*
Spotted-tailed quoll	15	0.06	0.83	40	-0.34	0.03*
Eastern quoll	14	-0.15	0.61	34	-0.12	0.51

n is the number of samples and *q* is the normal variate.

*Denotes significant trend (where *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$).

Table 4.3 Spearman correlation coefficient for $\delta^{15}\text{N}$ for the thylacine (1852-1930), Tasmanian devil (1921-2012), spotted-tailed quoll (1914-2008) and eastern quoll (1921-2012) in Tasmania.

Species	Collagen $\delta^{15}\text{N}$ (‰)			Keratin $\delta^{15}\text{N}$ (‰)		
	<i>n</i>	<i>q</i>	<i>P</i>	<i>n</i>	<i>q</i>	<i>P</i>
Thylacine	17	-0.01	0.97	10	-0.26	0.5
Tasmanian devil	38	0.17	0.31	23	0.45	0.03*
Spotted-tailed quoll	15	-0.03	0.92	40	-0.01	0.98
Eastern quoll	14	-0.32	0.27	34	0.04	0.85

n is the number of samples and *q* is the normal variate.

*Denotes significant trend (where *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$).

For each extant species, $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were divided into four categories based on the year of collection to identify temporal isotopic shifts over a shorter time frame. The Tasmanian devil showed differences between categorised decades for $\delta^{13}\text{C}$ (General Linear Model: $\text{df}=3$, $F=7.167$, $P<0.001$; Table 4.4) and $\delta^{15}\text{N}$ (General Linear Model: $\text{df}=3$, $F=7.330$, $P<0.001$; Table 4.5). Tasmanian devil specimens collected from 1980-1999 had significantly (Table 4.3) lower (Figure 4.5b) $\delta^{13}\text{C}$ values than those collected in 1960-1979 and 2000-2012. Tasmanian devil $\delta^{15}\text{N}$ records from 1980-1999 were significantly ($P<0.01$; Table 4.3) higher (Figure 4.5a) than specimens collected in 1900-1959 and 1960-1979. The spotted-tailed quolls showed differences between categorised decades for $\delta^{15}\text{N}$ (General Linear Model: $\text{df}=3$, $F=10.398$, $P<0.001$; Table 4.5), but not $\delta^{13}\text{C}$ (General Linear Model: $\text{df}=3$, $F=1.405$, $P=0.25$; Table 4.4). Spotted-tailed quoll $\delta^{15}\text{N}$ values from 1980-1999 were significantly (Table 4.5) lower (Figure 4.5d) than specimens collected from 1900-1959 and 2000-2012. Spotted-tailed quoll $\delta^{15}\text{N}$ values from 1960-1979 were also significantly (Table 4.5) lower (Figure 4.5d) than specimens collected from 2000-2012. The eastern quoll showed no significant differences in $\delta^{13}\text{C}$ (General Linear Model: $\text{df}=3$, $F=1.386$, $P=0.26$; Table 4.4) and $\delta^{15}\text{N}$ (General Linear Model: $\text{df}=3$, $F=0.354$, $P=0.79$; Table 4.5) values between the four time periods.

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

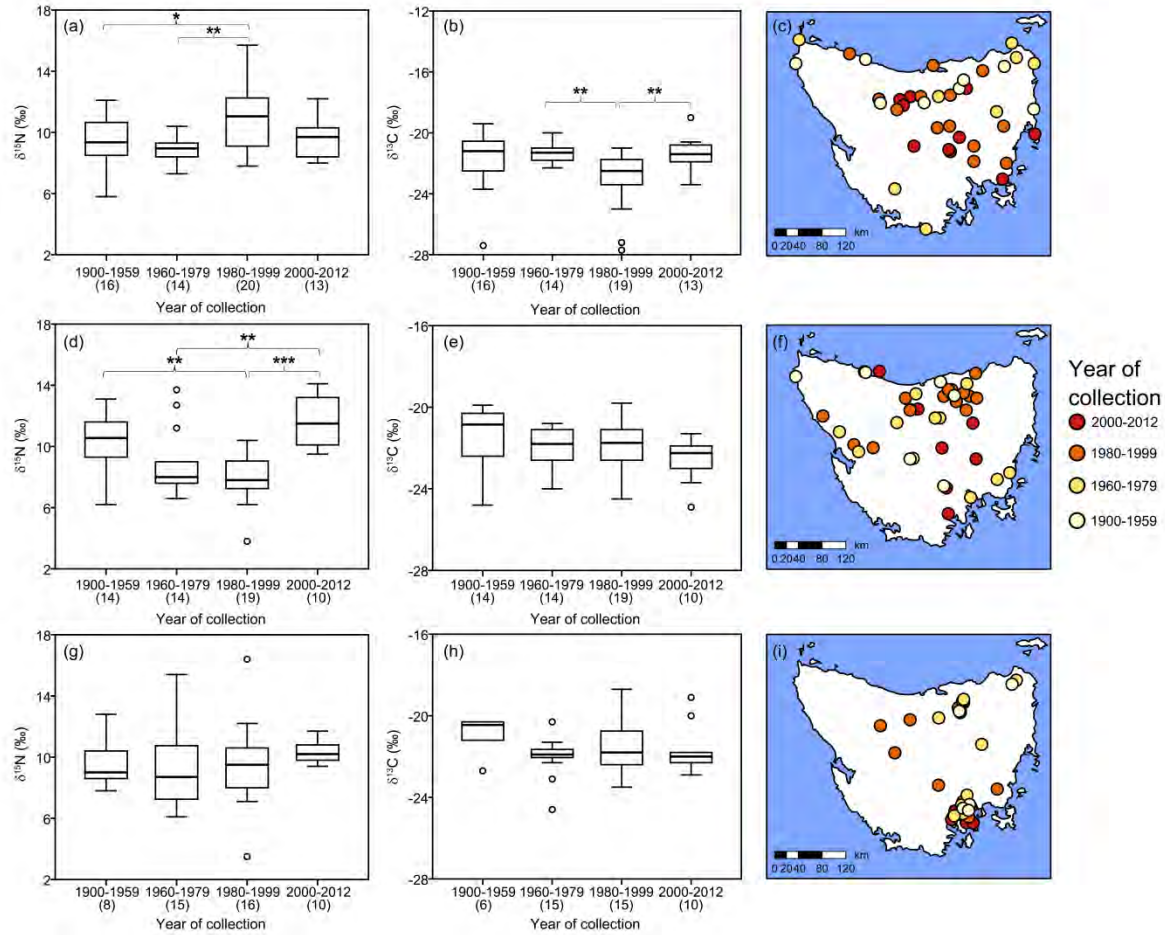


Figure 4.6 Box and whisker plot of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of (a, b) Tasmanian devil, (d, e) spotted-tailed quoll and (g, h) eastern quoll specimens collected between 1930 and 2012 in Tasmania. Specimen $\delta^{13}\text{C}$ values were combined for years dating 1803-1939, 1940-1959, 1980-1999 and 2000-2012. Values were corrected for the Suess effect and inter-tissue differences (see section 4.3.5 and 4.3.6, respectively). Box shows median, lower and upper quartiles, whiskers cover $10^{\text{th}} \pm 90^{\text{th}}$ percentile range, and circles are statistical outliers. Overall specimen sample size is displayed in brackets. For tissue type see Appendix A1. Significant differences between groups are denoted in *, where *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$. Geographic distribution of tissue samples collected of (c) Tasmanian devil, (f) spotted-tailed quoll and (i) eastern quoll based on museum records. Samples are categorised for each species by year of collection. Locality records were unavailable for most historic samples.

Table 4.4 Post-hoc Tukey test comparing $\delta^{13}\text{C}$ values for Tasmanian devil, spotted-tailed quoll and eastern quoll specimens collected in Tasmania during four time periods: 1900-1959, 1960-1979, 1980-1999 and 2000-2012. The $\delta^{13}\text{C}$ values for one Tasmanian devil specimen (QVM:1984:1:0260) and two eastern quoll specimens (65344 and A1516) were identified as extreme outliers based on box and histogram plots and were excluded from the analysis. *P* values and sample sizes are listed for each comparison between grouped decades.

	Year collected	1960-1979	1980-1999	2000-2012
Tasmanian devil	1900-1959	<i>P</i> =0.56, <i>n</i> =16,14	<i>P</i> =0.06, <i>n</i> =16,19	<i>P</i> =0.48, <i>n</i> =16,13
	1960-1979		<i>P</i> <0.01, <i>n</i> =14,19**	<i>P</i> =1.00, <i>n</i> =14,13
	1980-1999			<i>P</i> <0.01, <i>n</i> =19,13**
Spotted-tailed quoll	1900-1959	<i>P</i> =0.75, <i>n</i> =14,14	<i>P</i> =0.74, <i>n</i> =14,19	<i>P</i> =0.18, <i>n</i> =14,10
	1960-1979		<i>P</i> =1.00, <i>n</i> =14,19	<i>P</i> =0.67, <i>n</i> =14,10
	1980-1999			<i>P</i> =0.60, <i>n</i> =19,10
Eastern quoll	1900-1959	<i>P</i> =0.20, <i>n</i> =6,15	<i>P</i> =0.60, <i>n</i> =6,15	<i>P</i> =0.51, <i>n</i> =6,10
	1960-1979		<i>P</i> =0.75, <i>n</i> =15,15	<i>P</i> =0.92, <i>n</i> =15,10
	1980-1999			<i>P</i> =0.99, <i>n</i> =15,10

Note: Significant differences between groups are denoted in *, where ****P* < 0.001; ***P* < 0.01; **P* < 0.05; ns, *P* > 0.05

Table 4.5 Post-hoc Tukey test comparing $\delta^{15}\text{N}$ values of Tasmanian devil, spotted-tailed quoll and eastern quoll specimens collected in Tasmania at four time periods: 1900-1959, 1960-1979, 1980-1999 and 2000-2012. *P* values and sample sizes are listed for each comparison between grouped decades.

	Year collected	1960-1979	1980-1999	2000-2012
Tasmanian devil	1900-1959	<i>P</i> =0.90, <i>n</i> =16,14	<i>P</i> =0.02, <i>n</i> =16,20*	<i>P</i> =0.93, <i>n</i> =16,13
	1960-1979		<i>P</i> <0.01, <i>n</i> =14,20**	<i>P</i> =0.61, <i>n</i> =14,13
	1980-1999			<i>P</i> =0.15, <i>n</i> =20,13
Spotted-tailed quoll	1900-1959	<i>P</i> =1.43, <i>n</i> =14,14	<i>P</i> <0.01, <i>n</i> =14,19**	<i>P</i> =0.31, <i>n</i> =14,10
	1960-1979		<i>P</i> =0.59, <i>n</i> =14,19	<i>P</i> <0.01, <i>n</i> =14,10**
	1980-1999			<i>P</i> <0.001, <i>n</i> =19,10***
Eastern quoll	1900-1959	<i>P</i> =1.00, <i>n</i> =8,15	<i>P</i> =1.00, <i>n</i> =8,15	<i>P</i> =1.12, <i>n</i> =8,10
	1960-1979		<i>P</i> =1.00, <i>n</i> =15,16	<i>P</i> =0.75, <i>n</i> =15,10
	1980-1999			<i>P</i> =0.85, <i>n</i> =16,10

Note: Significant differences between groups are denoted in *, where ****P* < 0.001; ***P* < 0.01; **P* < 0.05; ns, *P* > 0.05

4.5 DISCUSSION

This study suggests that medium-sized (13-30 kg) prey species may have formed the bulk of the thylacine's diet in Tasmania which, if this is the case, would challenge previous notions of their diet and prey size. The dramatically reduced abundance and range of medium-sized mammalian herbivores following European settlement has important implications for the thylacine population that may have heavily relied on these species as a source of food. The extreme difficulty of accessing prey specimens acquired around the time that thylacines were alive meant that some potential prey species were excluded while others were represented by a small sample size so caution must be taken in interpreting this data. A high degree of trophic niche overlap, as identified here between the thylacine and both quoll species in their $\delta^{15}\text{N}$ values, is known to occur when space and resources become limiting factors in the presence of multiple predators (Pimm 2002). This may have led to high competitive pressure among these apex predators unless strategies were used to reduce resource competition. For instance, specialising in different habitats, as suggested by differences in $\delta^{13}\text{C}$ between the thylacine and all other marsupial carnivores, may have reduced direct competition. Surprisingly, intersexual isotopic differences were not detected in the thylacine despite their marked sexual dimorphism, with adult male thylacines on average being 10 kg heavier and up to 10 cm longer than females (Paddle 2000). However, our sample size to test for intersexual differences was limited as most thylacine specimens incorporated in this study did not have a known sex. There has been a long-term shift in stable isotope ratios of extant marsupial carnivores, which is possibly due to anthropogenic impacts such as hunting, agriculture and changing habitat and fire regimes. Our temporal perspective of extant marsupial carnivore isotopic niche, spanning nearly 200 years, has provided invaluable insight into the ecological flexibility of these threatened species and may help improve conservation strategies.

4.5.1 Diet and prey size of the thylacine

Our findings suggest that medium-sized mammalian herbivores may have been the dominant native prey of adult thylacines in Tasmania following European settlement. This may have included red-necked wallabies, Tasmanian pademelons and common wombats. Red-necked wallabies were the most commonly reported prey species of thylacines based on anecdotal accounts, though post-settlement records also suggest thylacine preyed on the smaller macropod species, the Tasmanian pademelon (Paddle 2000). Common wombats share similar habitat preferences to thylacines (i.e. temperate forested areas, sclerophyll forest, coastal scrub and heathland), increasing the likelihood of their co-occurrence (McIlroy 1995; 2008). Post-European hunting of red-necked wallabies and Tasmanian pademelons for meat, skins and sport may have limited their availability (Animal Welfare Advisory Committee Australia 2003). Thylacines may have been prevented from catching larger prey species due to morphological constraints (Jones and Stoddart 1998; Wroe *et al.* 2007; Attard *et al.* 2011) or low availability of larger animals (Pearse and Wapstra 1988; Green 1989). As the availability of medium and large prey species were in decline thylacines may have needed to shift their foraging efforts to smaller, energetically less profitable prey.

Pleistocene cave deposits associated with human hunting in Tasmania reveal a fauna assemblage dominated by red-necked wallabies (75%) and common wombats (25%) (McWilliams *et al.* 1999; Cosgrove and Allen 2001). It is possible that these species were highly abundant before European settlement (Garvey 2010), and as such, may have been preferentially targeted by thylacines. As medium-sized herbivores likely formed a staple diet of adult thylacines, as indicated in our study, the extinction and loss of thylacines from the ecosystem may be linked to the decrease in vigilance reported in macropod species today (Blumstein and Daniel 2003). Reduced anti-predator behaviour of prey species in the absence of a predator is a common occurrence in island communities like Tasmania, as they usually support fewer top predators than the adjacent mainland and are thus more directly affected by the removal of a predator (Blumstein 2002; Blumstein and Daniel 2005).

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

Small- to medium-sized *Macropus* were the main taxa associated with thylacine fossils recovered from caves in South Australia and Victoria (Case 1985). Common wombats occurred in 30% of caves where mainland thylacine fossils were present. We speculate that thylacines from mainland Australia may have shared similar prey preferences as that reported here for Tasmanian thylacines. Bettongs and potoroos also frequently occurred in association with mainland thylacine deposits but unfortunately were unable to be incorporated in the stable isotope mixing model as we did not have specimens from this time period. It is important to note that associations between predator and prey deposits cannot definitely determine maximum prey size as large prey may be too heavy to take back to the den and are instead eaten on site once killed.

Our stable isotope data suggest that the largest Tasmanian marsupial, the forester kangaroo, may not have formed a major prey item of thylacines following European settlement; however, our limited number of forester kangaroo samples makes this speculative. Forester kangaroos were viewed by early settlers as a main prey item (Paddle 2000). The stomach contents of a dissected male thylacine were described as "filled with a quantity of kangaroo weighing 5 lbs" (Paterson 1805). Whether the kangaroo was actively hunted or found as carrion by this thylacine is unknown. Additionally, this singular account does not necessarily mean that forester kangaroos formed a staple food item for the species.

The other large and potentially significant prey of the thylacine was the Tasmanian emu. This endemic emu subspecies became locally extinct in some areas by the 1830's (Dove 1924) and was severally depleted by the 1840's (Le Souef 1904). The species is believed to have survived in the wild until 1865 (Green 1989). Pleistocene remains of the Tasmanian emu indicate that they were similar in size to the modern emu (Heupink *et al.* 2011). The extinction of this species predates most of the thylacine specimens collected for this study though even if they had survived emus may have been too large for thylacines to procure.

Small prey species included in this study (i.e. two herbivore species and all carnivores and insectivores) formed a negligible contribution to the diet of thylacines. A short-beaked echidna was identified in the stomach contents of one thylacine in the early 1800's (Harris 1808); though our isotopic results show that short-beaked echidnas did not form a significant portion of the thylacine's diet. There were five species of possum in Tasmania, with two possum species represented in this study; the common brushtail possum and common ringtail possum. Possums were the main prey type to represent the group Herbivores B in our mixing model, and were shown to form a minor component of the thylacine's diet. The common brushtail possum was thought to form part of the thylacine's diet, potentially because they frequently forage on the ground (Kerle 2001), whereas the highly agile and arboreal common ringtail possum (Cronin 1991) would have been more difficult to catch.

Many assumptions of the thylacine's diet during early settlement are based on the behaviour of captive animals (Paddle 2000). In the 19th century, a captive thylacine reportedly refused to eat wombats despite the high abundance of wombats in the region where the thylacine was caught. In the late 1920's a different captive thylacine refused to eat wallaby carcasses. These singular accounts may not reflect typical behaviour of wild individuals, however potentially illustrates variability in individual dietary preferences.

4.5.2 Niche overlap among sympatric marsupial carnivores

The wide isotopic width of thylacines indicates that they were feeding across an entire trophic level and likely showed variation in habitat use patterns between individuals. The $\delta^{15}\text{N}$ values of the Tasmanian devil showed that this species fed at a slightly higher trophic level compared to the thylacine and also spanned over a wider trophic space. We also report a large trophic overlap between the thylacine and both quoll species. The wide isotopic niche shows that a range of different kinds of prey were taken by each carnivore and may reflect their opportunistic foraging behaviour (Guiler 1970), though

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

an overlap in $\delta^{15}\text{N}$ values may not necessarily mean that the same species were taken by each. Tasmanian devils hunt or scavenge a wide variety of prey including the common wombat, red-necked wallaby, birds, sheep and lamb (Green 1967; Guiler 1970; Pemberton 2008). As the only specialised scavenger in Tasmania (Jones 2003), the Tasmanian devil may have made use of the carcass remains of prey killed by the thylacine, including bone and teeth unlikely to be consumed by thylacines. Both quoll species are opportunistic hunters and will occasionally scavenge for food (Fleay 1932; Sharland 1963; Mansergh 1995; Jones and Barmuta 1998). Mammalian prey forms the bulk of the spotted-tailed quoll diet although they also prey on other taxa including birds, reptiles and invertebrates (Settle 1978; Belcher 1995). The diet of eastern quolls includes small- to medium-sized mammals, birds, skinks, invertebrates and plant material (Jones and Barmuta 1998).

Divergence of resource-exploiting traits (e.g. canine strength and size of the temporalis muscle) has occurred in this guild over an evolutionary time scale, potentially to reduce interspecific competition (Jones 1997). Despite these morphological distinctions, interspecific competition has been documented in modern food webs of marsupial carnivores, and has led to disparities in population densities among the guild (Jones and Barmuta 1998). Interference competition in the form of kleptoparasitism, intraguild killing, spatiotemporal avoidance or a combination of these may occur among sympatric predators if they utilise similar prey resources (Palomares and Caro 1999; Glen and Dickman 2005). Kleptoparasitism refers to the interspecific stealing of already procured food (Ruxton and Moody 1997), whereas intraguild killing involves the killing or eating of potential competitors for a resource that is usually limited (Polis *et al.* 1989). Tasmanian devils display dominance behaviour at carcasses and have the capacity to steal food from quolls (Jones 1998; Jones and Barmuta 2000). The specialised auditory region of dasyurids (Archer 1976) may have given them a competitive advantage over thylacines by helping them locate prey and avoid predators and bounty hunters (Wroe 1996).

By far the largest carnivore, the thylacine may have suppressed the population size of other marsupial carnivores by killing them or their young. Thylacine predation on both quoll species was suggested within several decades following the thylacine's extinction, yet there were no direct evidence of marsupial carnivore predation by the thylacine (Paddle 2000). Suppression of predators by larger competitors is common in terrestrial ecosystems (Carbone *et al.* 1997; Mills and Gorman 2003). Tasmanian devils currently have no known natural predators (Jones 1998) and have been observed killing the much smaller eastern quoll (Jones *et al.* 2004b). Tasmanian devils are less vigilant with increasing body size and age (Jones 1998) and were small enough to be killed by thylacines. The overlap in $\delta^{15}\text{N}$ values between thylacines and quolls would suggest that thylacines were not largely eating marsupial carnivores. Tasmania's carnivorous megafauna, specifically the marsupial lion (*Thylacoleo carnifex*) may well have hunted the thylacine and other large species (Owen 2003).

Interspecific differences in $\delta^{13}\text{C}$ values between the thylacine and all remaining Tasmanian marsupial carnivores may be attributed to distinctions in their habitat or prey utilisation. C_4 photosynthesisers (e.g., tropical and warm-climate grasses) have relatively high $\delta^{13}\text{C}$ values (-8‰ to -16‰), C_3 photosynthesisers (e.g. all trees and most shrubs, herbs and cool climate grasses) have low $\delta^{13}\text{C}$ values (-23‰ to -34‰) and CAM photosynthesisers have $\delta^{13}\text{C}$ values anywhere between these extremes (Smith and Epstein 1971; Smith and Brown 1973; Deines 1980; O'Leary 1981; 1988). The lower $\delta^{13}\text{C}$ values of the thylacine may suggest that they tended to inhabit denser, wooded regions compared to the Tasmanian devil, spotted-tailed quoll and eastern quoll (Pate and Noble 2000). This distinction in predator $\delta^{13}\text{C}$ values may also partly be attributed to the canopy effect, whereby the $\delta^{13}\text{C}$ of leaves in closed forests gradually decrease from the canopy to the floor (Medina and Minchin 1980; Schleser and Jayasekera 1985; van der Merwe and Medina 1989; Hanba *et al.* 1997). The spotted-tailed quoll is distinguished by a greater degree of arboreal activity than the other marsupial carnivores, allowing them to gain access to arboreal prey species (Jones and Barmuta

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

2000). The spotted-tailed quoll has higher $\delta^{13}\text{C}$ values than ground-dwelling carnivores in closed forests partly because they consume a higher proportion of arboreal prey that feed from the upper canopy. There are many documented cases of small predators modifying their behaviour to reduce the risk of encounters with larger predators (Switalski 2003; Berger and Gese 2007; Thompson and Gese 2007; Merkle *et al.* 2009) and this may contribute to different habitat use patterns in sympatric carnivores. Herbivorous prey in mosaic habitats may include grazer and browser species that would have different $\delta^{13}\text{C}$ values. In this situation, predators in the same habitat would have different $\delta^{13}\text{C}$ values if they were consuming a different proportion of browser to grazer species. Therefore, the difference in predator $\delta^{13}\text{C}$ values could reflect different habitat preference or simply different prey in the same habitat.

4.5.3 Sexual dimorphism and resource use

Sexually dimorphic species are likely to possess differences in reproductive costs, metabolic rate and morphology. These distinctions may lead to sex-based differences in foraging strategies or habitat use, partly due to divergence in the energetic needs of each sex (Breed *et al.* 2006). Despite their marked sexual dimorphism, we saw no intersexual differences in their $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values. The sexual dimorphism of thylacines may alternatively be a product of their polygynous breeding system but this would not preclude them from needing different diets. In general, large male body size often increases mating success due to intra-sexual competition or female choice and the ability to exploit a wider range of prey (McPherson and Chenoweth 2012), while smaller females may be favoured to reduce energy needs for daily maintenance and are more efficient in hunting smaller prey (Moors 1980). Thylacines were territorial and occasionally formed breeding pairs and small family groups (Paddle 2000). Competition among males for breeding females may potentially have occurred in this species, with larger-sized males favoured (Clutton-Brock and Harvey 1978).

As no sex-based differences in $\delta^{15}\text{N}$ values were detected in any of the marsupial carnivores investigated in this study, it is likely that they share a similar diet, or at the least, are feeding from the same trophic level, whereas similarities in $\delta^{13}\text{C}$ values between sexes may indicate shared habitat use within each species.

4.5.4 Long-term change in stable isotope ratio of thylacines

The thylacine showed no change in their $\delta^{13}\text{C}$ or $\delta^{15}\text{N}$ values over the several decades explored, nor did they change after the initiation of the government bounty scheme. Processes that may account for the lack of change in the stable isotope ratio of thylacines include having a consistent trophic position, foraging habits/location and isotopic composition of the base of the food web. We note that the time frame investigated for the thylacine was considerably smaller than extant marsupial carnivores and the change observed in other carnivores was after the time the thylacine was removed from the ecosystem. Further research is required to assess the biological effects of anthropogenic disturbances on the thylacine population and will benefit from the inclusion of recovered fossil specimens that predate European arrival (Sleightholme 2011).

4.5.5 Long-term change in $\delta^{13}\text{C}$ of extant marsupial carnivores

The gradual decline in Tasmanian devil and spotted-tailed quoll keratin $\delta^{13}\text{C}$ values is suggestive of a change in the average isotopic composition of plants that serve as the base of the food web. Finer scaled analysis of the temporal dataset revealed a lowering of Tasmanian devil $\delta^{13}\text{C}$ values during 1980-1999. As $\delta^{13}\text{C}$ changes little up the food chain, the $\delta^{13}\text{C}$ values of terrestrial generalist predators should provide a good indication of the surrounding plant community. Plants in dense-canopied environments have $\delta^{13}\text{C}$ values that are at least 2‰ lower than plants found in open environments due to differences in the relative proportions of C_3 and C_4 plants in these ecosystems (Heaton 1999). In view of this information, the trend observed in marsupial carnivore

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

$\delta^{13}\text{C}$ values may be caused by (i) the movement of marsupial carnivores to closed environments (forests and rainforests) from open mosaic environments (grasslands and woody grasslands) as mosaic habitats changed to completely open agricultural landscapes, (ii) changes to the dominant plant forms and species in Tasmania through woody-weed invasion, or (iii) a combination of these.

4.5.5.1 Movement of marsupial carnivores from open to closed environments

The loss of mammalian carnivores worldwide has been greatest in open environments; these areas have undergone substantial land conversion from native vegetation to agricultural land and plantations (Laliberté and Ripple 2004) and have increased species risk of predation (Gittleman 1985; Warwick 1998). In Tasmania, deliberate killing of marsupial carnivores by landholders shortly after European settlement was largely in response to poultry and livestock losses (Fleay 1932; Green and Scarborough 1990; Burnett 1993; Watt 1993; Burnett and Marsh 2004). Spotted-tailed quolls were shot, trapped and poisoned by farmers regardless of whether or not they were threatening poultry (Watt 1993; Long and Nelson 2010) and bounty schemes were introduced as early as 1830 to remove Tasmanian devils from rural areas in northwest properties (Guiler 1982; Owen and Pemberton 2005). The selective removal of native predators from farming and agricultural regions would have placed selective pressure on the both species, favouring individuals that inhabited and foraged in relatively undisturbed dense forested habitats.

The effect of increased disturbance to open environments such as agriculture and livestock grazing may have forced animals in these areas to relocate to denser forest areas. The risk-disturbance hypothesis explains that animals should seek a balance between avoiding disturbance and pursuing activities that may increase fitness such as food and mating (Frid and Dill 2002). Prior to European settlement, native herbivores, including the forester kangaroo and common wombat undoubtedly played an important role in the ecology of lowland grasslands. They graze on the native grasses and used

tussocks for shelter (Lunt 1991). Spotted-tailed quolls are considered forest-dependent (Green and Scarborough 1990; Jones and Barmuta 2000) although they have been observed hunting in grassland and pastoral areas adjacent to forested areas (Jones and Rose 1996). The movement of carnivores from an open grassland community to a closed forest community will result in a decrease to their $\delta^{13}\text{C}$ values. Tasmanian devils were legally protected from 1941 and their population began to expand into former areas since the 1960's though much of their habitat has been modified for human purposes (Bradshaw and Brook 2005) and has experienced extreme changes in vegetation cover (Gilbert 1959; Jackson 1968).

4.5.5.2 *Woody plant invasion*

The second process that could account for the observed trends in extant marsupial carnivore $\delta^{13}\text{C}$ values is one in which there is a gradual shift in the dominant plant forms and species towards denser vegetation as a result of changes in fire regimes, livestock grazing pressure, fertilisation and climate. Woody plants typically possess the C_3 photosynthetic pathway while grasses primarily use the C_4 photosynthetic pathway (Heaton 1999; Cerling *et al.* 1997). Frequent fires are sufficient to limit the cover and spread of woody plants, while fire exclusion can lead to shrub dominance or complete conversion to woodlands in as little as a few decades. Aboriginal people used frequent, low-intensity burning regimes, and were usually initiated under conditions when the vegetation was too wet to burn (Marsden-Smedley 1998; Marsden-Smedley and Kirkpatrick 2000; Enright and Thomas 2008). The fire regime shifted following the displacement of Aboriginal people by Europeans to fewer fires, followed by regional scale fires (Marsden-Smedley and Kirkpatrick 2000; Enright and Thomas 2008). Cessation of Aboriginal fire regimes increased the chance of trees, such as *Eucalyptus* and rainforest undershrubs becoming dominant (Jackson 1968).

Increases in atmospheric CO_2 over the past 200 years has potentially given a significant advantage to C_3 species relative to C_4 plant species in many regions of the world with

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

respect to physiological activity, growth and competitive ability (Archer *et al.* 1995). Furthermore, when native pastures have an increase in available resources (e.g. adding fertilisers), the vegetation may change and can result in the loss of native grass cover (Mokany *et al.* 2006). In total, more than 83% of native grasslands have been replaced after nearly 180 years of agricultural exploitation and exist today as small fragmented patches (Fensham 1989). The encroachment of woody plants into grass-dominated communities is a global phenomenon and has far reaching consequences; altering plant resource availability, hydrology and patterns of nutrient cycling (Van Auken 2009). Woody plant invasion can also have bottom-up effects on native fauna through changes in habitat structure, prey abundance and species richness (Seamster 2010). Further understanding of these potentially long emerging threats is imperative to improve biodiversity conservation and land management practices.

4.5.6 Long-term change in $\delta^{15}\text{N}$ of extant marsupial carnivores

During the mid 20th century the spotted-tailed quoll experienced a lowering in their $\delta^{15}\text{N}$ values of nearly one trophic level, followed by an increase in recent times, whereas the Tasmanian devil experienced an increase in $\delta^{15}\text{N}$ values during 1980-1999. Two processes may account for the variability in the $\delta^{15}\text{N}$ values of both species over time; temporal changes in the isotopic composition of the base of the food web and/or changes in marsupial carnivore diet/trophic position.

4.5.6.1 Diet/trophic variability

A shift in the prey consumed may be accompanied by a change in trophic level of the consumer as shown by their $\delta^{15}\text{N}$ values. A decrease in consumer $\delta^{15}\text{N}$ values would indicate that they are consuming a greater proportion of lower trophic level prey than they were formerly. As opportunistic predators, the Tasmanian devil and spotted-tailed quoll should be able to switch to alternative prey items when there are shifts in the density of other trophic groups (Ben-David *et al.* 1997). Dietary shifts have been

reported seasonally in the Tasmanian devil and both quoll species (Jones and Barmuta 2000) and over several consecutive years in the spotted-tailed quoll (Belcher 1995) but have not been investigated over longer time scales.

The disappearance of large carnivores often leads to the simplification of ecosystems through cascading effects that reduce species diversity. The population collapse of Tasmanian devils in the early 1900's and extinction of the thylacine in 1936 (Guiler 1992) may have had a drastic cascading effect down the food chain, resulting in changes to prey abundance and distribution. Smaller predators, such as the Tasmanian devil and quoll species may have switched to larger prey when the thylacine went extinct (Dickman 1988). Consequently, prey size selection could depend more on competition among predator guild members than on inherent relationships between predator and prey sizes (Radloff and Du Toit 2004).

Multiple other factors may have driven the apparent decline in native prey populations in Tasmania over the past century (Edgar *et al.* 2005); overexploitation, habitat alteration, exotic species introduction, climate change and rainfall variation (Belcher 1995; Steffen and Scientific 2009). Many native mammalian herbivores were killed by Europeans, with more than 900, 000 possums and wallabies caught in 1923 for their pelts (Owen and Pemberton 2005). Most animals that were hunted for meat and fur were herbivores, which have lower $\delta^{15}\text{N}$ values compared to insectivorous or carnivorous species. It has been suggested that the arboreal lifestyle of spotted-tailed quolls may have allowed them to easily access these snared animals when they were suspended above the ground (Owen and Pemberton 2005). An overall reduction in spotted-tailed $\delta^{15}\text{N}$ values would occur if a higher proportion of low trophic level species were consumed, such as snared possums and wallabies. Controlled trapping and killing of native animals was introduced under the Animal and Birds Protection Act 1929 and may have assisted the recovery of herbivore species, making them more readily available to native predators (Guiler 1957). The trade of red-necked wallaby,

4. ASSESSING MARSUPIAL CARNIVORE DIET USING STABLE ISOTOPES

Tasmanian pademelon and common brushtail possum fur and/or meat continues today but is now under strict regulation, whereas the large forester kangaroo and common ringtail possum are no longer hunted commercially.

The supplementation of introduced herbivores in the diet of Tasmanian devils and spotted-tailed quolls in the form of carcasses of domestic animals (Guiler 1970; Owen and Pemberton 2005) or killing of lambs and poultry (Gould *et al.* 1974) may also have contributed to changes in predator $\delta^{15}\text{N}$ values. During the 19th and early 20th century, poultry were usually fed from the stubble fields and waste grain (Knibbs 1913), whereas sheep grazed in pastures. Given this herbivorous diet, sheep and poultry should have a low $\delta^{15}\text{N}$ value during early European settlement compared to meat eaters. The $\delta^{15}\text{N}$ values of a nursing lamb will be one trophic level higher than its mother until weaning begins as the lamb is consuming its mother's milk (Polischuk *et al.* 2001; Newsome *et al.* 2006; Dalerum *et al.* 2007). Introduced species are more likely to be hunted by native carnivores than native prey if they are more readily catchable, accessible or abundant, as has been observed in many regions of the world (Andelt 1985; Corbett and Newsome 1987; MacCracken and Hansen 1987; Newsome *et al.* 1989; Maehr *et al.* 1990; Pech *et al.* 1992). Only recently have nitrogen fertilisers been applied to pastures, which would increase the $\delta^{15}\text{N}$ value of livestock feeding in these areas and subsequently, marsupial carnivores feeding off livestock.

4.5.6.2 Environmental influences

A shift in the $\delta^{15}\text{N}$ of consumers may represent changing processes at the base of the food chain due to nitrogen inputs into the terrestrial system, uptake of nitrogen by plants or a combination of these. Agriculture contributes excessive amounts of nitrogen to terrestrial systems, primarily in the form of manure ($\sim 5\text{‰}$) and inorganic fertiliser ($\sim 0\text{‰}$) (Kendall 1998). The $\delta^{15}\text{N}$ of manure is often further elevated through ammonia volatilisation during storage and application, and N from both sources is subject to further $\delta^{15}\text{N}$ increases through denitrification (Kendall 1998). Leaching or gaseous

losses occur when nitrogen deposits exceed the biological demand of the ecosystem, resulting in the preferential loss of ^{14}N from terrestrial environments (Templer *et al.* 2007). Plants will take up the remaining relatively enriched ^{15}N as the ecosystem gets closer to reaching nitrogen saturation (Högberg and Johannisson 1993). Herbivores will have $\delta^{15}\text{N}$ values reflecting that of the leaf tissues consumed and are subsequently transferred up the food web (Evans 2001). Thus, the increased $\delta^{15}\text{N}$ values reported over recent decades in the Tasmanian devil and spotted-tailed quoll could be an indicator of animal waste pollution or nitrogen loss through denitrification.

4.5.6.3 Verifying cause of temporal $\delta^{15}\text{N}$ shift

The cause of temporal changes in top predator $\delta^{15}\text{N}$ values may be verified by tracking $\delta^{15}\text{N}$ values of prey species over the same time frame. An analogous temporal shift in prey and predator $\delta^{15}\text{N}$ values would lend support towards changes to nitrogen uptake by plants or nitrogen input into the ecosystem (e.g. fertiliser).

4.6 CONCLUSION

We infer that hunting of small to medium bodied mammals was likely a major element of thylacine subsistence in Tasmania, and may have included the Common wombat, Bennett's wallaby and Tasmanian pademelon. The dramatically reduced abundance and range of Tasmania's herbivorous mammals following European settlement has important implications for the thylacine population that were heavily reliant on medium-bodied prey. We also detected several differences in the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between the thylacine and sympatric marsupial carnivores, suggesting potential variation in the habitat use and/or diet between these species. By combining historic and contemporary marsupial carnivore samples dating back to the 19th Century, we provide a new perspective of the ecology of marsupial carnivores and insight into past vegetation changes in Tasmania that can be used to aid land management initiatives.

4.7 ACKNOWLEDGEMENTS

We are especially grateful to S. Sleightholme for assisting in the identification of suitable thylacine material for this study. Marsupial carnivore tissues used in this study were provided by the American Museum of Natural History, New York; Booth Museum of Natural History, Brighton and Hove, UK; Cambridge University Zoological Museum, Cambridge; Field Museum of Natural History, Chicago; Forschungsinstitut and Schaumuseum Senckenberg, Frankfurt; Gothenburg Natural History Museum, Sweden; Hunterian Museum and Art Gallery, Glasgow; Glasgow University, Glasgow; Leeds City Museum, Leeds; Museum Victoria, Victoria; Naturhistorisches Museum Wien, Vienna; National History Museum, Oslo; National Museum of Australia, Canberra; University of Tasmania, Tasmania; Royal College of Surgeons of England, London and the Queen Victoria Museum and Art Gallery, Tasmania. Dried skin samples of potential prey species were provided by Queen Victoria Museum and Art Gallery and Senckenberg Gesellschaft für Naturforschung, Görlitz. Fresh tissue samples were also provided by B. Fancourt, S. Peck, D. Stanioch and A. Palmer. Acquisition of road kill was approved by the Department of Primary Industries, Parks, Water and Environment (Permit No. TFA 11163). This study was supported by an Australian Research Council grant to T. Rogers, an Evolution and Ecology Research Centre Postgraduate Research Start-Up Grant to M. Attard and the Postgraduate Writing and Skills Transfer Award to M. Attard sponsored by the Evolution and Ecology Research Centre at the University of New South Wales.

Chapter 5

CONCLUSION: THE INTERACTION BETWEEN DIET AND EXTINCTION RISK AND THE FUTURE OF AUSTRALIA'S CARNIVOROUS MARSUPIALS



*"What escapes the eye is the most insidious kind of extinction - the extinction of
interactions"*

Dan Janzen

5.1 KEY FINDINGS OF THE THESIS

5.1.1 Thylacine prey size and extinction risk

The primary aim of this thesis was to identify and assess the role of prey size and resource competition in the extinction of the thylacine (*Thylacinus cynocephalus*) in Tasmania. The skull mechanics of two extinct members of Thylacinidae were compared with extant relatives for the first time using finite element modelling to determine the maximum prey size of the thylacine (Chapter 2 and 3). Using this approach, we found that the thylacine was not well-adapted to tackle prey approaching or exceeding their own body mass, particularly if they were hunting alone. The mainland fossil thylacinid, *Nimbacinus dicksoni*, appears to share greater similarities in predatory behaviour to generalist daysurids than the thylacine (Chapter 3), and supports the notion that the thylacine was a more specialised carnivore (Wroe and Musser 2001). By assessing the capacity of the thylacine skull to withstand loads generated by jaw adductors or struggling prey in relative contexts has provided new insight in how its skull once functioned and why it was shaped in a particular way.

In both biomechanics studies in this thesis (Chapter 2 and 3), the thylacine was the only marsupial carnivore to experience peak stress at the rostrum during a canine bite, which may be because of their unique skull shape (Wroe and Milne 2007). Carnivores with different snout lengths experienced a trade off in mechanical performance (Pierce *et al.* 2008). In theory, a longer snout is mechanically weaker at the canines than in shorter snouted species due to an increased distance between the canines and the point of jaw articulation. In trade, longer snouts offer an increased speed of attack through having a longer outlever and are typically considered better suited to small, agile prey (Wroe and Milne 2007). The skull of the thylacine was large but lightly constructed and possessed a long, narrow rostrum adapted to quickly snapping up live prey. Snout morphology did not appear to be a limiting factor for the thylacine in an earlier comparative study with placental dingo (*Canis lupus dingo*) (Wroe *et al.* 2007). However, predictions relating to

5. CONCLUSION: MARSUPIAL DIET, COMPETITION AND EXTINCTION RISK

the morphological function of extinct species benefit from comparisons to close living relatives (Lauder and Thomason 1995).

The prey composition of the thylacine was estimated using a stable isotope mixing model that incorporated native predator and prey tissues dated prior to the extinction of the thylacine (Chapter 4). Medium-sized mammalian prey were likely to form the bulk of the thylacine diet and may have included the common wombat (*Vombatus ursinus*), red-necked wallaby (*Macropus rufogriseus*) and Tasmanian pademelon (*Thylogale billardierii*). These species fall within the expected size range of prey needed to sustain large (>21.5 kg) mammalian carnivores within a terrestrial environment (Carbone *et al.* 1999). This would suggest that the thylacine diet was sufficient to meet their energetic requirements. However, the average body mass of prey identified isotopically did not exceed the body mass of thylacines, and may reflect a preference in prey size or physical limitation in the size of prey they could catch. Co-operative hunting may have permitted predation upon larger prey, such as the forester kangaroo (*Macropus giganteus*), but the stable isotope signature of this herbivore suggested that they were a less important component of the thylacine's diet.

The contention that thylacines hunted prey weighing less than their own body mass is supported by other comparative biomechanical and morphological studies. The bite force of the thylacine has been predicted to be higher than the dingo. However, its skull experienced greater stress under conditions that simulate the influence of struggling prey, suggesting they were limited to small- to medium-sized prey (Wroe *et al.* 2007). Mammalian predators that target larger prey have a greater risk of breaking and fracturing their canines from catching and killing larger prey compared to predators that specialise in smaller prey. Jones and Stoddart (1998) observed low rates of canine tooth wear and fracture in the thylacine compared to large prey specialists and proposed that the thylacine relied on prey weighing up to 5 kg. The stable isotope findings of this thesis suggest that slightly larger species formed the bulk of their diet (Chapter 4). Anecdotal and historical evidence reveal a decreasing availability of medium-sized

herbivores following European settlement (Animal Welfare Advisory Committee Australia 2003). This would have had unintended consequences for thylacines that heavily relied on these species for survival unless they were able to adapt to changing food resources and still meet their energetic needs.

5.1.2 Predatory behaviour of *Nimbacinus dicksoni*

Australia has long been dominated by marsupials, and among these one family now dominate carnivorous niches, the dasyurids, which include quolls or native ‘cats’ and the notorious Tasmanian devil, as well as dozens of smaller marsupial ‘mice’ species . However, in recent decades it has become clear that dasyurids were once relatively rare and another, related, but now extinct marsupial carnivore family, the thylacinids, were dominant. Although the number of known fossil species of thylacinid has greatly increased from 1 to 11, few are known from substantial cranial material and predictions of feeding ecology have been largely qualitative. However, one species the ~25-15 million year old *Nimbacinus dicksoni* is represented by a well-preserved skull. It is now thought that the thylacine was more specialized than large living dasyurids and far more restricted in dietary breadth, possibly making it more vulnerable to extinction. Using a sophisticated virtual 3D reconstruction techniques and a digital engineering approach we asked whether killing behaviour and prey size in the fossil *N. dicksoni* was more similar to that of the thylacine, or to similar-sized living dasyurids. We find that mechanical behaviour of the *N. dicksoni* skull was closest to that of the largest native ‘cat’, the spotted-tailed quoll and well-removed from that of the thylacine and conclude that unlike its recently extinct relative, the fossil species hunted a wide range of prey, up to and likely exceeding its own body mass.

5.1.3 A case for intraspecific competition

There are many instances where carnivores have been adversely influenced by other members of the guild when they use the same resources (Mills and Mills 1982; Johnson and Franklin 1994; Palomares *et al.* 1996; White and Garrott 1997). This thesis

5. CONCLUSION: MARSUPIAL DIET, COMPETITION AND EXTINCTION RISK

demonstrates that thylacines fed on similar sized prey as the Tasmanian devil (*Sarcophilus harrisii*) and spotted-tailed quoll (*Dasyurus maculatus*) (Chapter 1). There was also marked trophic overlap between thylacines and both quoll species, while the Tasmanian devil had a marginally higher trophic position than the thylacine (Chapter 4). The diet of the Tasmanian devil and both quoll species has been studied in detail and include species isotopically identified as the main prey of the thylacine. For example, Tasmanian devils have been observed catching live prey up to 30 kg in size, including common wombats and red-necked wallabies (Jones and Barmuta 1998). The diet of spotted-tailed quoll and eastern quoll (*Dasyurus viverrinus*) consist primarily of mammals, though female spotted-tailed quolls also eat a high proportion of birds (Jones and Barmuta 1998). The presence of the thylacine may have affected the space use by the two largest remaining carnivores, the Tasmanian devil and spotted tailed quoll, as signified by differences in their $\delta^{13}\text{C}$ values (Chapter 4). If the space use of smaller carnivores was influenced by thylacine presence, then the dramatic decrease and eventual extinction of the thylacine may have allowed the smaller carnivores to increase their numbers.

Surprisingly, we did not identify intersexual differences in the stable isotope composition of the thylacine despite their marked sexual dimorphism however additional samples will be required to confirm these findings. Similarly, none of the extant marsupial carnivores displayed intersexual differences in stable isotope values, which suggests potential intraspecific competition for food and habitat resources.

5.1.4 Ecological responses to prolonged human disturbances

Our stable isotope findings augment the limited understanding of the changes in the terrestrial Tasmanian food web over the last ~180 years, since few records exist that might provide insights into historical food web interactions and energy flow pathways (Fensham 1989; Marsden-Smedley 1998). Century-long transition in keratin $\delta^{13}\text{C}$ values were found for the Tasmanian devil and spotted-tailed quoll and provide evidence of

major ecosystem responses to prolonged anthropogenic disturbances. Grassy woodlands and grasslands have been the most seriously affected vegetation type in Tasmania, being replaced by crops, pastures and aggressive woody plants since Europeans arrived over 200 years ago (Fensham 1989). Our isotopic data reflect significant changes between the proportion of C₃ and C₄ vegetation inhabited by marsupial carnivores over time, supporting a gradual transition in habitats utilised by apex predators towards denser woody areas.

We identified two processes that may separately or in combination be responsible for decadal changes in Tasmanian devil and spotted-tailed quoll $\delta^{15}\text{N}$ values following European settlement. Temporal variation in $\delta^{15}\text{N}$ consumer tissues is normally interpreted as a shift in their trophic position, reflecting a change in the relative proportions of each prey species assimilated. Severe habitat modification and predation by humans, foxes and feral cats is likely to have reduced the number of native terrestrial prey available to marsupial carnivores. As opportunistic predators, the Tasmanian devil and spotted-tailed quoll take advantage of carrion and are able to hunt a range of prey including species exceeding their own body mass (Jones and Barmuta 1998), allowing them to make full use of available prey. Changes in the community assemblage are likely to be reflected in the tissues of generalist predators as they typically switch their diet in response to changes in prey species abundance (Murdoch 1969). Alternatively, this perceived trophic level shift may actually be the result of changing isotopic compositions at the base of the food web. For example, nitrogen inputs into the soil from agriculture (e.g. manure and inorganic fertiliser) can lead to the preferential loss of ^{14}N from terrestrial environments through leaching or gaseous losses (Templer *et al.* 2007). Plants will take up the remaining relatively enriched $\delta^{15}\text{N}$ (Högberg and Johansson 1993) that is then assimilated into the tissue of primary consumers. Ecosystem-based restoration efforts typically involve the establishment or rehabilitation of self-sustaining native communities that reflect historical conditions (Lichatowich *et al.* 1995). The lack of baseline information on vegetation assemblage and food web

5. CONCLUSION: MARSUPIAL DIET, COMPETITION AND EXTINCTION RISK

interactions presents a major challenge in evaluating changes of a given ecosystem (Vander Zanden *et al.* 2003). The isotope-based food web and environment reconstructions presented in this thesis reveal long-term ecological changes that may be used to set historically relevant restoration targets in Tasmania.

5.2 RECOMMENDATIONS FOR FUTURE WORK

5.2.1 Were thylacines individual specialists?

Thylacines in Tasmania occupied a wide trophic range and may be described as a generalist population (Chapter 4). In recent years, attention has been drawn to the idea that individuals can occupy a specialised niche within a generalist population (Bolnick *et al.* 2003). Individual specialisation is common among generalist carnivore populations (Shaffer 1971; Pierotti and Annett 1987; Thiemann *et al.* 2011) and has significant ecological and evolutionary implications (Van Valkenburgh 2007). Scientific evidence to support individual dietary variation in the thylacine is largely limited. Robson and Young (1990) reported individual variation in tooth microwear, which may potentially be attributed to dietary differences within the species. However dietary interpretations based on this study were inconclusive due to their low sample size. Intra- and inter-individual dietary variation was not addressed in this study as the isotopic data for each individual only looked at a 'snapshot' in time. Accurate designation of resource variation within and between thylacine individuals will be important in assessing the complete diversity of trophic interactions and will require longitudinal records of individual behaviour.

The excellent preservation of thylacine tissues in museums raises the possibility of obtaining temporal dietary records of thylacine individuals long after they have become extinct. The analysis of multiple tissue types with different turnover rates from the same individual can be used to assess temporal variation in individual diet, and has been frequently applied to wild populations (Dalerum and Angerbjörn 2005). Alternatively,

metabolically inactive tissues that grow progressively over time such as hair, vibrissae and nails can be used to provide chronological isotopic records of individuals from ancient and modern populations (White *et al.* 1999; O'Connell *et al.* 2001; Bearhop *et al.* 2003; Roy *et al.* 2005; Furness *et al.* 2006; Cherel *et al.* 2007; Harrison *et al.* 2007). The stable isotope signature of inert tissues remains unchanged once synthesised as the protein remains biochemically unchanged as it is laid down (Gannes *et al.* 1998). This allows stable isotopes measurements to be serially sampled along inert tissues to infer isotopic changes within an individual over the period of tissue growth (Hobson and Wassenaar 1997). For example, thylacine vibrissae collected as part of this thesis may be used to unlock sequential isotopic information of these individuals.

5.2.2 Temporal change in predator $\delta^{15}\text{N}$ values: is it a real 'trophic' shift?

To verify the cause of the temporal $\delta^{15}\text{N}$ shift observed in Tasmanian devil and spotted-tailed quoll, isotopic information of prey species will need to be compared over the same time period where the change in predator $\delta^{15}\text{N}$ values has occurred. A large collection of prey materials are available in museum archives that were collected from different times since European settlement. A temporal shift in primary consumer $\delta^{15}\text{N}$ values at the same time as that found in these predators would support the notion that this change is due to variation in $\delta^{15}\text{N}$ at the base of the food chain. Consistency in prey $\delta^{15}\text{N}$ values over time would more likely indicate that marsupial carnivores have altered their diet.

5.2.3 Caveats to stable isotope studies

The caveats of applying stable isotope methods to extinct species complicate their use in reconstructing their ecology and environment. Firstly, the isotopic fractionation (change in isotope ratios) between a consumer tissue and their diet must be accurately estimated to reconstruct the diet of extinct species and infer historic inter-specific trophic relationships (Ambrose *et al.* 2003). It is ideal to obtain species-specific trophic fractionations for a given tissue, however, in most cases this is not possible, and trophic

5. CONCLUSION: MARSUPIAL DIET, COMPETITION AND EXTINCTION RISK

fractionations from a related species, or averaged trophic fractionations from multiple species with similar feeding niches are used (McCutchan *et al.* 2003). Species-specific fractionation values are unable to be obtained for extinct species, so we applied values taken from placental carnivores in the literature to construct the diet of the thylacine. The examination of diet-bone collagen enrichment factors in marsupials, particularly marsupial carnivores, would be preferable to taking enrichment factors of placental carnivores from the literature. As of yet, diet-bone collagen enrichment factors has not been studied in marsupials and presents a new avenue for future research.

Secondly, the collective use of multiple tissues in this study may be problematic due to variation in the amino acid concentration or composition of tissues within an individual, which can lead to small isotopic differences between tissues (Hedges 2003). Tissues may also vary isotopically due to variation in the composition of different dietary constitutions. Our pilot study compared stable isotope ratios in Tasmanian devil tissues to control for potential isotopic differences between tissues, and thereby allow multiple tissue types to be used in conjunction. Most studies used to determine isotopic fractionation between consumer tissues use captive animals fed fixed diets to control for potential biases in amino acid composition between diets (Gratton and Forbes 2006; MacNeil *et al.* 2006). As some of the tissues of interest are highly invasive to sample from live animals (e.g. bone), the tissue enrichment values used in this study were based on tissues collected opportunistically from deceased animals. As these animals were not fed fixed diets, variations in stable isotope enrichment in predator tissues may due, at least in part, to differences in food quality and diet (Oelbermann and Scheu 2002). This pilot study would benefit from using a higher sample size and including additional taxa to verify the findings.

An important caveat was that we did not control for the specific region in Tasmania where specimens were collected due to the rarity of historic specimens. Heterogeneity in isotope values among conspecifics may be due to variation in consumer prey preferences, or may be caused by temporal and/or spatial variation in prey availability

or isotopic composition (Layman *et al.* 2007). In the latter case, individuals specialised on the same resource by feeding consistently in different areas may differ greatly in isotope values if there is spatial heterogeneity in resource isotopes, so that habitat-derived variation in consumer isotopes may be mistaken as diet variation (Flaherty and Ben-David 2010). Similarly, individuals sampled at different time periods that specialise in the same resource may have temporal heterogeneity in consumer isotope values. Knowledge of spatial and temporal variability in prey isotope values will greatly improve the interpretation of consumer isotopic space (Layman *et al.* 2007), yet are near impossible to obtain for historical food webs. Nevertheless, the potential role of diet and competition in the marsupial carnivores investigated cannot be ignored, and the available data suggests interesting and important temporal trends, which should be explored further.

5.2.4 Looking into the future: the management of critically endangered carnivores

Since the extinction of the thylacine there has been a renewed need to understand the behaviour of critically endangered species. The diet of Australia's large threatened dasyurids is quite broad and variable as their diet consists of multiple species that can change over space and time (Jones and Barmuta 1998; Dawson *et al.* 2007). A comprehensive understanding of the present diet and habitat use of these top predators will be critical for the effective management of both wild and captive populations by (i) providing clarity on their ecological function, (ii) identifying important habitats and prey species, and (iii) assessing ecological impacts of free-range enclosures and natural populations on prey as well as mesopredators (middle trophic level predators) that are normally controlled through competition or predation by larger predators.

Analysis of prey remains from fecal samples or stomach contents have provided important insights into the diet of marsupials including Tasmanian devils (Taylor 1986; Marshall and Cosgrove 1990; Pemberton *et al.* 2008), yet have several disadvantages

5. CONCLUSION: MARSUPIAL DIET, COMPETITION AND EXTINCTION RISK

and potential biases. These conventional dietary techniques are limited in their ability to resolve temporal patterns in diet due to small sample sizes and nonrandom sampling, and potential pseudoreplication when independent samples are from the same subject (Reynolds and Aebischer 1991; Deb 1997; Darimont and Reimchen 2002). Partial remains and the prevalence of indigestible material such as bone limits the accuracy of assessing the relative importance of particular prey species to an individual's diet (Burton and Koch 1999) and represent their most recently digested food rather than diet *per se*. Stable isotope measurements in animal tissues overcome some of the shortcomings of traditional dietary studies because they reflect an individual's diet over time. A further advantage is that unlike conventional techniques, results based on stable isotope data reflect assimilated and not ingested foods (Hobson *et al.* 1996). Through our investigation we found that $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ can be used to discriminate among prey species belonging to different niches (Chapter 4) and may be further used to address the diet of other terrestrial predators in Australia. Stable isotope techniques are most effectively used in combination with conventional approaches to verify diet (Dehn *et al.* 2007) and we highly recommend its use in future ecological studies.

APPENDIX A

Specimen records and prey information

Appendix A1 Specimen records for all carnivorous marsupials included in the study, including the thylacine, Tasmanian devil, spotted-tailed quoll and eastern quoll.

Information is provided on the sex (M = male, F = female, U = unknown), year of death/acquisition, locality and institution where each specimen.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Thylacinus cynocephalus</i>	SMF15682	1852	Tasmania	M	FSS	Whisker
<i>Thylacinus cynocephalus</i>	4088	1869	Tasmania	U	LCM	Bone
<i>Thylacinus cynocephalus</i>	4089	1869	Tasmania	U	LCM	Bone
<i>Thylacinus cynocephalus</i>	4091	1869	Tasmania	U	LCM	Bone
<i>Thylacinus cynocephalus</i>	LEED 1869.46.7	1869	Tasmania	U	LCM	Hair
<i>Thylacinus cynocephalus</i>	LEED 4087	1869	Tasmania	U	LCM	Bone
<i>Thylacinus cynocephalus</i>	LEED 4090	1869	Tasmania	U	LCM	Bone
<i>Thylacinus cynocephalus</i>	As. 7/2	1871	Tasmania	U	CZM	Hair
<i>Thylacinus cynocephalus</i>	As. 7/3	1871	Tasmania	M	CZM	Bone
<i>Thylacinus cynocephalus</i>	As. 7/4	1871	Tasmania	U	CZM	Hair
<i>Thylacinus cynocephalus</i>	As. 7/5	1871	Tasmania	F	CZM	Bone
<i>Thylacinus cynocephalus</i>	As. 7/8	1871	Tasmania	F	CZM	Bone
<i>Thylacinus cynocephalus</i>	C28746	1883	Tasmania	M	MV	Hair
<i>Thylacinus cynocephalus</i>	NZM6279	1885	Tasmania	U	NHM	Bone
<i>Thylacinus cynocephalus</i>	NZM6288	1885	Tasmania	U	NHM	Bone
<i>Thylacinus cynocephalus</i>	FMNH 81522	1888	Tasmania	F	FMNH	Bone

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Thylacinus cynocephalus</i>	NZM411	1888	Tasmania	M	NHM	Bone
<i>Thylacinus cynocephalus</i>	z503	1888	Tasmania	F	HM	Whisker
<i>Thylacinus cynocephalus</i>	300130	1897	Tasmania	M	BM	Hair
<i>Thylacinus cynocephalus</i>	C5746	1901	Tasmania	M	MV	Hair
<i>Thylacinus cynocephalus</i>	26.9.1910	1910	Tasmania	U	WML	Bone
<i>Thylacinus cynocephalus</i>	GMN782	1911	Tasmania	U	GNHM	Whisker
<i>Thylacinus cynocephalus</i>	A295	1920	North-west coast, Tasmania	U	TMAG	Bone
<i>Thylacinus cynocephalus</i>	A297	1920	North-west coast, Tasmania	U	TMAG	Bone
<i>Thylacinus cynocephalus</i>	RCSOM/A 368.5	1922	Woolnorth, Tasmania	M	RCS	Bone
<i>Thylacinus cynocephalus</i>	14788	1923	Tasmania	U	AMNH	Bone
<i>Thylacinus cynocephalus</i>	100102	1925	Tasmania	F	BM	Bone
<i>Thylacinus cynocephalus</i>	NMA 1991.0016.0001	1930	Pieman River, Tasmania	U	NMA	Hair
<i>Thylacinus cynocephalus</i>	77701	Unknown	Tasmania	U	AMNH	Bone
<i>Thylacinus cynocephalus</i>	A300	Unknown	North-west coast, Tasmania	U	TMAG	Bone
<i>Thylacinus cynocephalus</i>	A1277	Unknown	Tasmania	U	TMAG	Bone
<i>Thylacinus cynocephalus</i>	A1278	Unknown	Tasmania	U	TMAG	Bone
<i>Thylacinus cynocephalus</i>	A890	Unknown	Florentine Valley	M	TMAG	Bone
<i>Thylacinus cynocephalus</i>	NZM2010	Unknown	Tasmania	U	NHM	Bone
<i>Sarcophilus harrisii</i>	16200	1852	Tasmania	U	FSS	Whisker
<i>Sarcophilus harrisii</i>	SMF 16200	1852	Tasmania	U	FSS	Whisker
<i>Sarcophilus harrisii</i>	NZM2011	1888	Tasmania	M	NZM	Bone

APPENDIX

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Sarcophilus harrisii</i>	QVM:OLD:1:2017	1903	Launceston - Pt B, Tasmanian Northern Midlands, Doctors Creek, N. Tasmania	M	QVM	Whisker
<i>Sarcophilus harrisii</i>	A263	1908	Tasmania	U	TMAG	Bone
<i>Sarcophilus harrisii</i>	1921 - G Estate	1921	Gunns Estate, Launceston, Tasmania	U	QVM	Bone
<i>Sarcophilus harrisii</i>	65670	1922	Arthur River, Tasmania	M	AMNH	Bone
<i>Sarcophilus harrisii</i>	65671	1922	Arthur River, Tasmania	M	AMNH	Whisker
<i>Sarcophilus harrisii</i>	65672	1922	Arthur River, Tasmania	F	AMNH	Bone
<i>Sarcophilus harrisii</i>	65673	1922	Arthur River, Tasmania	F	AMNH	Whisker
<i>Sarcophilus harrisii</i>	65675	1922	Arthur river mill, Tasmania	F	AMNH	Whisker
<i>Sarcophilus harrisii</i>	65676	1922	Arthur river mill, Tasmania	M	AMNH	Whisker
<i>Sarcophilus harrisii</i>	QVM:1941:1:0179	1941	Launceston, Underwood, Tasmania	U	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1941:1:0226	1941	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1942:1:0157	1942	Winnaleah, Tasmania	U	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1946:1:0019	1946	Meander, Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	RCSOM/A 369.83	1951	Wynard, Tasmania	U	RCS	Bone
<i>Sarcophilus harrisii</i>	RCSOM/A 369.87	1951	Wynard, Tasmania	U	RCS	Bone
<i>Sarcophilus harrisii</i>	QVM:1963:1:0243	1962	Westbury, Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	1963.173.2	1963	Tasmania	U	WML	Bone

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Sarcophilus harrisii</i>	A2844	1963	Greens Creek, Tasmania	M	TMAG	Bone
<i>Sarcophilus harrisii</i>	A2827	1964	Woolnorth, Tasmania	M	TMAG	Bone
<i>Sarcophilus harrisii</i>	QVM:1964:1:0151	1964	Gladstone, Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1964:1:0157	1964	Ansons bay, , Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1964:1:0203	1964	Gladstone, Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	A774	1967	Pedder Hut, Tasmania	U	TMAG	Whisker
<i>Sarcophilus harrisii</i>	A2824	1969	Cape Portland, Tasmania	F	TMAG	Bone
<i>Sarcophilus harrisii</i>	QVM:1981:1:0129	1976	Avoca, Tasmanian Northern Midlands	F	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1982:1:0198	1980	Greens Beach, Tasmania	F	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM1988:1:0041	1980	Oxford, Tasmania	M	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1982:1:0016	1982	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1982:1:0017	1982	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1982:1:0159	1982	Meander Valley, Tasmanian Northern Midlands	U	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1984:1:0260	1984	Cressy, Tasmania	U	QVM	Hair
<i>Sarcophilus harrisii</i>	QVM:1986:1:0021	1986	Dorset, Ben Lomond, Scottsdale, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1987:1:0056	1987	Greens Beach, Tasmania	U	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1987:1:0085	1987	Greens Beach, Tasmania	F	QVM	Hair

APPENDIX

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Sarcophilus harrisii</i>	QVM:1988:1:0040	1988	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Sarcophilus harrisii</i>	160043	1991	Tasmania	M	FMNH	Whisker
<i>Sarcophilus harrisii</i>	160044	1991	Liavenec-Breona Hwy, Tasmania	F	FMNH	Bone
<i>Sarcophilus harrisii</i>	160045	1991	Tasmania	F	FMNH	Whisker
<i>Sarcophilus harrisii</i>	160046	1991	Mud Walls road, near Colebrook, Tasmania	M	FMNH	Bone
<i>Sarcophilus harrisii</i>	QVM:1996:1:0001	1992	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	F	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1996:1:0006	1992	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1996:1:0004	1993	Cradle Mountain, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	QVM:1996:1:0008	1993	Cradle Mountain, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	160047	1996	Campbell Town Road, 10 km W Lake Leake, Tasmania	F	FMNH	Whisker
<i>Sarcophilus harrisii</i>	160049	1996	Smithton, Tasmania	F	FMNH	Whisker
<i>Sarcophilus harrisii</i>	SH02	2000	Tasmania	F	QVM	Bone
<i>Sarcophilus harrisii</i>	SH04	2000	Mole creek, Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	SH10	2000	Tasmania	M	QVM	Bone
<i>Sarcophilus harrisii</i>	SH14	2000	Tasmania	F	QVM	Bone
<i>Sarcophilus harrisii</i>	SH12	2003	Adjacent to Coles Bay, Tasmania	U	QVM	Bone

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Sarcophilus harrisii</i>	SH13	2004	Bothwell, Tasmania	F	QVM	Bone
<i>Sarcophilus harrisii</i>	Freezer No. CO2744	2008	Lake Cresent Road, Bothwell near Lake Cresent	U	TMAG	Bone
<i>Sarcophilus harrisii</i>	Freezer No. CO2745	2008	Tasmania	U	TMAG	Bone
<i>Sarcophilus harrisii</i>	A3303	2010	Bothwell, Tasmania	M	TMAG	Bone
<i>Sarcophilus harrisii</i>	TAS12_019	2012	Cradle Mountain, Tasmania	F	Fieldwork	Bone
<i>Sarcophilus harrisii</i>	TAS12_020	2012	Cradle Mountain, Tasmania	M	Fieldwork	Bone
<i>Dasyurus maculatus</i>	A6 10/3	1869	Tasmania	F	CZM	Bone
<i>Dasyurus maculatus</i>	A552	1914	Macquarie Plains, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	QVM:1993:1:0032	1919	Waratah/Wynyard, Tasmania	U	QVM	Whisker
<i>Dasyurus maculatus</i>	65682	1922	Arthur river mill, Tasmania	M	AMNH	Whisker
<i>Dasyurus maculatus</i>	65683	1922	Arthur river mill, Tasmania	F	AMNH	Whisker
<i>Dasyurus maculatus</i>	65684	1922	Arthur river mill, Tasmania	M	AMNH	Whisker
<i>Dasyurus maculatus</i>	66162	1922	Arthur river mill, Tasmania	M	AMNH	Whisker
<i>Dasyurus maculatus</i>	65685	1923	Authur River Sawmill, Tasmania	M	AMNH	Whisker
<i>Dasyurus maculatus</i>	65687	1923	Authur River Sawmill, Tasmania	M	AMNH	Whisker
<i>Dasyurus maculatus</i>	65688	1923	Arthur River, Tasmania	M	AMNH	Bone
<i>Dasyurus maculatus</i>	QVM:1940:1:0164	1940	George Town, Tasmania	U	QVM	Bone
<i>Dasyurus maculatus</i>	A3492	1951	Tarrleah, Tasmania	U	TMAG	Bone
<i>Dasyurus maculatus</i>	RCSOMA 370.42	1951	Wynard, Tasmania	U	RCS	Bone

APPENDIX

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus maculatus</i>	A22	1954	Central Highlands, Tarraleah, Tasmanian Southern Ranges	F	TMAG	Whisker
<i>Dasyurus maculatus</i>	A553	1957	East Tamar Highway	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A21	1960	Buckland, Glamorgan-Spring Bay, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	QVM:1963:1:0018	1963	Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1963:1:0019	1963	Liffy Road, North Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1963:1:0232	1963	Liffey Falls, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	SMF 15507	1963	Bell Bay-Lannceston, Tasmania	M	FSS	Whisker
<i>Dasyurus maculatus</i>	A701	1965	West Hobart, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A903	1974	Zeehan, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A906	1974	Zeehan, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A910	1974	Crotty, Tasmania	U	TMAG	Bone
<i>Dasyurus maculatus</i>	QVM:1975:1:0039	1974	North Tasmania	U	QVM	Bone
<i>Dasyurus maculatus</i>	QVM:1978:1:0301	1978	Wyena, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1978:1:0545	1978	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Dasyurus maculatus</i>	A1259	1979	Buckland, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A1330	1979	Triabunna, Tasmania	U	TMAG	Bone
<i>Dasyurus maculatus</i>	QVM:1982:1:0050	1982	Northern Tasmania	U	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1984:1:0226	1984	Launceston, Underwood, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1984:1:0245	1984	Patersonia, Tasmania	M	QVM	Hair

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus maculatus</i>	QVM:1985:1:0053	1985	Underwood, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1987:1:0087	1987	Hollybank Forest Reserve, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1987:1:0109	1987	Mount Direction, Tasmania	F	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1988:1:0024	1987	Bridpost, Tasmania	F	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1988:1:0013	1988	West Tamar, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1988:1:0044	1988	Ben Lomond, Launceston, Tasmania	M	QVM	Hair
<i>Dasyurus maculatus</i>	QVM:1994:1:0010	1991	Launceston, Tasmania	M	QVM	Bone
<i>Dasyurus maculatus</i>	QVM:1995:1:0003	1991	Waratah/Wynyard, Tasmania	M	QVM	Bone
<i>Dasyurus maculatus</i>	QVM:1993:1:0217	1992	Northern Midlands, Evandale, Tasmania	M	QVM	Bone
<i>Dasyurus maculatus</i>	QVM:1994:1:0004	1992	Kentish, Tasmanian Northern slopes, Sheffield, Tasmania	M	QVM	Bone
<i>Dasyurus maculatus</i>	A1532	1993	Cardigan Flats, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A1540	1994	Hillwood, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	QVM:1996:1:0002	1994	West Tamar, Tasmania	M	QVM	Bone
<i>Dasyurus maculatus</i>	QVM:1996:1:0003	1995	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Dasyurus maculatus</i>	160039	1996	Queenstown, Tasmania	M	FMNH	Bone
<i>Dasyurus maculatus</i>	SQ04	2000	Mole Creek Hwy Near Lobster Falls, Tasmania	M	QVM	Whisker
<i>Dasyurus maculatus</i>	SQ05	2000	Mole Creek Hwy Near Lobster Falls, Tasmania	M	QVM	Whisker

APPENDIX

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus maculatus</i>	SQ06	2000	A1 Epping Forest, Tasmania	M	QVM	Whisker
<i>Dasyurus maculatus</i>	SQ03	2001	Mole creek, Tasmania	M	QVM	Whisker
<i>Dasyurus maculatus</i>	A2489	2004	Oatlands, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	SQ02	2004	A2 Hwy, , Tasmania	M	QVM	Whisker
<i>Dasyurus maculatus</i>	A2490	2006	Huon Valley, Castle Forbes Bay, Tamanian Southern Ranges	M	TMAG	Whisker
<i>Dasyurus maculatus</i>	A2491	2006	Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A2659	2008	Berwent valley, Tasmania	U	TMAG	Whisker
<i>Dasyurus maculatus</i>	A2660	2008	Bothwell, Tasmania	U	TMAG	Whisker
<i>Dasyurus hallucatus</i>	A6 11/2	1869	Tasmania	F	CZM	Bone
<i>Dasyurus hallucatus</i>	1921 No ID "native cat" Gunns Estate	1921	Gunns Estate, Launceston, Tasmania	U	QVM	Bone
<i>Dasyurus hallucatus</i>	65344	1922	Hobart, Tasmania	U	AMNH	Bone
<i>Dasyurus hallucatus</i>	65689	1922	Hobart, Tasmania	F	AMNH	Bone
<i>Dasyurus hallucatus</i>	65690	1922	Huon, Tasmania	M	AMNH	Bone
<i>Dasyurus hallucatus</i>	65693	1922	Huon, Tasmania	M	AMNH	Bone
<i>Dasyurus hallucatus</i>	65694	1922	Huon, Tasmania	M	AMNH	Bone
<i>Dasyurus hallucatus</i>	QVM:1976:1:0106	1955	Lilydale, Tasmania	M	QVM	Bone
<i>Dasyurus hallucatus</i>	A17	1956	Mount Wellington, Tasmania	U	TMAG	Whisker
<i>Dasyurus hallucatus</i>	A16	1959	Ridgeway, Tasmania	U	TMAG	Whisker
<i>Dasyurus hallucatus</i>	A11	1960	Westbury, Tasmania	U	TMAG	Whisker

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus viverrinus</i>	A399	1962	Sandfly, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A484	1963	Collinsvale, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A492	1963	Westbury, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	QVM:1964:1:0155	1964	Gladstone, Tasmania	F	QVM	Hair
<i>Dasyurus viverrinus</i>	QVM:1964:1:0164	1964	Gladstone, Tasmania	F	QVM	Hair
<i>Dasyurus viverrinus</i>	QVM:1964:1:0169	1964	Gladstone, Tasmania	F	QVM	Hair
<i>Dasyurus viverrinus</i>	QVM:1964:1:0175	1964	Gladstone, Tasmania	M	QVM	Hair
<i>Dasyurus viverrinus</i>	A905	1973	Campbell town, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A916	1974	Cygnets, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A1370	1982	Brightwater Road, Howden, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A1383	1983	Hamilton, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A1372	1984	Molesworth, Derwent Valley, Tasmania	U	TMAG	Bone
<i>Dasyurus viverrinus</i>	A1372	1984	Molesworth, Derwent Valley, South East TAS	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	QVM:1985:1:0032	1985	Launceston, Tasmania	M	QVM	Hair
<i>Dasyurus viverrinus</i>	A1517	1986	Lake St Clair, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	A1519	1986	Buckland, Glamorgan-Spring Bay, Tasmania	M	TMAG	Whisker
<i>Dasyurus viverrinus</i>	QVM:1986:1:0029	1986	Underwood, Tasmania	F	QVM	Hair
<i>Dasyurus viverrinus</i>	QVM:1986:1:0032	1986	Rocherlea, Tasmania	M	QVM	Hair
<i>Dasyurus viverrinus</i>	QVM:1988:1:0029	1988	Rocherlea, Tasmania	M	QVM	Hair
<i>Dasyurus viverrinus</i>	160040	1991	Brown's Mt Road, Capania, Tasmania	M	FMNH	Bone

APPENDIX

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus viverrinus</i>	QVM:1994:1:0011	1991	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	M	QVM	Bone
<i>Dasyurus viverrinus</i>	A1516	1993	Petchey's Bay, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	QVM:1993:1:0006	1993	Launceston, Ravenswood, Tasmanian Northern Midlands	U	QVM	Bone
<i>Dasyurus viverrinus</i>	QVM:1994:1:0025	1993	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	M	QVM	Bone
<i>Dasyurus viverrinus</i>	QVM:1994:1:0025	1993	Kentish, Tasmanian central highlands, Cradle Mountain, Tasmania	M	QVM	Bone
<i>Dasyurus viverrinus</i>	QVM:2006:1:0046	1994	Meander Valley, Tasmanian Northern Midlands	F	QVM	Bone
<i>Dasyurus viverrinus</i>	A1530	1995	Hobart, Tasmania	U	TMAG	Whisker
<i>Dasyurus viverrinus</i>	RK14	2011	Bruny Island, Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK15	2011	South Hobart, Tasmania	F	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK16	2011	Allen's Rivulet, , Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK18	2011	Kettering, Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK19	2012	Cradoc, Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK20	2012	Cradoc, Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK25	2012	Brwhhy Island, Tasmania	F	UTAS	Whisker

Appendix A1 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus viverrinus</i>	RK27	2012	Mountain River, Tasmania	M	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK28	2012	Crabtree, Tasmania	F	UTAS	Whisker
<i>Dasyurus viverrinus</i>	RK29	2012	Tasmania	M	UTAS	Whisker

* Institution Abbreviations: AMNH (American Museum of Natural History), BM (Booth Museum), CZM (Cambridge Zoological Museum), FMNH (Field Museum of Natural History), FSS (Forschungsinstitut and Schaumuseum Senckenberg), GNMH (Gothenburg Natural History Museum), HM (Hunterian Museum), LCM (Leeds City Museum), MV (Museum Victoria), NHM (National History Museum), NMA (National Museum of Australia), NZM (Natural Zoological museum), QVM (Queen Victoria Museum and Art Gallery), RCS (Royal College of Surgeons of England), UTAS (University of Tasmania), WML (World Museum Liverpool). Tissues collected during 2011 fieldwork are listed under 'fieldwork'.

APPENDIX

Appendix A2 List of specimens representing potential prey species of the thylacine.

Skin was analysed for all prey specimens. The list includes specimen records including sex (male=M; female=F, unknown=U), year of death/acquisition and locality. All specimens were held at the Queen Victoria Museum and Art Gallery.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution*	Tissue type
<i>Dasyurus maculatus</i>	SMF 15507	1963	Bell Bay-Lannceston, Tasmania	M	FSS	Skin
<i>Isoodon obesulus</i>	QVM:1964:1:0010	1964	St Patericks river, Tasmania	M	QVM	Skin
<i>Macropus giganteus</i>	NA	1931	Tasmania	U	QVM	Skin
<i>Macropus rufogriseus</i>	QVM:1959:1:0039	1946	Avoca, Tasmanian Northern Midlands	F	QVM	Skin
<i>Macropus rufogriseus</i>	QVM:1964:1:0120	1964	Leana, Gladston, Tasmania	F	QVM	Skin
<i>Ornithorhynchus anatinus</i>	NA	1940	Tasmania	U	QVM	Skin
<i>Ornithorhynchus anatinus</i>	QVM:1948:1:0011	1948	Mole creek, Tasmania	M	QVM	Skin
<i>Ornithorhynchus anatinus</i>	QVM1946:1:0013	1946	Deloraine, Tasmania	F	QVM	Skin
<i>Perameles gunnii</i>	QVM:1945:1:0080	1938	Launceston, N. Tasmania	M	QVM	Skin
<i>Potorous tridactylus</i>	NA	1939	Myrtle Bank, Tasmania	M	QVM	Skin
<i>Potorous tridactylus</i>	QVM:1939:1:0100	1939	Sheffield, N.W. Tasmania	U	QVM	Skin
<i>Potorous tridactylus</i>	QVM:1993:1:0052	1908	Tasmania	U	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM:1939:1:0096	1939	Sheffield, N.W. Tasmania	U	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM:1939:1:0097	1939	Sheffield, N.W. Tasmania	U	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM:1940:1:0193	1940	Mt. Arthur, Tasmania	U	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM:1946:1:0088	1946	Deloraine, Tasmania	F	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM:1993:1:0073	1917	Tasmania	U	QVM	Skin
<i>Pseudocheirus peregrinus</i>	QVM1946:1:0092	1946	Westbury, Tasmania	M	QVM	Skin

Appendix A2 continued.

Scientific name	Specimen number	Year of death	Locality/Source	Sex	Institution *	Tissue type
<i>Sarcophilus harrisii</i>	QVM:1941:1:0179	1941	Launceston, Underwood, Tasmania	U	QVM	Skin
<i>Sarcophilus harrisii</i>	QVM:OLD:1:2017	1903	Launceston - Pt B, Tasmanian Northern Midlands, Doctors Creek, N. Tasmania	M	QVM	Skin
<i>Sarcophilus harrisii</i>	SMF 16200	1852	Tasmania	U	FSS	Skin
<i>Tachyglossus aculeatus</i>	QVM:1946:1:0014	1946	Tasmania	F	QVM	Skin
<i>Thylogale billardierii</i>	QVM:1964:1:0121	1964	Leana, Gladston, Tasmania	M	QVM	Skin
<i>Thylogale billardierii</i>	QVM:1964:1:0123	1964	Leana, Gladston, Tasmania	M	QVM	Skin
<i>Thylogale billardierii</i>	QVM:1964:1:0196	1964	Leana, Gladston, Tasmania	M	QVM	Skin
<i>Trichosurus vulpecula</i>	QVM:1963:1:0282	1959	Gladstone, Tasmania	F	QVM	Skin
<i>Vombatus ursinus</i>	2191	1905	Tasmania	M	QVM	Skin

APPENDIX

Appendix A3 Diet and body mass of potential prey species of the thylacine included in this study. Average body mass are given for males (M) and females (F) where available.

Species	Scientific name	Diet category	Diet description	Adult body mass range; average (kg)*	Reference
Common brushtail possum	<i>Trichosurus vulpecula</i>	Browser	Primarily <i>Eucalyptus</i> and <i>Acacia</i> foliage, flowers, shoots, fruits and seeds.	1.2-4.5	Owen and Thomson (1965); Freeland and Winter (1975); Cronin (1991)
Common ringtail Possum	<i>Pseudocheirus peregrinus</i>	Browser	Primarily <i>Eucalyptus</i> leaves but will also eat foliage, flowers and fruits from other tree and shrub species.	0.6-1.1	Freeland and Winter (1975); Fitzgerald (1984); Kerle (1984); Cronin (1991)
Common wombat	<i>Vombatus ursinus</i>	Grazer/ Browser	Primarily native grasses but shrubs, roots, sedges, bark, herbs and moss are also eaten	20.0-40.0; 26	Cronin (1991); Rishworth <i>et al.</i> (1995); Triggs, B. (1996); Skerratt <i>et al.</i> (2004); Evans <i>et al.</i> (2006)
Eastern Barred Bandicoot	<i>Perameles gunnii</i>	Insectivore	Crickets, beetles and earthworms	0.5-1.5	Cronin (1991); Mallick <i>et al.</i> (2000)
Forester kangaroo	<i>Macropus giganteus</i>	Grazer	Variety of grasses	25.0-66.0; M=66, F=37	Strahan (1995); Menkhorst (2001)
Platypus	<i>Ornithorhynchus anatinus</i>	Insectivore	Annelid worms, insect larvae, freshwater shrimps, tadpoles, water bugs, beetles and yabbies	0.7-2.6	Grant (1982); Cronin (1991); Grant and Temple-Smith (1998)
Red-necked wallaby	<i>Macropus rufogriseus</i>	Grazer	Grass and herbs	11.0-27.0; M=27, F=16	Cronin (1991); Watts (1993); Strahan (1995); Menkhorst (2001)
Short-beaked echidna	<i>Tachyglossus aculeatus</i>	Insectivore	Ants and termites	2.0-7.0	Abensperg-Traun (1991); Cronin (1991); Menkhorst (2001) Nicol and Andersen (2007)
Southern brown bandicoot	<i>Isodon obesulus</i>	Insectivore	Insects and their larvae, underground fungi, worms, lizards and berries	0.4-1.6; M=3.5, F=0.7	Cronin (1991); Quin (1988); Menkhorst (2001)
Spotted-tailed quoll	<i>Dasyurus maculatus</i>	Carnivore	Rats, gliding possums, wallabies, reptiles and insects, birds and eggs	1.8-7.0; M=3.5, F=1.8	Cronin (1991); Jones (1995); Menkhorst (2001)
Tasmanian devil	<i>Sarcophilus harrisii</i>	Carnivore	Wallabies, small mammals and birds	6.0-11.0; M=8.7, F=6.1	Guiler (1978); Cronin (1991); Jones (1995)
Tasmanian pademelon	<i>Thylogale billardierii</i>	Grazer	Prefers short green grass, but also eats herbs and green shoots	2.4-12.0; M=9, F=5.8	Cronin (1991); Belcher (1995); Strahan (1995); Menkhorst (2001)

*Average body mass for females (F) and males (M), where available. The average species body mass is listed for common wombats as no information was available on each sex.

Appendix A4 Sample size, mean $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ($\pm\text{SE}$), and C/N ratios (mean $\pm$ SD) of museum-preserved skin of potential prey species. All specimens were collected between 1835 and 1964.

Common name	Scientific name	<i>n</i>	$\delta^{13}\text{C}$ ‰	$\delta^{15}\text{N}$ ‰	C/N ratio
Common brushtail possum	<i>Trichosurus vulpecula</i>	1	-19.9	6.2	3.5
Common ringtail possum	<i>Pseudocheirus peregrinus</i>	6	-20.9 ± 1.5	6.0 ± 1.9	3.4 ± 0.1
Common wombat	<i>Vombatus ursinus</i>	1	-22.9	8.7	3.3
Eastern barred bandicoot	<i>Perameles gunnii</i>	1	-20.5	11.6	3.3
Forester kangaroo	<i>Macropus giganteus</i>	1	-19.3	7.0	3.5
Short-beaked echidna	<i>Tachyglossus aculeatus</i>	1	-19.7	11.4	3.4
Long-nosed potoroo	<i>Potorous tridactylus</i>	3	-20.5 ± 0.4	14.5 ± 0.6	3.4 ± 0.2
Platypus	<i>Ornithorhynchus anatinus</i>	3	-21.2 ± 0.5	14.0 ± 1.9	3.3 ± 0.1
Red-necked wallaby	<i>Macropus rufogriseus</i>	2	-21.7 ± 0.3	7.7 ± 0.2	3.4 ± 0.0
Southern brown bandicoot	<i>Isodon obesulus</i>	1	-19.5	10.8	3.2
Spotted-tailed quoll	<i>Dasyurus maculatus</i>	1	-19.2	13.3	3.4
Tasmanian devil	<i>Sarcophilus harrisii</i>	3	-20.7 ± 0.5	13.6 ± 1.2	3.5 ± 0.2
Tasmanian pademelon	<i>Thylogale billardierii</i>	3	-22.2 ± 0.5	7.8 ± 0.8	3.5 ± 0.1

APPENDIX B

Tissue enrichment of carbon and nitrogen stable isotopes in marsupial carnivore tissues

Tissues were sampled from 10 Tasmanian devils, including two road kill collected in February 2011, and eight frozen specimens acquired by the Queen Victoria Museum and Art Gallery and the Tasmanian Museum and Art Gallery between 2000 and 2012. The manner of death for museum derived animals was largely unknown, but we were able to attribute the deaths of most individuals to vehicle collisions based on visual inspection of the bodies. Protocols for collection of frozen tissues varied from that described for historic specimens. A toe was removed from each fresh or frozen specimens and the bone defleshed by rinsing in distilled water for ~3 weeks. Skin and hair were removed from the upper thigh of each animal, and the longest posterior vibrissa was plucked. For skin samples, the subcutaneous tissue (fat tissue layer) underneath the dermis was removed by scraping with a scalpel. Refer to section 4.3.3 for further information on protocols used for sample preparation and analysis.

Raw data for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and C/N ratios are listed in Appendix B1. The C/N ratio is a simple measure of chemical quality in preserved samples. Bone collagen that has a C/N ratio outside the range of 2.9 to 3.6 are considered to have an excellent level of preservation, while ratios outside this range can give anomalous $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values (DeNiro 1985). For Tasmanian devil tissue comparisons, three bone collagen samples had a C/N ratio slightly above this range, with the highest value at 4.22‰ (Appendix B1), and were all included in subsequent analysis.

Appendix B1 Raw stable isotope data for bone collagen, skin collagen, vibrissa and hair samples collected opportunistically from deceased wild adult Tasmanian devils (n=10). Specimen sex (M = male, F = female, U = unknown), stable isotope ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) and C/N ratios are provided.

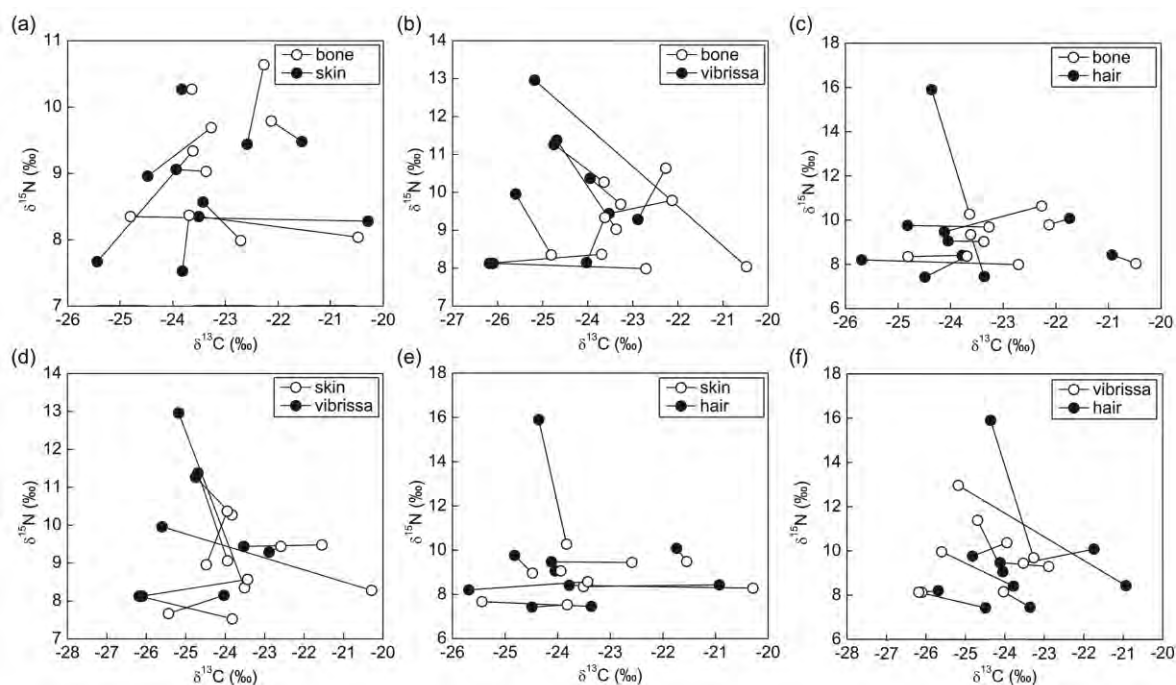
Subject	Sex	Bone			Skin			Vibrissa			Hair	
		$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	C/N ratio	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	C/N ratio	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	C/N ratio	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
1	F	-20.5	8.0	3.7	-23.5	8.4	3.2	-25.2	13.0	3.4	-20.9	8.4
2	M	-24.8	8.4	4.2	-20.3	8.3	3.3	-25.6	10.0	3.4	-23.8	8.4
3	M	-23.4	9.0	3.6	-23.9	9.1	3.5	-24.7	11.4	3.4	-24.1	9.1
4	U	-22.1	9.8	3.9	-21.6	9.5	3.1	-23.5	9.4	3.6	-21.7	10.1
5	F	-23.6	10.3	3.6	-23.8	10.3	3.3	-24.8	11.3	3.6	-24.4	15.9
6	F	-22.3	10.6	3.8	-22.6	9.4	3.3	-22.9	9.3	3.6	-24.1	9.5
7	U	-23.3	9.7	3.4	-24.5	9.0	4.1	-24.0	10.4	3.4	-24.8	9.8
8	U	-23.6	9.3	3.2	-25.4	7.7	5.1	-24.0	8.2	3.4	-23.4	7.5
9	F	-22.7	8.0	3.3	-23.4	8.6	3.2	-26.1	8.1	3.6	-25.7	8.2
10	M	-23.7	8.4	3.3	-23.8	7.5	3.4	-26.2	8.1	3.6	-24.5	7.4

Note: All samples had C/N ratios within the range 3.1-5.1.

APPENDIX

We tested stable isotope data for normality using the Shapiro–Wilk statistic. A Wilcoxon matched-pairs signed-ranks test was used to compare $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values between tissues within each individual. As samples were collected around the same time, we did not account for the Suess effect prior to analysis. The isotopic spacing between paired samples of each tissue are displayed in Appendix B2 and outcomes for the Wilcoxon matched pairs test between each tissue type for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ are displayed in Appendix B3.

The interpretation of the Tasmanian devil tissue enrichment estimates are limited by our low sample size, however provide a general indication of isotopic differences between tissue types. Paired samples of skin and bone (see Appendix B2a) showed no difference in isotopic signatures ($P=0.20$ for $\delta^{13}\text{C}$ and $P=0.14$ for $\delta^{15}\text{N}$; Appendix B3), and conformed to expectations based on controlled feeding studies (Hare *et al.* 1991). The turnover rate of skin collagen usually reflects stable isotope values averaged over the previous 2-3 months of an animal's life (Tieszen *et al.* 1983), while bone collagen can represent diet of the lifetime of the individual (Stenhouse and Baxter 1979). Skin and the protein fraction of bone comprise primarily of Type 1 collagen (Epstein Jr and Munderloh 1978; Wheelless 2009), and therefore are expected to possess similar stable isotope values after controlling for diet. We found no significant difference in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ between bone and hair ($P = 0.07$ and 0.72 for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, respectively; Appendix B3). Hair and vibrissae have a similar biochemical composition (Lehninger 1973), being composed of keratin, and showed no difference in stable isotope values ($P = 0.09$ and 0.28 for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, respectively; Appendix B3). Differences in $\delta^{13}\text{C}$ values of bone and vibrissa ($P=0.01$, Appendix B3) may be attributed to these tissues representing diet over different time period; the vibrissae tip of Tasmanian devils reflect the isotopic signature of the animal from a 'snapshot' in time from ~9 months prior to sampling (unpublished data), whereas bone collagen undergoes constant remodelling over the lifetime of an animal (Finucane 2007).



Appendix B2 Stable carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) isotope ratios of paired (a) bone and skin, (b) bone and whisker, (c) bone and hair, (d) skin and whisker, (e) skin and hair, and (f) vibrissa and hair from ten wild adult Tasmanian devils. Lines are drawn between tissue stable isotope values from the same individual.

Appendix B3 Wilcoxon matched pairs signed rank test comparing inter-tissue isotopic spacing for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ from wild Tasmanian devils ($n=10$). Valid N excludes paired tests that have the same stable isotope values.

	$\delta^{13}\text{C}$					$\delta^{15}\text{N}$				
	Mean $\pm$ SD	Valid N	T	Z	P-value	Mean $\pm$ SD	Valid N	T	Z	P-value
$\Delta_{(\text{bone-skin})}$	0.3 $\pm$ 2.0	10	15.00	1.27	0.20	0.4 $\pm$ 0.7	9	10.00	1.48	0.14
$\Delta_{(\text{bone-vibrissa})}$	1.7 $\pm$ 1.4	10	0.00	2.80	0.01*	-0.8 $\pm$ 1.9	10	18.00	0.97	0.33
$\Delta_{(\text{bone-hair})}$	0.7 $\pm$ 1.3	10	10.00	1.78	0.07	0.3 $\pm$ 0.8	10	24.00	0.36	0.72
$\Delta_{(\text{skin-vibrissa})}$	1.4 $\pm$ 1.9	10	7.00	2.09	0.04*	-1.2 $\pm$ 1.5	10	6.00	2.20	0.03*
$\Delta_{(\text{skin-hair})}$	0.4 $\pm$ 1.9	10	16.00	1.17	0.24	-0.1 $\pm$ 0.4	9	14.00	1.01	0.31
$\Delta_{(\text{whisker-hair})}$	1.0 $\pm$ 1.6	10	11.00	1.68	0.09	-1.1 $\pm$ 1.6	10	17.00	1.07	0.28

Note: Significance levels (*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns, $P > 0.05$) are given.

APPENDIX C

Terrestrial carnivore tissue to diet enrichment

Appendix C1 Literature search for specimen-specific plasma fractionation factors in mammals, sorted by environment, infraclass and trophic level. The animal section describes the species common and scientific name, environment (T = terrestrial, M = marine), Infraclass (M = marsupial, P = placental) and tissue. The discrimination factor section describes the discrimination factors ($\Delta^{13}\text{C}$ and $\Delta^{15}\text{N}$) calculated in each case. Adapted from Caut *et al.* (2009).

Animal	Diet						Discriminatio on factor		Reference				
	Common name	Scientific name	Env	Infra- class	Tissue	Tropic level	Fixed diet	$\delta^{13}\text{C}$ (‰)		$\delta^{15}\text{N}$ (‰)	Tim e (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)
Long-nosed bandicoot		<i>Perameles nasuta</i>	T	M	Plasma	Carnivor e	High protein corn	-18.6	3.8	28- 35	3.20	2.10	Klaassen <i>et al.</i> (2005)
Long-nosed bandicoot		<i>Perameles nasuta</i>	T	M	Plasma	Carnivor e	High protein sweet potato	-24.8	3.3	28- 35	1.40	2.80	Klaassen <i>et al.</i> (2005)
Long-nosed bandicoot		<i>Perameles nasuta</i>	T	M	Plasma	Carnivor e	Low protein corn	-13.9	1.1	28- 35	-1.80	3.80	Klaassen <i>et al.</i> (2005)
Long-nosed bandicoot		<i>Perameles nasuta</i>	T	M	Plasma	Carnivor e	Low protein sweet potato	-25.8	3.8	28- 35	4.60	2.40	Klaassen <i>et al.</i> (2005)
American black bear		<i>Ursus americanus</i>	T	P	Plasma	Omnivor e	Salmon	-20.4	11.2	40	1.40	2.30	Hilderbrand <i>et al.</i> (1996)
American black bear		<i>Ursus americanus</i>	T	P	Plasma	Omnivor e	Mule deer	-25.5	7.0	40	3.90	3.50	Hilderbrand <i>et al.</i> (1996)

Appendix C1 continued.

Animal	Diet						Discrimination factor						
	Common name	Scientific name	Env	Infra-class	Tissue	Tropic level	Fixed diet	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Tim e (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)	Reference
American black bear		<i>Ursus americanus</i>	T	P	Plasma	Omnivore	Apples	-24.1	2.8	40	3.3	3.8	Hilderbrand <i>et al.</i> (1996)
American black bear		<i>Ursus americanus</i>	T	P	Plasma	Omnivore	65% herring, 35% vegetation	-21.0	9.5	40	2.7	4.5	Hilderbrand <i>et al.</i> (1996)
American black bear		<i>Ursus americanus</i>											
American black bear		<i>Ursus americanus</i>	T	P	Plasma	Omnivore	35% herring, 65% vegetation	-21.5	6.3	40	1.7	4.0	Hilderbrand <i>et al.</i> (1996)
Grizzly bear		<i>Ursus arctos horribilis</i>	T	P	Plasma	Omnivore	Chinook salmon	-20.1	11.2	21	0.1	3.8	Felicetti <i>et al.</i> (2003)
Grizzly bear		<i>Ursus arctos horribilis</i>	T	P	Plasma	Omnivore	Apples	-26.7	0.7	21	4.5	6.3	Felicetti <i>et al.</i> (2003)
Grizzly bear		<i>Ursus arctos horribilis</i>	T	P	Plasma	Omnivore	Commercial bear chow	-19.0	3.8	21	0.4	6.2	Felicetti <i>et al.</i> (2003)
Grizzly bear		<i>Ursus arctos horribilis</i>	T	P	Plasma	Omnivore	Pelleted chow 15.4% protein	-22.2	2.3	21	1.0	6.2	Felicetti <i>et al.</i> (2003)
Grizzly bear		<i>Ursus arctos horribilis</i>											
Grizzly bear		<i>Ursus arctos horribilis</i>	T	P	Plasma	Omnivore	Pelleted chow 3.3% protein	-22.8	3.6	21	2.4	5.4	Felicetti <i>et al.</i> (2003)
Rat		<i>Rattus sp.</i>	T	P	Plasma	Omnivore	Commercial diet	-21.1	5.1	?	0.7	3.9	Yoneyama <i>et al.</i> (1983)

Appendix C1 continued.

Animal	Diet							Discrimination factor		Reference			
	Common name	Scientific name	Env	Infra-class	Tissue	Trophic level	Fixed diet	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)		Tim e (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)
Red fox		<i>Vulpes vulpes</i>	T	P	Plasma	Omnivore	Commercial pellet food	-18.7	4.90	210	0.60	4.20	Roth and Hobson (2000)
Gray seals		<i>Halichoerus grypus</i>	M	P	Plasma	Carnivore	Herring	-19.5	13.30	120-300	0.50	2.90	Lesage et al. (2002)
		<i>Halichoerus grypus</i>	M	P	Plasma	Carnivore	Herring/Capelin	-19.7	14.10	120-300	0.80	3.30	Lesage et al. (2002)
Harbor seals		<i>Phoca vitulina</i>	M	P	Plasma	Carnivore	Herring	-19.6	13.30	120-300	1.10	3.20	Lesage et al. (2002)
Harbor seals		<i>Phoca vitulina</i>	M	P	Plasma	Carnivore	Herring	-20.0	12.90	120-300	0.70	2.70	Lesage et al. (2002)
Harbor seals		<i>Phoca vitulina</i>	M	P	Plasma	Carnivore	Capelin	-19.4	14.40	120-300	0.50	3.00	Lesage et al. (2002)
Harp seal		<i>Phoca groenlandica</i>	M	P	Plasma	Carnivore	Herring	-19.9	13.90	120-300	1.00	3.60	Lesage et al. (2002)
Northern fur seal		<i>Callorhinus ursinus</i>	M	P	Plasma	Carnivore	Fish (herring & capelin)	-18.9	11.60	180	1.00	5.20	Kurle (2002)
Arctic fox		<i>Vulpes lagopus</i>	T	P	Plasma	Carnivore	Mix of marine and terrestrial	-	-	70	0.47	3.57	Lecomte et al. (2011)
Arctic fox		<i>Vulpes lagopus</i>	T	P	Plasma	Carnivore	Marine	-	-	70	1.15	2.80	Lecomte et al. (2011)
Arctic fox		<i>Vulpes lagopus</i>	T	P	Plasma	Carnivore	Terrestrial	-	-	70	0.79	3.03	Lecomte et al. (2011)

Appendix C2 Literature search for specimen-specific blood and red blood cell fractionation factors in mammals, sorted by environment, infraclass and trophic level. The animal section describes the species common and scientific name, environment (T = terrestrial, M = marine), Infraclass (M = marsupial, P = placental) and tissue (red blood cells, blood). The discrimination factor section describes the discrimination factors ($\Delta\delta^{13}\text{C}$ and $\Delta\delta^{15}\text{N}$) calculated in each case. Adapted from Caut *et al.* (2009).

Animal	Diet						Discrimination factor						
	Common name	Scientific name	Env	Infraclass	Tissue	Trophic level	Fixed diet	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Time (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)	Reference
Long-nosed bandicoot		<i>Perameles nasuta</i>	T	M	Red blood cells	Carnivore	High protein sweet potato	-24.80	3.30	28-35	-0.20	2.10	Klaassen <i>et al.</i> (2005)
Mice		<i>Mus sp.</i>	T	P	Blood	Omnivore	Diet 2018	-20.50	2.80	172	1.40	2.70	Arneson <i>et al.</i> (2005)
Mice		<i>Mus sp.</i>	T	P	Blood	Omnivore	Diet 1	-26.50	6.10	172	1.40	3.20	Arneson <i>et al.</i> (2005)
Mice		<i>Mus sp.</i>	T	P	Blood	Omnivore	Diet 2	-14.80	9.10	172	-1.90	2.90	Arneson <i>et al.</i> (2005)
Rat		<i>Rattus sp.</i>	T	P	Red blood cells	Omnivore	Commercial diet	-21.10	5.10	?	0.20	2.00	Yoneyama <i>et al.</i> (1983)
Red fox		<i>Vulpes vulpes</i>	T	P	Red blood cells	Omnivore	Commercial pellet	-18.70	4.90	210	0.70	2.6	Roth and Hobson (2000)
Pallas's long-tongued bat		<i>Glossophaga soricina</i>	T	P	Blood	Herbivore	Soy diet	-22.90	2.50	540	0.10	3.50	Mirón <i>et al.</i> (2006)

Appendix C2 continued.

Animal		Diet					Discrimination factor					
Common name	Scientific name	Env	Infra-class	Tissue	Tropic level	Fixed diet	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Time (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)	Reference
Palla's long-tongued bat	<i>Glossophaga soricina</i>	T	P	Blood	Herbivor _e	Milk diet	-20.60	3.80	540	1.30	4.90	Mirón <i>et al.</i> (2006)
Palla's long-tongued bat	<i>Glossophaga soricina</i>	T	P	Blood	Herbivor _e	Amaranth diet	-18.90	4.00	105	2.00	4.40	Mirón <i>et al.</i> (2006)
(Harp, Harbour, Ringed) seal	<i>Pagophilus groenlandicus</i> , <i>Phoca vitulina</i> , <i>P. hispida</i>	M	P	Red blood cells	Carnivor _e	Herring	-20.30	13.00	730	1.70	1.70	Hobson <i>et al.</i> (1996)
Gray seals	<i>Halichoerus grypus</i>	M	P	Red blood cells	Carnivor _e	Herring	-19.50	13.30	120-300	1.20	1.60	Lesage <i>et al.</i> (2002)
Gray seals	<i>Halichoerus grypus</i>	M	P	Red blood cells	Carnivor _e	Herring/Capelin	-19.70	14.10	120-300	1.70	1.70	Lesage <i>et al.</i> (2002)
Harbor seals	<i>Phoca vitulina</i>	M	P	Red blood cells	Carnivor _e	Herring	-19.60	13.30	120-300	1.50	1.70	Lesage <i>et al.</i> (2002)
Harbor seals	<i>Phoca vitulina</i>	M	P	Red blood cells	Carnivor _e	Herring	-20.00	12.90	120-300	1.10	2.00	Lesage <i>et al.</i> (2002)
Harbor seals	<i>Phoca vitulina</i>	M	P	Red blood cells	Carnivor _e	Capelin	-19.40	14.40	120-300	1.40	1.40	Lesage <i>et al.</i> (2002)

APPENDIX

Appendix C2 continued.

Animal		Diet						Discrimination factor				
Common name	Scientific name	Env	Infra-class	Tissue	Tropic level	Fixed diet	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Time (d)	$\Delta^{13}\text{C}$ (‰)	$\Delta^{15}\text{N}$ (‰)	Reference
Harp seal	<i>Phoca groenlandica</i>	M	P	Red blood cells	Carnivore	Herring	-19.90	13.90	120-300	1.90	2.00	Lesage <i>et al.</i> (2002)
Northern fur seal	<i>Callorhinus ursinus</i>	M	P	Red blood cells	Carnivore	Fish (herring & capelin)	-18.90	11.60	180	0.20	3.90	Kurle (2002)
Arctic fox	<i>Vulpes lagopus</i>	T	P	Red blood cells	Carnivore	Mix of marine and terrestrial	-	-	70	0.49	2.56	Lecomte <i>et al.</i> (2011)
Arctic fox	<i>Vulpes agopus</i>	T	P	Red blood cells	Carnivore	Marine	-	-	70	0.24	0.38	Lecomte <i>et al.</i> (2011)
Arctic fox	<i>Vulpes lagopus</i>	T	P	Red blood cells	Carnivore	Terrestrial	-	-	70	0.56	1.58	Lecomte <i>et al.</i> (2011)

Appendix C3 Previously published isotopic fractionation between bone collagen and diet of terrestrial carnivorous mammal communities and species.

Predator common name	Predator scientific name	Prey	Site	$\Delta\delta^{13}\text{C}_{\text{collagen-diet}}$ (‰)		$\Delta\delta^{15}\text{N}_{\text{collagen-diet}}$ (‰)		Reference
				Mean	SD	Mean	SD	
Carnivorous mammals (cheetah, honey badger, domestic dog, bat-eared fox, genet, white-tipped mongoose, golden jackal, spotted hyena, lion and leopard)	<i>Acinonyx jubatus</i> ; <i>Millivora capensis</i> ; <i>Canis familiaris</i> ; <i>Otocyon megalotis</i> ; <i>Genetta sp.</i> ; <i>Ichneumon albicauda</i> ; <i>Canis aureau</i> ; <i>Crocuta crocuta</i> ; <i>Panthera leo</i> ; <i>Panthera leo</i> ; <i>Panthera pardus</i>	Herbivorous mammals	East Africa	-	-	4.80	-	Ambrose and DeNiro (1986)
Carnivorous mammals (leopard, tiger, lion, caracal, mountain lion, arctic fox, fisher, bobcat, coyote, striped hyaena, jackal, honey badger, fennec, grey fox)	<i>Leo pardus</i> ; <i>Leo tigris</i> ; <i>Leo leo</i> ; <i>Felis caracal</i> ; <i>Felis concolor</i> ; <i>Alopex lagopus</i> ; <i>Martes pennanti</i> ; <i>Lynx rufus</i> ; <i>Canis latrans</i> ; <i>Hyaena hyaena</i> ; <i>Canis sp.</i> ; <i>Mellivora capensis</i> ; <i>Fennecus zerda</i> ; <i>Urocyon cinereoargenteus</i>	Herbivorous mammals	Kenya	1.20	-	4.00	-	Schoeninger (1985)
Coyote	<i>Canis latrans</i>	Deer	Ontario	-	-	2.80	0.90	Schwarz (1991)

Appendix C3 continued.

Predator common name	Predator scientific name	Prey	Site	$\Delta\delta^{13}\text{C}_{\text{collagen-diet}}$ (‰)		$\Delta\delta^{15}\text{N}_{\text{collagen-diet}}$ (‰)		Reference
				Mean	SD	Mean	SD	
Lynx	<i>Lynx lynx</i>	Hare, Antelope	South Africa	2.00	-	-	-	Van der Merwe (1989)
Lynx	<i>Lynx lynx</i>	Roe deer	Bialoweiza forrest	1.20	-	4.20	-	Bocherens and Drucker (2003)
Lynx	<i>Lynx lynx</i>	1/3 all ungulates	Bialoweiza forrest	0.40	-	4.10	-	Bocherens and Drucker (2003)
Lynx	<i>Lynx lynx</i>	Ungulates	Bialoweiza forrest	1.10	-	4.00	-	Bocherens and Drucker (2003)
Wolf	<i>Canis lupus</i>	Red deer	Bialoweiza forrest	1.30	-	3.40	-	Bocherens and Drucker (2003)
Wolf	<i>Canis lupus</i>	1/3 all ungulates	Bialoweiza forrest	0.70	-	3.90	-	Bocherens and Drucker (2003)
Wolf	<i>Canis lupus</i>	Ungulates	Bialoweiza forrest	1.00	-	3.60	-	Bocherens and Drucker (2003)
Wolf	<i>Canis lupus</i>	Herbivorous mammals	Saint-Germain-la-Rivier	0.90	-	4.40	-	Drucker (2001)
Wolf	<i>Canis lupus</i>	Herbivorous mammals	Les Jamblands	1.30	-	5.50	-	Drucker (2001)
North American gray wolf	<i>Canis lupus</i> L.	Moose	Isle Royale	1.30	0.60	4.60	0.70	Fox-Dobbs <i>et al.</i> (2007)

Appendix C3 continued.

Predator common name	Predator scientific name	Prey	Site	$\Delta\delta^{13}\text{C}_{\text{collagen-diet}} (\text{‰})$		$\Delta\delta^{15}\text{N}_{\text{collagen-diet}} (\text{‰})$		Reference
				Mean	SD	Mean	SD	
Alexander Archipelago wolf	<i>Canis lupus ligon</i>	Caribo	Interior Alaska	1.20	-	2.40	-	Szepanski <i>et al.</i> (1999)
Wolf	<i>Canis lupus</i>	Deer	Ontario	-	-	2.70	-	Schwarcz (1991)
Mean*				1.13	0.60	3.89	0.80	

REFERENCES

- Abensperg-Traun, M. (1991) Survival strategies of the Echidna *Tachyglossus aculeatus* shaw 1792 (Monotremata: Tachyglossidae). *Biological Conservation*, **58**, 317-328.
- Alcover, J. A., Sans, A. and Palmer, M. (2002) The extent of extinctions of mammals on islands. *Journal of Biogeography*, **25**, 913-918.
- Allen, J. and Greenwood, J. (1988) Frequency-Dependent Selection by Predators. *Philosophical Transactions of the Royal Society of London. B, Biological Sciences*, **319**, 485-503.
- Ambrose, S. H., Buikstra, J. and Krueger, H. W. (2003) Status and gender differences in diet at Mound 72, Cahokia, revealed by isotopic analysis of bone. *Journal of Anthropological Archaeology*, **22**, 217-226.
- Andelt, W. F. (1985) Behavioral ecology of coyotes in south Texas. *Wildlife Monographs*, **94**, 1-45.
- Archer, M. (1976) The basicranial region of marsupicarnivores (Marsupialia), interrelationships of carnivorous marsupials, and affinities of the insectivorous marsupial peramelids. *Zoological Journal of the Linnean Society*, **59**, 217-322.
- Archer, M., Arena, R., Bassarova, M., Black, K., Brammall, J., Cooke, B., Creaser, P., Crosby, K., Gillespie, A., Godthelp, H., Gott, M., Hand, S. J., Kear, B., Krikmann, A., Mackness, B., Muirhead, J., Musser, A., Myers, T., Pledge, N., Wang, Y. and Wroe, S. (1999) The evolutionary history and diversity of Australian mammals. *Australian Mammalogy*, **21**, 1-45.
- Archer, M., Brammall, J., Field, J., Hand, S. J. and Hook, C. (2002) *The Evolution of Australia: 110 million years of change*. Australian Museum. Sydney.
- Archer, M., Derrick, A., Bassarova, M., Beck, R. M. D., Black, K., Walter, E. B., Brewer, P., Bernard, N. C., Crosby, K. and Gillespie, A. (2006) Current status of species-level representation in faunas from selected fossil localities in the

- Riversleigh World Heritage Area, northwestern Queensland. *Alcheringa: An Australasian Journal of Palaeontology*, **30**, 1-17.
- Archer, M., Hand, S. J. and Godthelp, H. (1994) Riversleigh. The Story of Animals in Ancient Rainforests of Inland Australia. *Reed Books, Sydney*, **1995**, 77-90.
- Archer, M., Godthelp, H., Hand, S.J., Attenborough, D. (2000) *Australia's lost world: Riversleigh, world heritage site*. Reed New Holland. Sydney.
- Archer, S., Schimel, D. S. and Holland, E. A. (1995) Mechanisms of shrubland expansion: land use, climate or CO₂? *Climatic Change*, **29**, 91-99.
- Arneson, L. S. and MacAvoy, S. E. (2005) Carbon, nitrogen, and sulfur diet-tissue discrimination in mouse tissues. *Canadian Journal of Zoology*, **83**, 989-995.
- Attard, M. R. G., Chamoli, U., Ferrara, T. L., Rogers, T. L. and Wroe, S. (2011) Skull mechanics and implications for feeding behaviour in a large marsupial carnivore guild: the thylacine, Tasmanian devil and spotted-tailed quoll. *Journal of Zoology*, **285**, 292-300.
- Animal Welfare Advisory Committee Australia (2003) Animal welfare standard for the hunting of wallabies in Tasmania. Nature Conservation Branch, Hobart.
- Australian Bureau of Statistics (ABS) (2008) Australian Historical Population Statistics, 2008. Cat. No. 3105.3100.3165.3001. ABS, Canberra.
- Bailey, C. (2001) *Tiger tales: Stories of the Tasmanian tiger*. HarperCollins. Sydney.
- Baillie, J. E. M., Hilton-Taylor, C. and Stuart, S. N., (2004) A global species assessment. 2004 IUCN Red List of Threatened Species. The IUCN Species Survival Commission, Cambridge.
- Barker, R. and Caughley, G. (1990) Distribution and abundance of kangaroos (Marsupialia: Macropodidae) at the time of European contact: Tasmania. *Australian Mammalogy*, **13**, 157-166.
- Bearhop, S., Furness, R. W., Hilton, G. M., Votier, S. C. and Waldron, S. (2003) A forensic approach to understanding diet and habitat use from stable isotope analysis of (avian) claw material. *Functional Ecology*, **17**, 270-275.

REFERENCES

- Becker, B. H., Newman, S. H., Inglis, S. and Beissinger, S. R. (2007) Diet-feather stable isotope ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) fractionation in Common murre and other seabirds. *The Condor*, **109**, 451-456.
- Beddard, F. E. (1891) 4. On the pouch and brain of the male thylacine. *Proceedings of the Zoological Society of London*, **59**, 138-148.
- Begon, M., Harper, J. L. and Townsend, C. R. (1990) *Ecology, Individuals, Population, Communities*. Blackwell, London.
- Belcher, C. (1995) Diet of the tiger quoll (*Dasyurus maculatus*). *Wildlife Research*, **22**, 341-357.
- Belcher, C., Nelson, J. and Darrant, J. (2007) Diet of the tiger quoll (*Dasyurus maculatus*) in south-eastern Australia. *Australian Journal of Zoology*, **55**, 117-122.
- Bell, P. R., Snively, E. and Shychoski, L. (2009) A comparison of the jaw mechanics in hadrosaurid and ceratopsid dinosaurs using finite element analysis. *The Anatomical Record: Advances in Integrative Anatomy and Evolutionary Biology*, **292**, 1338-1351.
- Belovsky, G. E., (1987) Extinction models and mammalian persistence. In: *Viable populations for conservation*: 35-57. M. E. Soulé (Ed.). Cambridge University Press, Cambridge.
- Ben-David, M., Flynn, R. W. and Schell, D. M. (1997) Annual and seasonal changes in diets of martens: evidence from stable isotope analysis. *Oecologia*, **111**, 280-291.
- Benazzi, S., Bookstein, F. L., Strait, D. S. and Weber, G. W. (2011) A new OH5 reconstruction with an assessment of its uncertainty. *Journal of Human Evolution*, **61**, 75-88.
- Berger, J., Stacey, P. B., Bellis, L. and Johnson, M. P. (2001a) A mammalian predator-prey imbalance: grizzly bear and wolf extinction affect avian neotropical migrants. *Ecological Applications*, **11**, 947-960.

- Berger, J., Swenson, J. E. and Persson, I. (2001b) Recolonizing carnivores and naive prey: conservation lessons from Pleistocene extinctions. *Science*, **291**, 1036-1039.
- Berger, K. M. and Gese, E. M. (2007) Does interference competition with wolves limit the distribution and abundance of coyotes? *Journal of Animal Ecology*, **76**, 1075-1085.
- Besl, P. J. and McKay, N. D. (1992) A method for registration of 3-D shapes. *IEEE Transactions on pattern analysis and machine intelligence*, **14**, 239-256.
- Binks, C. J. (1980) *Explorers of Western Tasmania*. Mary Fisher Bookshop. Launceston, Tasmania.
- Birchall, J., O'Connell, T. C., Heaton, T. H. E. and Hedges, R. E. M. (2005) Hydrogen isotope ratios in animal body protein reflect trophic level. *Journal of Animal Ecology*, **74**, 877-881.
- Blumstein, D. T. (2002) Moving to suburbia: ontogenetic and evolutionary consequences of life on predator-free islands. *Journal of Biogeography*, **29**, 685-692.
- Blumstein, D. T. and Daniel, J. C. (2003) Foraging behavior of three Tasmanian macropodid marsupials in response to present and historical predation threat. *Ecography*, **26**, 585-594.
- Blumstein, D. T. and Daniel, J. C. (2005) The loss of anti-predator behaviour following isolation on islands. *Proceedings of the Royal Society B: Biological Sciences*, **272**, 1663-1668.
- Bocherens, H. and Drucker, D. (2003) Trophic level isotopic enrichment of carbon and nitrogen in bone collagen: case studies from recent and ancient terrestrial ecosystems. *International Journal of Osteoarchaeology*, **13**, 46-53.
- Bodmer, R. E., Eisenberg, J. F. and Redford, K. H. (2002) Hunting and the likelihood of extinction of Amazonian mammals. *Conservation Biology*, **11**, 460-466.
- Bogliani, G., Sergio, F. and Tavecchia, G. (1999) Wood pigeons nesting in association with hobby falcons: advantages and choice rules. *Animal Behaviour*, **57**, 125-131.

REFERENCES

- Bolnick, D. I., Svanbaack, R., Fordyce, J. A., Yang, L. H., Davis, J. M., Hulsey, C. D. and Forister, M. L. (2003) The ecology of individuals: incidence and implications of individual specialization. *American Naturalist*, **161**, 1-28.
- Bourke, J., Wroe, S., Moreno, K., McHenry, C. and Clausen, P. (2008) Effects of gape and tooth position on bite force and skull stress in the dingo (*Canis lupus dingo*) using a 3-dimensional finite element approach. *PLoS ONE*, **3**, e2200.
- Bowman, D. (2001) Future eating and country keeping: what role has environmental history in the management of biodiversity? *Journal of Biogeography*, **28**, 549-564.
- Bradshaw, C. J. A. and Brook, B. W. (2005) Disease and the devil: density-dependent epidemiological processes explain historical population fluctuations in the Tasmanian devil. *Ecography*, **28**, 181-190.
- Braithwaite, R. W. and Begg, R. J., (1995) Northern quoll *Dasyurus hallucatus*. In: *The Mammals of Australia: National Photographic Index of Australian Wildlife*: 65-66. Reed Books, Sydney
- Brashares, J. S., Arcese, P. and Sam, M. K. (2001) Human demography and reserve size predict wildlife extinction in West Africa. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **268**, 2473-2478.
- Breed, G. A., Bowen, W. D., McMillan, J. I. and Leonard, M. L. (2006) Sexual segregation of seasonal foraging habitats in a non-migratory marine mammal. *Proceedings of the Royal Society B: Biological Sciences*, **273**, 2319-2326.
- Brodie III, E. D. and Brodie Jr, E. D. (1999) Predator-prey arms races. *Bioscience*, **49**, 557-568.
- Bromham, L., Lanfear, R., Cassey, P., Gibb, G. and Cardillo, M. (2012) Reconstructing past species assemblages reveals the changing patterns and drivers of extinction through time. *Proceedings of the Royal Society B: Biological Sciences*, **279**, 4024-4032.
- Brose, U., Jonsson, T., Berlow, E. L., Warren, P., Banasek-Richter, C., Bersier, L.-F., Blanchard, J. L., Brey, T., Carpenter, S. R. and Blandenier, M.-F. C. (2006)

- Consumer-resource body-size relationships in natural food webs. *Ecology*, **87**, 2411-2417.
- Brown, J. H. and Maurer, B. A. (1986) Body size, ecological dominance and Cope's rule. *Nature*, **324**, 248-250.
- Brown, M. J., (1988) Distribution and conservation of King Billy Pine. Forestry Commision, Hobart.
- Brown, O. J. F. (2006) Tasmanian devil (*Sarcophilus harrisii*) extinction on the Australian mainland in the mid-Holocene: multicausality and ENSO intensification. *Alcheringa: An Australasian Journal of Palaeontology*, **30**, 49-57.
- Buchmann, O. L. K. and Guiler, E. R., (1977) Behavior and ecology of the Tasmanian devil, *Sarcophilus harrisii*. In: *The biology of marsupials*: 155-168. B. Stonehouse and D. Gilmore (Eds.). Macmillan, Baltimore.
- Bugoni, L., McGill, R. A. R. and Furness, R. W. (2008) Effects of preservation methods on stable isotope signatures in bird tissues. *Rapid Communications in Mass Spectrometry*, **22**, 2457-2462.
- Bulte, E. H., Horan, R. D. and Shogren, J. F. (2003) Is the Tasmanian tiger extinct? A biological-economic re-evaluation. *Ecological Economics*, **45**, 271-279.
- Bump, J. K., Fox-Dobbs, K., Bada, J. L., Koch, P. L., Peterson, R. O. and Vucetich, J. A. (2007) Stable isotopes, ecological integration and environmental change: wolves record atmospheric carbon isotope trend better than tree rings. *Proceedings of the Royal Society B: Biological Sciences*, **274**, 2471-2480.
- Burnett, S., (1993) The conservation status of the Tiger quoll, *Dasyurus maculatus gracilis* in North Queensland. Queensland Department of Environment and Heritage, Brisbane.
- Burnett, S. and Marsh, H., (2004) Conservation of the Spotted-tailed Quoll, *Dasyurus maculatus*: a conceptual and applied model with particular reference to populations of the endangered *D. m. gracilis*. In: *Conservation of Australia's Forest Fauna*: 624-638. D. Lunney (Ed.). Royal Zoological Society of New South Wales, Mosman.

REFERENCES

- Burns, J., Trumble, S., Castellini, M. and Testa, J. (1998) The diet of Weddell seals in McMurdo Sound, Antarctica as determined from scat collections and stable isotope analysis. *Polar Biology*, **19**, 272-282.
- Burton, R. K. and Koch, P. L. (1999) Isotopic tracking of foraging and long-distance migration in northeastern Pacific pinnipeds. *Oecologia*, **119**, 578-585.
- Caraco, T. and Wolf, L. (1975) Ecological determinants of group sizes of foraging lions. *American Naturalist*, **109**, 343-352.
- Carbone, C., Du Toit, J. and Gordon, I. (1997) Feeding success in African wild dogs: does kleptoparasitism by spotted hyenas influence hunting group size? *Journal of Animal Ecology*, **66**, 318-326.
- Carbone, C., Mace, G. M., Roberts, S. C. and MacDonald, D. W. (1999) Energetic constraints on the diet of terrestrial carnivores. *Nature*, **402**, 286-288.
- Carbone, C., Teacher, A. and Rowcliffe, J. M. (2007) The costs of carnivory. *PLoS Biol*, **5**, e22.
- Cardillo, M. and Bromham, L. (2001) Body Size and Risk of Extinction in Australian Mammals. *Conservation Biology*, **15**, 1435-1440.
- Cardillo, M., Mace, G. M., Jones, K. E., Bielby, J., Bininda-Emonds, O. R. P., Sechrest, W., Orme, C. D. L. and Purvis, A. (2005) Multiple causes of high extinction risk in large mammal species. *Science*, **309**, 1239-1241.
- Cardillo, M., Purvis, A., Sechrest, W., Gittleman, J. L., Bielby, J. and Mace, G. M. (2004) Human population density and extinction risk in the world's carnivores. *PLoS Biology*, **2**, e197.
- Case, J. A. (1985) Differences in prey utilization by Pleistocene marsupial carnivores, *Thylacoleo carnifex* (Thylacoleonidae) and *Thylacinus cynocephalus* (Thylacinidae). *Australian Mammalogy*, **8**, 45-52.
- Caut, S., Angulo, E. and Courchamp, F. (2009) Variation in discrimination factors ($\Delta^{15}\text{N}$ and $\Delta^{13}\text{C}$): the effect of diet isotopic values and applications for diet reconstruction. *Journal of Applied Ecology*, **46**, 443-453.
- Ceballos, G. and Ehrlich, P. R. (2002) Mammal population losses and the extinction crisis. *Science*, **296**, 904-907.

- Cerling, T. E., Harris, J. M., MacFadden, B. J., Leakey, M. G., Quade, J., Eisenmann, V. and Ehleringer, J. R. (1997) Global vegetation change through the Miocene/Pliocene boundary. *Nature*, **389**, 153-158.
- Cerling, T., Harris, J., Hart, J., Kaleme, P., Klingel, H., Leakey, M., Levin, N., Lewison, R. and Passey, B. (2008) Stable isotope ecology of the common hippopotamus. *Journal of Zoology*, **276**, 204-212.
- Chamoli, U. and Wroe, S. (2011) Allometry in the distribution of material properties and geometry of the felid skull: Why larger species may need to change and how they may achieve it. *Journal of Theoretical Biology*, **283**, 217-226.
- Charnov, E. L., Orians, G. H. and Hyatt, K. (1976) Ecological implications of resource depression. *The American Naturalist*, **110**, 247-259.
- Cherel, Y., Hobson, K. A., Guinet, C. and Vanpe, C. (2007) Stable isotopes document seasonal changes in trophic niches and winter foraging individual specialization in diving predators from the Southern Ocean. *Journal of Animal Ecology*, **76**, 826-836.
- Chiba, S. and Roy, K. (2011) Selectivity of terrestrial gastropod extinctions on an oceanic archipelago and insights into the anthropogenic extinction process. *Proceedings of the National Academy of Sciences*, **108**, 9496-9501.
- Christensen, J. T. and Richardson, K. (2008) Stable isotope evidence of long-term changes in the North Sea food web structure. *Marine Ecology Progress Series*, **368**, 1-8.
- Christiansen, P. and Wroe, S. (2007) Bite forces and evolutionary adaptations to feeding ecology in carnivores. *Ecology*, **88**, 347-358.
- Clausen, P., Wroe, S., McHenry, C., Moreno, K. and Bourke, J. (2008) The vector of jaw muscle force as determined by computer-generated three dimensional simulation: A test of Greaves' model. *Journal of biomechanics*, **41**, 3184-3188.
- Clementz, M. T., Hoppe, K. A. and Koch, P. L. (2003) A paleoecological paradox: the habitat and dietary preferences of the extinct tethythere *Desmostylus*, inferred from stable isotope analysis. *Journal Information*, **29**, 506-519.

REFERENCES

- Clutton-Brock, T. and Harvey, P. H. (1978) Mammals, resources and reproductive strategies. *Nature*, **273**, 191.
- Cohen, J. E., Pimm, S. L., Yodzis, P. and Saldaña, J. (1993) Body sizes of animal predators and animal prey in food webs. *Journal of Animal Ecology*, **62**, 67-78.
- Colles, A., Liow, L. H. and Prinzing, A. (2009) Are specialists at risk under environmental change? Neoecological, paleoecological and phylogenetic approaches. *Ecology Letters*, **12**, 849-863.
- Cook, S., Weinstein, A. and Klawitter, J. (1982) Materials Science A Three-dimensional Finite Element Analysis of a Porous Rooted Co-Cr-Mo Alloy Dental Implant. *Journal of dental research*, **61**, 25-29.
- Corbett, L. K. and Newsome, A. E. (1987) The feeding ecology of the Dingo. III. Dietary relationships with widely fluctuating prey populations in arid Australia: An hypothesis of alternation of predation. *Oecologia*, **74**, 215-227.
- Cosgrove, R. F. and Allen, J. (2001) *Prey choice and hunting strategies in the Late Pleistocene: Evidence from Southwest Tasmania*. Pandanus Books. Canberra.
- Côté, S. D., Rooney, T. P., Tremblay, J.-P., Dussault, C. and Waller, D. M. (2004) Ecological impacts of deer overabundance. *Annual review of ecology, evolution, and systematics*, **35**, 113-147.
- Courchamp, F., Langlais, M. and Sugihara, G. (2001) Cats protecting birds: modelling the mesopredator release effect. *Journal of Animal Ecology*, **68**, 282-292.
- Crompton, A. W., Barnet, J., Lieberman, D. E., Owerkowicz, T., Skinner, J. and Baudinette, R. V. (2008) Control of jaw movements in two species of macropodines (*Macropus eugenii* and *Macropus rufus*). *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology*, **150**, 109-123.
- Cronin, L. (1991) *Key guide to Australian mammals*. Reed Books, Sydney.
- Crooks, K. R. and Soulé, M. E. (1999) Mesopredator release and avifaunal extinctions in a fragmented system. *Nature*, **400**, 563-566.

- Crowley, B. E., Godfrey, L. R. and Irwin, M. T. (2011) A glance to the past: subfossils, stable isotopes, seed dispersal, and lemur species loss in southern Madagascar. *American Journal of Primatology*, **73**, 25-37.
- Cullen Jr, L., Bodmer, R. E. and Valladares Pádua, C. (2000) Effects of hunting in habitat fragments of the Atlantic forests, Brazil. *Biological Conservation*, **95**, 49-56.
- Dalerum, F. and Angerbjörn, A. (2005) Resolving temporal variation in vertebrate diets using naturally occurring stable isotopes. *Oecologia*, **144**, 647-658.
- Dalerum, F., Bennett, N. C. and Clutton-Brock, T. H. (2007) Longitudinal differences in ^{15}N between mothers and offspring during and after weaning in a small cooperative mammal, the meerkat (*Suricata suricatta*). *Rapid Communications in Mass Spectrometry*, **21**, 1889-1892.
- Damuth, J. (1981) Population density and body size in mammals. *Nature*, **290**, 699-700.
- Darimont, C. T. and Reimchen, T. E. (2002) Intra-hair stable isotope analysis implies seasonal shift to salmon in gray wolf diet. *Canadian Journal of Zoology*, **80**, 1638-1642.
- Davidson, A. D., Hamilton, M. J., Boyer, A. G., Brown, J. H. and Ceballos, G. (2009) Multiple ecological pathways to extinction in mammals. *Proceedings of the National Academy of Sciences*, **106**, 10702-10705.
- Davies, J. L. (1965) *Atlas of Tasmania*. Lands and Surveys Department, Hobart.
- Davies, K. F., Margules, C. R. and Lawrence, J. F. (2000) Which traits of species predict population declines in experimental forest fragments? *Ecology*, **81**, 1450-1461.
- Davis, C. (1940) Preliminary survey of the vegetation near New Harbour, South-west Tasmania. *Papers and Proceedings of the Royal Society of Tasmania*, **40**, 1-10.
- Dawson, J. P., Claridge, A. W., Triggs, B. and Paull, D. J. (2007) Diet of a native carnivore, the spotted-tailed quoll (*Dasyurus maculatus*), before and after an intense wildfire. *Wildlife Research*, **34**, 342-351.
- Deb, D. (1997) Trophic uncertainty vs parsimony in food web research. *Oikos*, **78**, 191-194.

REFERENCES

- Dehn, L.-A., Sheffield, G. G., Follmann, E. H., Duffy, L. K., Thomas, D. L. and O'Hara, T. M. (2007) Feeding ecology of phocid seals and some walrus in the Alaskan and Canadian Arctic as determined by stomach contents and stable isotope analysis. *Polar Biology*, **30**, 167-181.
- Deines, P., (1980) The isotopic composition of reduced organic carbon. In: *Handbook of environmental isotope geochemistry*: 329–406. P. Fritz and J. C. Fontes (Eds.). Elsevier, Amsterdam.
- DeNiro, M. J. (1985) Postmortem preservation and alteration of in vivo bone collagen isotope ratios in relation to palaeodietary reconstruction. **317**, 806-809.
- DeNiro, M. J. and Epstein, S. (1978) Influence of diet on the distribution of carbon isotopes in animals. *Journal of The Geochemical Society and The Meteoritical Society*, **45**, 341-351.
- DeNiro, M. J. and Epstein, S. (1981) Influence of diet on the distribution of nitrogen isotopes in animals. *Journal of The Geochemical Society and The Meteoritical Society*, **45**, 341-351.
- Deniro, M. J. and Schoeniger, M. J. (1983) Stable carbon and nitrogen isotope ratios of bone collagen: Variations within individuals, between sexes, and within populations raised on monotonous diets. *Journal of Archaeological Science*, **10**, 199-203.
- Diamond, J., (1984) "Normal" extinctions of isolated populations. In: *Extinctions*: 191-246. M. H. Nitecki (Ed.). Chicago, USA: Chicago University Press.
- Dickman, C. (1988) Body size, prey size, and community structure in insectivorous mammals. *Ecology*, **69**, 569-580.
- Dove, H. S. (1924) Notes on the Tasmanian Emu. *Emu*, **23**, 221-222.
- Dovers, S., (2000) Still settling Australia: environment, history, and policy. In: *Environmental History and Policy: Still Settling Australia*: 2-23. S. Dover (Ed.). Oxford University Press, Melbourne.
- Drossel, B., Higgs, P. G. and McKane, A. J. (2001) The influence of predator-prey population dynamics on the long-term evolution of food web structure. *Journal of Theoretical Biology*, **208**, 97-107.

- Dumont, E. R., Grosse, I. R. and Slater, G. J. (2009) Requirements for comparing the performance of finite element models of biological structures. *Journal of Theoretical Biology*, **256**, 96-103.
- Edgar, G. J., Samson, C. R. and Barrett, N. S. (2005) Species extinction in the marine environment: Tasmania as a regional example of overlooked losses in biodiversity. *Conservation Biology*, **19**, 1294-1300.
- Edgar, R. and Belcher, C., (1995) Spotted-tailed quoll *Dasyurus maculatus* (Kerr, 1792). In: *The Mammals of Australia*: 67–69. R. Strahan (Ed.). Reed Books, Sydney.
- Eggers, T. and Hefin, J. T. (2000) You are what you eat...or are you? *Trends in ecology & evolution*, **15**, 265-266.
- Enright, N. J. and Thomas, I. (2008) Pre-European Fire Regimes in Australian Ecosystems. *Geography Compass*, **2**, 979-1011.
- Epstein Jr, E. and Munderloh, N. (1978) Human skin collagen. Presence of type I and type III at all levels of the dermis. *Journal of Biological Chemistry*, **253**, 1336-1337.
- Estes, J. A., Terborgh, J., Brashares, J. S., Power, M. E., Berger, J., Bond, W. J., Carpenter, S. R., Essington, T. E., Holt, R. D. and Jackson, J. B. (2011) Trophic downgrading of planet earth. *Science*, **333**, 301-306.
- Evans, M. C., Macgregor, C. and Jarman, P. J. (2006) Diet and feeding selectivity of common wombats. *Wildlife Research*, **33**, 321-330.
- Evans, R. D. (2001) Physiological mechanisms influencing plant nitrogen isotope composition. *Trends in plant science*, **6**, 121-126.
- Ewer, R. F. (1969) Some observations on the killing and eating of prey by two dasyurid marsupials: the Mulgara, *Dasyercus cristicauda*, and the Tasmanian devil, *Sarcophilus harrisii*. *Zeitschrift für Tierpsychologie*, **26**, 23–38.
- Ezard, T. H. G., Aze, T., Pearson, P. N. and Purvis, A. (2011) Interplay between changing climate and species' ecology drives macroevolutionary dynamics. *Science*, **332**, 349-351.

REFERENCES

- Felicetti, L. A., Schwartz, C. C., Rye, R. O., Haroldson, M. A., Gunther, K. A., Phillips, D. L. and Robbins, C. T. (2003) Use of sulfur and nitrogen stable isotopes to determine the importance of whitebark pine nuts to Yellowstone grizzly bears. *Canadian Journal of Zoology*, **81**, 763-770.
- Fensham, R. (1989) The pre-European vegetation of the Midlands, Tasmania: a floristic and historical analysis of vegetation patterns. *Journal of Biogeography*, **16**, 29-45.
- Feranec, R. S. (2003) Stable isotopes, hypsodonty, and the paleodiet of *Hemiauchenia* (Mammalia: Camelidae): a morphological specialization creating ecological generalization. *Paleobiology*, **29**, 230-242.
- Feranec, R. S. (2004) Isotopic evidence of saber-tooth development, growth rate, and diet from the adult canine of *Smilodon fatalis* from Rancho La Brea. *Palaeogeography, Palaeoclimatology, Palaeoecology*, **206**, 303-310.
- Féret, M., Gauthier, G., Béchet, A., Giroux, J.-F. and Hobson, K. A. (2003) Effect of a spring hunt on nutrient storage by greater snow geese in Southern Quebec. *The Journal of Wildlife Management*, **67**, 796-807.
- Fernández-Mosquera, D., Vila-Taboada, M. and Grandal-d'Anglade, A. (2001) Stable isotopes data ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) from the cave bear (*Ursus spelaeus*): a new approach to its palaeoenvironment and dormancy. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **268**, 1159-1164.
- Figueirido, B. and Janis, C. M. (2011) The predatory behaviour of the thylacine: Tasmanian tiger or marsupial wolf? *Biology Letters*, **7**, 937-940.
- Figueirido, B., Palmqvist, P. and Pérez-Claros, J. (2008) Ecomorphological correlates of craniodental variation in bears and paleobiological implications for extinct taxa: an approach based on geometric morphometrics. *Journal of Zoology*, **277**, 70-80.
- Finucane, B. C. (2007) Mummies, maize, and manure: multi-tissue stable isotope analysis of late prehistoric human remains from the Ayacucho Valley, Peru. *Journal of Archaeological Science*, **34**, 2115-2124.
- Fitzgerald, A., (1984) Diet of the possum (*Trichosurus vulpecula*) in three Tasmanian forest types and its relevance to the diet of possums in New Zealand forests. In:

- Possums and gliders*: 137–143. A. Smith and I. Hume (Eds.). Surrey Beatty and Sons, Sydney.
- Flaherty, E. A. and Ben-David, M. (2010) Overlap and partitioning of the ecological and isotopic niches. *Oikos*, **119**, 1409-1416.
- Fleay, D. (1932) The rare dasyures (native cats). *Victorian Naturalist*, **49**, 63-69.
- Fogel, M. L., Tuross, N. and Owsley, D. W., (1989) Nitrogen isotope tracers of human lactation in modern and archaeological populations. In: *Annual Report to the Director, Geophysical Laboratory*: 111-117. Carnegie Institution, Washington.
- Fortin, D., Beyer, H. L., Boyce, M. S., Smith, D. W., Duchesne, T. and Mao, J. S. (2005) Wolves influence elk movements: behaviour shapes a trophic cascade in Yellowstone national park. *Ecology*, **86**, 1320-1330.
- France, R. and Peters, R. (1997) Ecosystem differences in the trophic enrichment of ^{13}C in aquatic food webs. *Canadian Journal of Fisheries and Aquatic Sciences*, **54**, 1255-1258.
- Freeland, W. J. and Winter, J. W. (1975) Evolutionary consequences of eating: *Trichosurus vulpecula* (marsupialia) and the genus *Eucalyptus*. *Journal of Chemical Ecology*, **1**, 439-455.
- Frid, A. and Dill, L. M. (2002) Human-caused disturbance stimuli as a form of predation risk. *Conservation Ecology*, **6**, 11 [Online, URL: <<http://www.consecol.org/vol6/iss1/art11>>.]
- Fuller, B. T., Fuller, J. L., Harris, D. A. and Hedges, R. E. M. (2006) Detection of breastfeeding and weaning in modern human infants with carbon and nitrogen stable isotope ratios. *American Journal of Physical Anthropology*, **129**, 279-293.
- Furness, R. W., Crane, J. E., Bearhop, S., Garthe, S., Kakela, A., Kakela, R., Reijo, K., Kelly, A., Kubetzki, U., Votier, S. C. and Waldron, S. (2006) Techniques to link individual migration patterns of seabirds with diet specialization, condition and breeding performance. *ARDEA*, **94**, 631-639.
- Galliez, M., de Souza Leite, M., Queiroz, T. L. and dos Santos Fernandez, F. A. (2009) Ecology of the water opossum *Chironectes minimus* in Atlantic forest streams of southeastern Brazil. *Journal of Mammalogy*, **90**, 93-103.

REFERENCES

- Gannes, L. Z., del Rio, C. M. and Koch, P. (1998) Natural abundance variations in stable isotopes and their potential uses in animal physiological ecology. *Comparative Biochemistry and Physiology*, **119**, 725-737.
- Garvey, J. (2010) Economic anatomy of the Bennett's wallaby (*Macropus rufogriseus*): Implications for understanding human hunting strategies in late Pleistocene Tasmania. *Quaternary International*, **211**, 144-156.
- Gendron, R. P. (1987) Models and mechanisms of frequency-dependent predation. *American Naturalist*, **130**, 603-623.
- Geng, J. P., Tan, K. and Liu, G. R. (2001) Application of finite element analysis in implant dentistry: a review of the literature. *The Journal of prosthetic dentistry*, **85**, 585-598.
- Gilbert, J. (1959) Forest succession in the Florentine valley, Tasmania. *Papers and Proceedings of the Royal Society of Tasmania*, **93**, 129-152.
- Gilbert, J. M. (1979) Fires in the Tasmanian bush. *The Tasmanian Tramp*, **23**, 118-122.
- Gilbert, R. O. (1987) *Statistical Methods for Environmental Pollution Monitoring*. Van Nostrand Reinhold. New York.
- Gittleman, J. L. (1985) Carnivore body size: ecological and taxonomic correlates. *Oecologia*, **67**, 540-554.
- Gittleman, J. L., Funk, S. M., MacDonald, D. W. and Wayne, R. K. (2001) *Carnivore conservation*. Cambridge University Press. Cambridge.
- Glen, A. and Dickman, C. (2006) Diet of the spotted-tailed quoll (*Dasyurus maculatus*) in eastern Australia: effects of season, sex and size. *Journal of Zoology*, **269**, 241-248.
- Glen, A. S. and Dickman, C. R. (2005) Complex interactions among mammalian carnivores in Australia, and their implications for wildlife management. *Biological Reviews*, **80**, 387-401.
- Goswami, A., Milne, N., Wroe, S., Goswami, A., Milne, N. and Wroe, S. (2011) Biting through constraints: cranial morphology, disparity and convergence across living and fossil carnivorous mammals. *Proceedings of the Royal Society B: Biological Sciences*, **278**, 1831-1839.

- Gould, J., Dixon, J. M., Gould, J., Ornithologist, G. B., Gould, J. and Ornithologue, G.-B. (1974) *Australian marsupials and monotremes*. Macmillan, Melbourne.
- Gowlland, R. and Gowlland, K. (1976) *Trampled Wilderness. The History of South-west Tasmania*, 3rd edn. Richmond Printers. Devonport.
- Granato, G. E. (2006) *Kendall-Theil Robust Line (KTRLine-version 1.0)--a Visual Basic Program for Calculating and Graphing Robust Nonparametric Estimates of Linear-regression Coefficients Between Two Continuous Variables*. US Department of the Interior, US Geological Survey.
- Grant, T. (1982) Food of the platypus, *Ornithorhynchus anatinus* (Monotremata: Ornithorhynchidae), from various water bodies in New South Wales. *Australian Mammalogy*, **5**, 235-236.
- Grant, T. and Temple-Smith, P. (1998) Field biology of the platypus (*Ornithorhynchus anatinus*): historical and current perspectives. *Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences*, **353**, 1081-1091.
- Gratton, C. and Forbes, A. E. (2006) Changes in $\delta^{13}\text{C}$ stable isotopes in multiple tissues of insect predators fed isotopically distinct prey. *Oecologia*, **147**, 615-624.
- Greaves, W. S. (1980) The mammalian jaw mechanism - the high glenoid cavity. *The American Naturalist*, **116**, 432-440.
- Green, R. H. (1967) Notes on the devil (*Sarchophilus harrisii*) and the quoll (*Dasyurus viverrinus*) in north-eastern Tasmania. *Records of the Queen Victoria Museum*, **27**, 1-12.
- Green, R. H. (1989) *Birds of Tasmania*. Potoroo Publishing. Launceston, Tasmania.
- Green, R. H. and Scarborough, T. J. (1990) The spotted-tailed quoll *Dasyurus maculatus* (Dasyuridae, Marsupialia) in Tasmania. *The Tasmanian Naturalist*, **100**, 1-14.
- Greenwood, J. J. and Elton, R. A. (1979) Analysing experiments on frequency-dependent selection by predators. *The Journal of Animal Ecology*, **48**, 721-737.
- Griffiths, D. (1980) Foraging costs and relative prey size. *The American Naturalist*, **116**, 743-752.

REFERENCES

- Groombridge, B. (1993) *1994 IUCN Red List of Threatened Animals*. IUCN.. Gland, Switzerland and Cambridge.
- Groves, C., Wilson, D. and Reeder, D. (2005) *Mammal species of the world*. John Hopkins University Press, Baltimore.
- Guiler, E. (1970) Observations on the Tasmanian devil, *Sarcophilus harrisii* (Marsupialia : Dasyuridae) I. Numbers, home, range, movements and food in two populations. *Australian Journal of Zoology*, **18**, 49-62.
- Guiler, E. R. (1957) The present status of some Tasmanian mammals in relation to the fur industry of Tasmania. *Papers and Proceedings of the Royal Society of Tasmania*, **91**, 117-128.
- Guiler, E. R. (1961) The former distribution and decline of the thylacine. *Australian Journal of Science*, **23**, 207-210.
- Guiler, E. R. (1978) Observations on the Tasmanian devil, *Sarcophilus harrisii* (Dasyuridae: Marsupiala) at Granville Harbour, 1966-75. . *Papers and Proceedings of the Royal Society of Tasmania*, **112**, 161-163.
- Guiler, E. R. (1982) Temporal and spatial distribution of the Tasmanian devil, *Sarcophilus harrisii* (Dasyuridae: Marsupialia). *Papers and Proceedings of the Royal Society of Tasmania*, **116**, 153-163.
- Guiler, E. R. (1985) *Thylacine: The tragedy of the Tasmanian tiger*. Oxford University Press, Melbourne.
- Guiler, E. R. (1992) *The Tasmanian Devil*. St. David's Park Publishing. Hobart.
- Guiler, E. R. and Godard, P. (1998) *Tasmanian tiger: A lesson to be learnt*. Abrolhos Publishing, Perth.
- Haemig, P. D. (2001) Symbiotic nesting of birds with formidable animals: a review with applications to biodiversity conservation. *Biodiversity and Conservation*, **10**, 527-540.
- Hanba, Y. T., Mori, S., Lei, T. T., Koike, T. and Wada, E. (1997) Variations in leaf $\delta^{13}\text{C}$ along a vertical profile of irradiance in a temperate Japanese forest. *Oecologia*, **110**, 253-261.

- Hannah, L., Midgley, G., Andelman, S., Araújo, M., Hughes, G., Martinez-Meyer, E., Pearson, R. and Williams, P. (2007) Protected area needs in a changing climate. *Frontiers in Ecology and the Environment*, **5**, 131-138.
- Hare, E. P., Fogel, M. L., Stafford, T. W., Mitchell, A. D. and Hoering, T. C. (1991) The isotopic composition of carbon and nitrogen in individual amino acids isolated from modern and fossil proteins. *Journal of Archaeological Science*, **18**, 277-292.
- Harris, G. P. (1808) Description of two new species of *Didelphis* from Van Diemen's Land. *Transactions of the Linnean Society of London*, **9**, 174-178.
- Harrison, S. M., Zazzo, A., Bahar, B., Monahan, F. J., Moloney, A. P., Scrimgeour, C. M. and Schmidt, O. (2007) Using hooves for high-resolution isotopic reconstruction of bovine dietary history. *Rapid Communications in Mass Spectrometry*, **21**, 479-486.
- Hawkins, C. E., Baars, C., Hesterman, H., Hocking, G. J., Jones, M. E., Lazenby, B., Mann, D., Mooney, N., Pemberton, D., Pyecroft, S., Restani, M. and Wiersma, J. (2006) Emerging disease and population decline of an island endemic, the Tasmanian devil *Sarcophilus harrisii*. *Biological Conservation*, **131**, 307-324.
- Heaton, T. H. E. (1999) Spatial, species, and temporal variations in the $^{13}\text{C}/^{12}\text{C}$ ratios of C_3 plants: Implications for palaeodiet studies. *Journal of Archaeological Science*, **26**, 637-649.
- Heberle, G. (2004) Reports of alleged thylacine sightings in Western Australia. *Conservation Science Western Australia*, **5**, 1-5.
- Hedges, R. E. M. (2003) On bone collagen—apatite-carbonate isotopic relationships. *International Journal of Osteoarchaeology*, **13**, 66-79.
- Hemmingsen, A. M. (1960) Energy metabolism as related to body size and respiratory surfaces, and its evolution. Reports of the Steno Memorial Hospital and Nordisk Insulin Laboratorium (Copenhagen), **9**, 6-110.
- Heupink, T. H., Huynen, L. and Lambert, D. M. (2011) Ancient DNA suggests dwarf and 'giant' emu are conspecific. *PLoS ONE*, **6**, e18728.

REFERENCES

- Hilderbrand, G. V., Farley, S. D., Robbins, C. T., Hanley, T. A., Titus, K. and Servheen, C. (1996) Use of stable isotopes to determine diets of living and extinct bears. *Canadian Journal of Zoology*, **74**, 2080-2088.
- Hilton, G. M., Thompson, D. R., Sagar, P. M., Cuthbert, R. J., Cherel, Y. and Bury, S. J. (2006) A stable isotopic investigation into the causes of decline in a sub-Antarctic predator, the rockhopper penguin *Eudyptes chrysocome*. *Global Change Biology*, **12**, 611-625.
- Hirons, A. C., Schell, D. M. and Finney, B. P. (2001) Temporal records of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in North Pacific pinnipeds: inferences regarding environmental change and diet. *Oecologia*, **129**, 591-601.
- Hobson, K. A. (1999) Tracing origins and migration of wildlife using stable isotopes: a review. *Oecologia*, **120**, 314-326.
- Hobson, K. A. and Montevecchi, W. A. (1991) Stable isotopic determinations of trophic relationships of great auks. *Oecologia*, **87**, 528-531.
- Hobson, K. A., Schell, D. M., Renouf, D. and Noseworthy, E. (1996) Stable carbon and nitrogen isotopic fractionation between diet and tissues of captive seals: implications for dietary reconstructions involving marine mammals. *Canadian Journal of Fisheries and Aquatic Sciences*, **53**, 528-533.
- Hobson, K. A. and Wassenaar, L. I. (1997) Linking breeding and wintering grounds of neotropical migrant songbirds using stable hydrogen isotopic analysis of feathers. *Oecologia*, **109**, 142-148.
- Hocking, G. J. and Driessen, M. M. (1996) Mammals of Northeast Tasmania. *Records of the Queen Victoria Museum and Art Gallery*, **103**, 163-168.
- Högberg, P. and Johannisson, C. (1993) ^{15}N Abundance of forests is correlated with losses of nitrogen. *Plant and Soil*, **157**, 147-150.
- Holliday, J. A. and Stepan, S. J. (2004) Evolution of hypercarnivory: the effect of specialization on morphological and taxonomic diversity. *Paleobiology*, **30**, 108-128.

- Husseman, J. S., Murray, D. L., Power, G., Mack, C., Wenger, C. and Quigley, H. (2003) Assessing differential prey selection patterns between two sympatric large carnivores. *Oikos*, **101**, 591-601.
- IUCN, (2012) IUCN Red List of Threatened Species. Version 2012.2 Online, URL: <<http://www.iucnredlist.org>>.]
- Jackson, W. D. (1968) Fire, air, water and earth—an elemental ecology of Tasmania. *Proceedings of the Ecological Society of Australia*, **3**, 9-16.
- Jerozolinski, A. and Peres, C. A. (2003) Bringing home the biggest bacon: a cross-site analysis of the structure of hunter-kill profiles in Neotropical forests. *Biological Conservation*, **111**, 415-425.
- Johnsingh, A. (1992) Prey selection in three large sympatric carnivores in Bandipur. *Mammalia*, **56**, 517-526.
- Johnson, C. N. and Wroe, S. (2003) Causes of extinction of vertebrates during the Holocene of mainland Australia: arrival of the dingo, or human impact? *The Holocene*, **13**, 941-948.
- Johnson, W. E. and Franklin, W. L. (1994) Spatial resource partitioning by sympatric grey fox (*Dusicyon griseus*) and culpeo fox (*Dusicyon culpaeus*) in southern Chile. *Canadian Journal of Zoology*, **72**, 1788-1793.
- Jones, M. E. (1995) Guild structure of the large marsupial carnivores in Tasmania. PhD thesis. University of Tasmania, Hobart.
- Jones, M. E. (1997) Character displacement in Australian dasyurid carnivores: size relationships and prey size patterns. *Ecology*, **78**, 2569-2587.
- Jones, M. E. (1998) The function of vigilance in sympatric marsupial carnivores: the eastern quoll and the Tasmanian devil. *Animal Behaviour*, **56**, 1279-1284.
- Jones, M. E., (2003) Convergence in ecomorphology and guild structure among marsupial and placental carnivores. In: *Predators with pouches: the biology of carnivorous marsupials*: 285-269. M. E. Jones, C. Dickman and M. Archer (Eds.). CSIRO Publishing, Collingwood.

REFERENCES

- Jones, M. E. and Barmuta, L. A. (1998) Diet overlap and abundance of sympatric dasyurid carnivores: a hypothesis of competition? *The Journal of Animal Ecology*, **67**, 410-421.
- Jones, M. E. and Barmuta, L. A. (2000) Niche differentiation among sympatric Australian dasyurid carnivores. *Journal of Mammalogy*, **81**, 434-447.
- Jones, M. E., M., O., A., B. C., Morris, K., Murray, A. J., Woolley, P. A., Firestone, K. B., Johnson, B. and Burnett, S., (2003) Carnivore concerns: problems, issues and solutions for conserving Australasia's marsupial carnivores. In: *Predators with Pouches: the Biology of Carnivorous Marsupials* 418-430. M. E. Jones, C. R. Dickman and M. Archer (Eds.). CSIRO Publishing, Melbourne.
- Jones, M. E., Paetkau, D., Geffen, E. and Moritz, C. (2004a) Genetic diversity and population structure of Tasmanian devils, the largest marsupial carnivore. *Molecular Ecology*, **13**, 2197-2209.
- Jones, M. E. and Rose, R. K., (1996) Preliminary assessment of distribution and habitat associations of the spotted-tailed quoll (*Dasyurus maculatus maculatus*) and eastern quoll (*D. viverrinus*) in Tasmania to determine conservation and reservation status. Nature Conservation Branch, Parks and Wildlife Service. Report to the Tasmanian Regional Forest Agreement Environment and Heritage Technical Committee, Hobart, Tasmania.
- Jones, M. E., Smith, G. C. and Jones, S. M. (2004b) Is anti-predator behaviour in Tasmanian eastern quolls (*Dasyurus viverrinus*) effective against introduced predators? *Animal Conservation*, **7**, 155-160.
- Jones, M. E. and Stoddart, D. M. (1998) Reconstruction of the predatory behaviour of the extinct marsupial thylacine (*Thylacinus cynocephalus*). *Journal of Zoology*, **246**, 239-246.
- Kamilar, J. M. and Paciulli, L. M. (2008) Examining the extinction risk of specialized folivores: a comparative study of colobine monkeys. *American Journal of Primatology*, **70**, 816-827.
- Karanth, K. U. and Sunquist, M. E. (1995) Prey selection by tiger, leopard and dhole in tropical forests. *Journal of Animal Ecology*, **64**, 439-450.

- Kelly, J. F. (2000) Stable isotopes of carbon and nitrogen in the study of avian and mammalian trophic ecology. *Canadian Journal of Zoology*, **78**, 1-27.
- Kelt, D.A. and Van Vuren, D.H. (2001) The ecology and macroecology of mammalian home range area. *American Naturalist*, **157**, 637–645.
- Kerle, A. (2001) *Possums: the brushtails, ringtails and greater glider*. University of New South Wales Press, Sydney.
- Kerle, J. A., (1984) Variation in the ecology of *Trichosurus*: its adaptive significance. In: *Possums and gliders*: 115-128. A. Smith and I. Hume (Eds.). Surrey Beatty and Sons, Sydney.
- Klaassen, M., Thums, M. and Hume, I. D. (2005) Effects of diet change on carbon and nitrogen stable-isotope ratios in blood cells and plasma of the long-nosed bandicoot (*Perameles nasuta*). *Australian Journal of Zoology*, **52**, 635-647.
- Kleiber, M. (1961) *The fire of life. An introduction to animal energetics*. Wiley, New York.
- Knibbs, G. (1913) *Official Year Book Of The Commonwealth of Australia 1901-1912*. The Bureau of Census and Statistics, Melbourne.
- Koch, P. L., Fogel, M. L. and Tuross, N., (2007) Tracing the diet of fossil animals using stable isotopes. In: *Stable isotopes in ecology and environmental science*: 63-92. R. H. Michener and K. Lajtha (Eds.). Oxford: Blackwell Scientific Publications.
- Koenigswald, W. W., Rensberger, J. M. and Pfretzschner, H. U. (1987) Changes in tooth enamel of early Paleocene mammals allowing increased diet diversity. *Nature*, **328**, 150-152.
- Kohn, M. J., McKay, M. P. and Knight, J. L. (2005) Dining in the Pleistocene—Who's on the menu? *Geology*, **33**, 649-652.
- Kondoh, M. (2007) Anti-predator defence and the complexity–stability relationship of food webs. *Proceedings of the Royal Society B: Biological Sciences*, **274**, 1617-1624.
- Korpimäki, E. (1987) Dietary shifts, niche relationships and reproductive output of coexisting Kestrels and Long-eared Owls. *Oecologia*, **74**, 277-285.

REFERENCES

- Krajewski, C., Buckley, L. and Westerman, M. (1997) DNA phylogeny of the marsupial wolf resolved. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **264**, 911-917.
- Krajewski, C., Driskell, A. C., Baverstock, P. R. and Braun, M. J. (1992) Phylogenetic Relationships of the Thylacine (Mammalia: Thylacinidae) among Dasyuroid Marsupials: Evidence from Cytochrome b DNA Sequences. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **250**, 19-27.
- Kurle, C. M. (2002) Stable-isotope ratios of blood components from captive northern fur seals (*Callorhinus ursinus*) and their diet: applications for studying the foraging ecology of wild otariids. *Canadian Journal of Zoology*, **80**, 902-909.
- Laliberté, A. S. and Ripple, W. J. (2004) Range contractions of North American carnivores and ungulates. *Bioscience*, **54**, 123-138.
- Lauder, G. V. and Thomason, J. J. (1995) *On the inference of function from structure*. Cambridge University Press, Cambridge.
- Layman, C. A., Arrington, D. A., Montaña, C. G. and Post, D. M. (2007) Can stable isotope ratios provide for community-wide measures of trophic structure? *Ecology*, **88**, 42-48.
- Le Souef, D. (1904) Extinct Tasmanian emu.. *Emu*, **3**, 229-231.
- Lecomte, N., Ahlstrøm, Ø., Ehrich, D., Fuglei, E., Ims, R. A. and Yoccoz, N. G. (2011) Intrapopulation variability shaping isotope discrimination and turnover: experimental evidence in arctic foxes. *PLoS ONE*, **6**, e21357.
- Lee-Thorp, J. A., Van Der Merwe, N. J. and Brain, C. (1989) Isotopic evidence for dietary differences between two extinct baboon species from Swartkrans. *Journal of Human Evolution*, **18**, 183-189.
- Lee, S. H., Schell, D. M., McDonald, T. L. and Richardson, W. J. (2005) Regional and seasonal feeding by bowhead whales *Balaena mysticetus* as indicated by stable isotope ratios. *Marine ecology. Progress series*, **285**, 271-287.
- Lehninger, A. L. (1973) *Biochemistry. 2nd Edition*. Worth. New York.

- Leonard, J. A., Vilà, C., Fox-Dobbs, K., Koch, P. L., Wayne, R. K. and Van Valkenburgh, B. (2007) Megafaunal extinctions and the disappearance of a specialized wolf ecomorph. *Current Biology*, **17**, 1146-1150.
- Lesage, V., Hammill, M. O. and Kovacs, K. M. (2002) Diet-tissue fractionation of stable carbon and nitrogen isotopes in phocid seals. *Marine Mammal Science*, **18**, 182-193.
- Letnic, M., Fillios, M. and Crowther, M. S. (2012) Could Direct Killing by Larger Dingoes Have Caused the Extinction of the Thylacine from Mainland Australia? *PLoS ONE*, **7**, e34877.
- Lichatowich, J., Mobrand, L., Lestelle, L. and Vogel, T. (1995) An approach to the diagnosis and treatment of depleted Pacific salmon populations in Pacific Northwest watersheds. *Fisheries*, **20**, 10-18.
- Long, E. S., Sweitzer, R. A., Diefenbach, D. R. and Ben-David, M. (2005) Controlling for anthropogenically induced atmospheric variation in stable carbon isotope studies. *Oecologia*, **146**, 148-156.
- Long, K. and Nelson, J., (2010) National Recovery Plan for the Spotted-tailed Quoll *Dasyurus maculatus*. Department of Sustainability and Environment, Melbourne.
- Longin, R. (1971) New method of collagen extraction for radiocarbon analysis. *Nature*, **230**, 241-242.
- Lowenstein, J. M., Sarich, V. M. and Richardson, B. J. (1981) Albumin systematics of the extinct mammoth and Tasmanian wolf. *Nature*, **291**, 409-411.
- Lowry, J. W. J. and Merrilees, D. (1969) Age of the desiccated carcass of a thylacine (Marsupialia, Dasyuroidea) from Thylacine Hole, Nullarbor region, Western Australia. *Helictite*, **7**, 15-16.
- Lunt, I. D. (1991) Management of remnant lowland grasslands and grassy woodlands for nature conservation: a review. *Victorian Naturalist*, **108**, 56-66.
- Mac Nally, R. C. (1983) On assessing the significance of interspecific competition to guild structure. *Ecology*, **64**, 1646-1652.

REFERENCES

- Mac Nally, R. C. and Doolan, J. M. (1986) An empirical approach to guild structure: habitat relationships in nine species of eastern-Australian cicadas. *Oikos*, **47**, 33-46.
- MacCracken, J. G. and Hansen, R. M. (1987) Coyote Feeding Strategies in Southeastern Idaho: Optimal Foraging by an Opportunistic Predator? *The Journal of Wildlife Management*, **51**, 278-285.
- MacFadden, B. J., Higgins, P., Clementz, M. T. and Jones, D. S. (2004) Diets, habitat preferences, and niche differentiation of Cenozoic sirenians from Florida: evidence from stable isotopes. *Paleobiology*, **30**, 297-324.
- MacNeil, M. A., Drouillard, K. G. and Fisk, A. T. (2006) Variable uptake and elimination of stable nitrogen isotopes between tissues in fish. *Canadian Journal of Fisheries and Aquatic Sciences*, **63**, 345-353.
- Maehr, D. S., Belden, R. C., Land, E. D. and Wilkins, L. (1990) Food habits of panthers in southwest Florida. *The Journal of Wildlife Management*, **54**, 420-423.
- Maguire, C. M. and Grey, J. (2006) Determination of zooplankton dietary shift following a zebra mussel invasion, as indicated by stable isotope analysis. *Freshwater Biology*, **51**, 1310-1319.
- Mallick, S. A., Driessen, M. M. and Hocking, G. J. (2000) Demography and home range of the eastern barred bandicoot (*Perameles gunnii*) in south-eastern Tasmania. *Wildlife Research*, **27**, 103-115.
- Mansergh, I. (1983) The status, distribution and abundance of *Dasyurus maculatus* (tiger quoll) in Australia, with particular reference to Victoria. *Australian Zoologist*, **21**, 109-122.
- Mansergh, I. M., (1995) Spotted-tailed quoll. In: *Mammals of Victoria*: 51-52. P. W. Menkhorst (Ed.). Oxford University Press, Melbourne.
- Marker, L. L., Muntifering, J. R., Dickman, A. J., Mills, M. G. L. and Macdonald, D. W. (2003) Quantifying prey preferences of free-ranging Namibian cheetahs. *South African Journal of Wildlife Research*, **33**, 43-53.

- Marsden-Smedley, J. B., (1998) Changes in southwestern Tasmanian fire regimes since the early 1800s. *Papers and Proceedings of the Royal Society of Tasmania*, **132**, 15-29.
- Marsden-Smedley, J. B. and Kirkpatrick, J. B. (2000) Fire management in Tasmania's Wilderness World Heritage Area: Ecosystem restoration using Indigenous-style fire regimes? *Ecological Management & Restoration*, **1**, 195-203.
- Marshall, B. and Cosgrove, R. (1990) Tasmanian devil (*Sarcophilus harrisii*) scat-bone: signature criteria and archaeological implications. *Archaeology in Oceania*, **25**, 102-113.
- Maxwell, S., Burbidge, A. A. and Morris, K. 1996. *The 1996 Action Plan for Australian Marsupials and Monotremes*. Australasian Marsupial and Monotreme Specialist Group, IUCN Species Survival Commission, Gland, Switzerland.
- McCutchan, J. H., Lewis, W. M., Kendall, C. and McGrath, C. C. (2003) Variation in trophic shift for stable isotope ratios of carbon, nitrogen, and sulfur *Oikos*, **102**, 378-390.
- McHenry, C. R., Clausen, P. D., Daniel, W. J., Meers, M. B. and Pendharkar, A. (2006) Biomechanics of the rostrum in crocodilians: A comparative analysis using finite-element modeling. *The Anatomical Record Part A: Discoveries in Molecular, Cellular, and Evolutionary Biology*, **288A**, 827-849.
- McHenry, C. R., Wroe, S., Clausen, P. D., Moreno, K. and Cunningham, E. (2007) Supermodeled sabercat, predatory behavior in *Smilodon fatalis* revealed by high-resolution 3D computer simulation. *Proceedings of the National Academy of Sciences*, **104**, 16010-16015.
- McIlroy, J. C., (1995) Common wombat. In: *The Mammals of Australia*: 204-205. R. Strahan (Ed.). Australian Museum/Reed Books, Chatswood.
- McIlroy, J. C., (2008) Common wombat: *Vombatus ursinus*. In: *The mammals of Australia*. 3rd ed.: 206-208. S. Van Dyck and R. Strahan (Eds.). Reed New Holland, Sydney.
- McKenzie, A. A. (1990) Co-operative hunting in the black-backed jackal (*Canis mesomelas*) Schreber. PhD thesis, University of Pretoria, South Africa.

REFERENCES

- McKinney, M. L. (1997) Extinction vulnerability and selectivity: Combining Ecological and Paleontological Views. *Annual review of ecology and systematics*, **28**, 495-516.
- McPherson, F. J. and Chenoweth, P. J. (2012) Mammalian sexual dimorphism. *Animal Reproduction Science*, **131**, 109-122.
- McWilliams, R., Allen, J., Cosgrove, R. and Holdaway, S. (1999) Southern Forests Archaeological Project Data Base. *School of Archaeological and Historical Studies*, La Trobe University, Melbourne.
- Medina, E. and Minchin, P. (1980) Stratification of $\delta^{13}\text{C}$ values of leaves in Amazonian rain forests. *Oecologia*, **45**, 377-378.
- Meers, M. B. (2002) Maximum bite force and prey size of *Tyrannosaurus rex* and their relationships to the inference of feeding behavior. *Historical Biology: A Journal of Paleobiology*, **16**, 1-12.
- Menkhorst, P. (2001) *A field guide to the mammals of Australia*. Oxford University Press, Melbourne.
- Menzies, B. R., Renfree, M. B., Heider, T., Mayer, F., Hildebrandt, T. B. and Pask, A. J. (2012) Limited genetic diversity preceded extinction of the Tasmanian tiger. *PLoS ONE*, **7**, e35433.
- Merkle, J. M. J., Stahler, D. S. D. and Smith, D. S. D. (2009) Interference competition between gray wolves and coyotes in Yellowstone National Park. *Canadian Journal of Zoology*, **87**, 56-63.
- Millard, A., (2000) A model for the effect of weaning on nitrogen isotope ratios in humans. In: *Perspectives in amino acid and protein geochemistry*: 51-59. Oxford University Press, New York.
- Miller, W., Drautz, D. I., Janecka, J. E., Lesk, A. M., Ratan, A., Tomsho, L. P., Packard, M., Zhang, Y., McClellan, L. R. and Qi, J. (2009) The mitochondrial genome sequence of the Tasmanian tiger (*Thylacinus cynocephalus*). *Genome Research*, **19**, 213-220.

- Mills, M. and Mills, M. (1982) Factors affecting the movement patterns of brown hyaenas, *Hyaena brunnea*, in the southern Kalahari. *South African Journal of Wildlife Research*, **12**, 111-117.
- Mills, M. G. and Gorman, M. L. (2003) Factors affecting the density and distribution of wild dogs in the Kruger National Park. *Conservation Biology*, **11**, 1397-1406.
- Minagawa, M. and Wada, E. (1984) Stepwise enrichment of ^{15}N along food chains: further evidence and the relation between $\delta^{15}\text{N}$ and animal age. *Journal of The Geochemical Society and The Meteoritical Society*, **48**, 1135-1140.
- Mirón, M. L. L., Herrera, M. L. G., Ramírez, P. N. and Hobson, K. A. (2006) Effect of diet quality on carbon and nitrogen turnover and isotopic discrimination in blood of a New World nectarivorous bat. *Journal of Experimental Biology*, **209**, 541-548.
- Mittelbach, M. and Crewdson, M. (2005) *Carnivorous nights: on the trail of the Tasmanian tiger*. Villard, New York.
- Moazen, M., Curtis, N., O'Higgins, P., Jones, M. E. H., E., E. S. and Fagan, M. J. (2008) A novel assessment of the role of sutures in a lizard skull. *Proceedings of the Royal Society of London, Series B*, **276**, 39-44.
- Mokany, K., Friend, D., Kirkpatrick, J. and Gilfedder, L., (2006) Managing Tasmanian Native Pastures: a technical guide for graziers. Department of Primary Industries, Parks, Water and Environment, Tasmania.
- Moore, J. W. and Semmens, B. X. (2008) Incorporating uncertainty and prior information into stable isotope mixing models. *Ecology Letters*, **11**, 470-480.
- Moors, P. J. (1980) Sexual dimorphism in the body size of mustelids (Carnivora): the roles of food habits and breeding systems. *Oikos*, **34**, 147-158.
- Morris, K., (1996) The 1996 action plan for Australian marsupials and monotremes. A. A. Burbidge and S. Maxwell (Eds.). Wildlife Australia, Canberra.
- Morrison, J. C., Sechrest, W., Dinerstein, E., Wilcove, D. S. and Lamoreux, J. F. (2007) Persistence of large mammal faunas as indicators of global human impacts. *Journal of Mammalogy*, **88**, 1363-1380.

REFERENCES

- Muirhead, J. (1997) Two new early Miocene thylacines from Riversleigh, northwestern Queensland. *Memoirs of the Queensland Museum* , **41**, 367-378.
- Muirhead, J. and Archer, M. (1990) *Nimbacinus dicksoni*, a plesiomorphic thylacine (Marsupialia: thylacinidae) from Tertiary deposits of Queensland and the Northern Territory. *Memoirs of the Queensland Museum*, **28**, 203-221.
- Muirhead, J. and Wroe, S. (1998) A new genus and species, *Badjcinus turnbulli* (Thylacinidae: Marsupialia), from the late Oligocene of Riversleigh, northern Australia, and an investigation of thylacinid phylogeny. *Journal of Vertebrate Paleontology*, **18**, 612-626.
- Munday, P. L. (2004) Habitat loss, resource specialization, and extinction on coral reefs. *Global Change Biology*, **10**, 1642-1647.
- Muñoz-Durán, J. (2002) Correlates of speciation and extinction rates in the Carnivora. *Evolutionary ecology research*, **4**, 963-991.
- Murdoch, W. W. (1969) Switching in general predators: experiments on predator specificity and stability of prey populations. *Ecological Monographs*, **39**, 335-354.
- Murray, K. A., Rosauer, D., McCallum, H. and Skerratt, L. F. (2011) Integrating species traits with extrinsic threats: closing the gap between predicting and preventing species declines. *Proceedings of the Royal Society B: Biological Sciences*, **278**, 1515-1523.
- Murray, P. (1997) *Thylacinus megiriani*, a new species of thylacine (Marsupialia: Thylacinidae) from the Ongeva local fauna of central Australia. *Records of the South Australian Museum*, **30**, 43-61.
- Murray, P. and Megirian, D. (2000) Two new genera and three new species of Thylacinidae (Marsupialia) from the Miocene of the Northern Territory, Australia. *The Beagle, Records of the Museums and Art Galleries of the Northern Territory*, **16**, 145-162.
- Myers, T. J. (2001) Marsupial body mass prediction. *Australian Journal of Zoology*, **49**, 99-118.

- Nalla, R. K., Kinney, J. H. and Ritchie, R. O. (2003) Mechanistic fracture criteria for the failure of human cortical bone. *Nature Materials*, **2**, 164-168.
- Nathan, C. V., (1975) Quolls at Vaucluse. In: *Sydney Morning Herald*. Sydney.
- Nedin, C. (1991) The dietary niche of the extinct Australian marsupial lion: *Thylacoleo carnifex* Owen. *Lethaia*, **24**, 115-118.
- Newsome, A., Parer, I. and Catling, P. (1989) Prolonged prey suppression by carnivores—predator-removal experiments. *Oecologia*, **78**, 458-467.
- Newsome, S., Etnier, M., Kurle, C., Waldbauer, J., Chamberlain, C. and Koch, P. (2007) Historic decline in primary productivity in western Gulf of Alaska and eastern Bering Sea: isotopic analysis of northern fur seal teeth. *Marine Ecology Progress Series*, **332**, 211-224.
- Newsome, S. D., Koch, P. L., Etnier, M. A. and Auriolles-Gamboa, D. (2006) Using carbon and nitrogen isotope values to investigate maternal strategies in northeast Pacific otariids. *Marine Mammal Science*, **22**, 556-572.
- Nicol, S. C. and Andersen, N. A. (2007) The life history of an egg-laying mammal, the echidna. *Ecoscience*, **14**, 275-285.
- O'Connell, T. C., Hedges, R. E. M., Healey, M. A. and Simpson, A. H. R. W. (2001) Isotopic comparison of hair, nail and bone: modern analyses. *Journal of Archaeological Science*, **28**, 1247-1255.
- O'Leary, M. H. (1981) Carbon isotope fractionation in plants. *Phytochemistry*, **20**, 553-567.
- O'Leary, M. H. (1988) Carbon isotopes in photosynthesis. *Bioscience*, **38**, 328-336.
- Oakwood, M. (1997) The ecology of the northern quoll. PhD thesis, Australian National University, Canberra.
- Oelbermann, K. and Scheu, S. (2002) Stable isotope enrichment ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) in a generalist predator (*Pardosa lugubris*, Araneae: Lycosidae): effects of prey quality. *Oecologia*, **130**, 337-344.
- Olden, J. D., Poff, N. L. and Bestgen, K. R. (2008) Trait synergisms and the rarity, extirpation, and extinction risk of desert fishes. *Ecology*, **89**, 847-856.

REFERENCES

- Oldfield, C. C., McHenry, C. R., Clausen, P. D., Chamoli, U., Parr, W. C. H., Stynder, D. D. and Wroe, S. (2012) Finite element analysis of ursid cranial mechanics and the prediction of feeding behaviour in the extinct giant *Agriotherium africanum*. *Journal of Zoology*, **286**, 163-170.
- Owen, D. (2003) *Thylacine: the tragic tale of the Tasmanian tiger*. Allen & Unwin, Sydney.
- Owen, D. and Pemberton, D. (2005) *The Tasmanian devil: a unique and threatened animal*. Allen & Unwin, Sydney.
- Owen, W. H. and Thomson, J. A. (1965) Notes on the comparative ecology of the Common Brushtailed and Mountain Possums in eastern Australia. *The Victorian naturalist*, **82**, 216-217.
- Owens, I. P. F. and Bennett, P. M. (2000) Ecological basis of extinction risk in birds: Habitat loss versus human persecution and introduced predators. *Proceedings of the National Academy of Sciences*, **97**, 12144-12148.
- Owens, N. J. P. (1987) Natural Variations in ^{15}N in the marine environment. *Advances in marine biology*, **24**, 389-451.
- Paddle, R. (2000) *The Last Tasmanian tiger: the History and Extinction of the Thylacine*. Cambridge University Press, Cambridge.
- Paine, R. T. (1980) Food webs: linkage, interaction strength and community infrastructure. *Journal of Animal Ecology*, **49**, 667-685.
- Palomares, F. and Caro, T. (1999) Interspecific killing among mammalian carnivores. *The American Naturalist*, **153**, 492-508.
- Palomares, F., Ferreras, P., Fedriani, J. and Delibes, M. (1996) Spatial relationships between Iberian lynx and other carnivores in an area of south-western Spain. *Journal of Applied Ecology*, **33**, 5-13.
- Parr, W., Wroe, S., Chamoli, U., Richards, H., McCurry, M., Clausen, P. and McHenry, C. (2012) Toward integration of geometric morphometrics and computational biomechanics: New methods for 3D virtual reconstruction and quantitative analysis of Finite Element Models. *Journal of Theoretical Biology*, **301**, 1-14.

- Pate, F. D. and Noble, A. H. (2000) Geographic distribution of C₃ and C₄ grasses recorded from stable carbon isotope values of bone collagen of South Australian herbivores. *Australian Journal of Botany*, **48**, 203-207.
- Paterson, W., (1805) An animal of a truly singular and novel description. 21 April 1805. In: *Sydney Gazette and New South Wales Advertiser*: 3.
- Pearse, R. J. and Wapstra, J. E., (1988) Conservation and management of the eastern grey kangaroo, *Macropus giganteus*, of Tasmania. Unpublished report.
- Pech, R., Sinclair, A., Newsome, A. and Catling, P. (1992) Limits to predator regulation of rabbits in Australia: evidence from predator-removal experiments. *Oecologia*, **89**, 102-112.
- Pellis, S. M. and Officer, R. C. E. (1987) An analysis of some predatory behaviour patterns in four species of carnivorous marsupials (Dasyuridae), with comparative notes on the Eutherian cat *Felis catus*. *Ethology*, **75**, 177-196.
- Pemberton, D. (1988) Soil erosion between Birch Inlet and Elliott Bay, southwestern Tasmania. *Papers and Proceedings of the Royal Society of Tasmania*, **122**, 109-114.
- Pemberton, D., (1989) Land Systems of Tasmania Region 7 - Southwest. Department of Agriculture, Hobart.
- Pemberton, D., Gales, S., Bauer, B., Gales, R., Lazenby, B. and Medlock, K. (2008) The diet of the Tasmanian Devil, *Sarcophilus harrisii*, as determined from analysis of scat and stomach contents. *Papers and Proceedings of the Royal Society of Tasmania*, **142**, 13-22.
- Pemberton, D. and Renouf, D. (1993) A field-study of communication and social-behavior of the Tasmanian devil at feeding sites. *Australian Journal of Zoology*, **41**, 507-526.
- Peron, F. (1809) *A voyage of discovery to the Southern Hemisphere, performed by order of the Emperor Napoleon, during the years 1801, 1802, 1803, and 1804*. Richard Phillips, London.
- Peterson, B. J. and Fry, B. (1987) Stable isotopes in ecosystem studies. *Annual review of ecology and systematics*, **18**, 293-320.

REFERENCES

- Peterson, M., (1990) Distribution and Conservation of Huon Pine. Forestry Commission, Hobart.
- Petren, K. and Case, T. J. (1996) An experimental demonstration of exploitation competition in an ongoing invasion. *Ecology*, **77**, 118-132.
- Pierce, S. E., Angielczyk, K. D. and Rayfield, E. J. (2008) Patterns of morphospace occupation and mechanical performance in extant crocodilian skulls: a combined geometric morphometric and finite element modeling approach. *Journal of Morphology*, **269**, 840-864.
- Pierotti, R. and Annett, C., (1987) Reproductive consequences of dietary specialization and switching in an ecological generalist. In: *Foraging behavior*: 417-442. A. C. Kamil, J. R. Krebs and H. R. Pulliam (Eds.). Plenum Press, New York.
- Pimm, S. L. (1992) *The balance of nature?: ecological issues in the conservation of species and communities*. University of Chicago Press, Chicago.
- Pimm, S. L. (2002) *Food webs. 2nd Edition*. University of Chicago Press, Chicago.
- Plane, M. (1976) The occurrence of *Thylacinus* in Tertiary rocks from Papua New Guinea. *BMR Journal of Australian Geology and Geophysics*, **1**, 78-79.
- Polis, G. A., Myers, C. A. and Holt, R. D. (1989) The ecology and evolution of intraguild predation: potential competitors that eat each other. *Annual review of ecology and systematics*, **20**, 297-330.
- Polischuk, S. C., Hobson, K. A. and Ramsay, M. A. (2001) Use of stable-carbon and -nitrogen isotopes to assess weaning and fasting in female polar bears and their cubs. *Canadian Journal of Zoology*, **79**, 499-511.
- Pollock, A. (1999) Notes on status, distribution and diet of northern quoll *Dasyurus hallucatus* in the Mackay-Bowen area, mideastern Queensland. *Australian Zoologist*, **31**, 388-395.
- Porro, L. B. (2009) Cranial biomechanics in the early dinosaur *Heterodontosaurus*. PhD thesis, University of Cambridge, Cambridge.
- Post, D. M. (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology*, **83** 703-718.

- Power, M. E. (1990) Effects of fish in river food webs. *Science(Washington)*, **250**, 811-814.
- Pressey, R. L., Cabeza, M., Watts, M. E., Cowling, R. M. and Wilson, K. A. (2007) Conservation planning in a changing world. *Trends in ecology & evolution*, **22**, 583-592.
- Preuschoft, H. and Witzel, U. (2005) Functional shape of the skull in vertebrates: Which forces determine skull morphology in lower primates and ancestral synapsids? *The Anatomical Record Part A: Discoveries in Molecular, Cellular, and Evolutionary Biology*, **283A**, 402-413.
- Prowse, T. A. A., Johnson, C. N., Lacy, R. C., Bradshaw, C. J. A., Pollak, J. P., Watts, M. J. and Brook, B. W. (2013) No need for disease: testing extinction hypotheses for the thylacine using multi-species metamodels. *Journal of Animal Ecology*, **82**, 355-364.
- Prugh, L. R., Stoner, C. J., Epps, C. W., Bean, W. T., Ripple, W. J., Laliberte, A. S. and Brashares, J. S. (2009) The rise of the mesopredator. *Bioscience*, **59**, 779-791.
- Purvis, A., Gittleman, J. L., Cowlishaw, G. and Mace, G. M. (2000) Predicting extinction risk in declining species. *Proceedings of the Royal Society B: Biological Sciences*, **267**, 1947-1952.
- Quin, D. G. (1988) Observations on the diet of the southern brown bandicoot, *Isodon obesulus* (Marsupialia: Peramelidae), in southern Tasmania. *Australian Mammalogy*, **11**, 15-25.
- Quinn, J. L. and Kokorev, Y. (2002) Trading-off risks from predators and from aggressive hosts. *Behavioral Ecology and Sociobiology*, **51**, 455-460.
- Radloff, F. G. T. and Du Toit, J. T. (2004) Large predators and their prey in a southern African savanna: a predator's size determines its prey size range. *Journal of Animal Ecology*, **73**, 410-423.
- Rayfield, E. J. (2005) Aspects of comparative cranial mechanics in the theropod dinosaurs *Coelophysis*, *Allosaurus* and *Tyrannosaurus*. *Zoological Journal of the Linnean Society*, **144**, 309-316.

REFERENCES

- Rayfield, E. J. (2007) Finite element analysis and understanding the biomechanics and evolution of living and fossil organisms. *Annual Review of Earth and Planetary Sciences*, **35**, 541-576.
- Rayfield, E. J., Norman, D. B., Horner, C. C., Horner, J. R., Smith, P. M., Thomason, J. J. and Upchurch, P. (2001) Cranial design and function in a large theropod dinosaur. *Nature*, **409**, 1033-1037.
- Reynolds, H. (2012) *A history of Tasmania*. Cambridge University Press. Melbourne.
- Reynolds, J. C. and Aebischer, N. J. (1991) Comparison and quantification of carnivore diet by faecal analysis: a critique, with recommendations, based on a study of the Fox *Vulpes vulpes*. *Mammal Review*, **21**, 97-122.
- Rho, J. Y., Hobatho, M. C. and Ashman, R. B. (1995) Relations of mechanical properties to density and CT numbers in human bone. *Medical Engineering and Physics*, **17**, 347-355.
- Richards, M. P., Pettitt, P. B., Trinkaus, E., Smith, F. H., Paunović, M. and Karavanić, I. (2000) Neanderthal diet at Vindija and Neanderthal predation: the evidence from stable isotopes. *Proceedings of the National Academy of Sciences*, **97**, 7663-7666.
- Rishworth, C., McIlroy, J. and Tanton, M. (1995) Diet of the Common Wombat, *Vombatus Ursinus*, in Plantations of Pinus Radiata. *Wildlife Research*, **22**, 333-339.
- Ritchie, E. G. and Johnson, C. N. (2009) Predator interactions, mesopredator release and biodiversity conservation. *Ecology Letters*, **12**, 982-998.
- Robertson, D. I. and Duncan, F., (1991) Distribution and Conservation of Deciduous Beech. Forestry Commission Tasmania and Department of Parks, Wildlife and Heritage, Hobart.
- Robson, S. K. and Young, W. G. (1990) A comparison of tooth microwear between an extinct marsupial predator, the Tasmanian tiger *Thylacinus cynocephalus* (Thylacinidae) and an extant scavenger, the Tasmanian devil *Sarcophilus harrisii* (Dasyuridae, Marsupialia). *Australian Journal of Zoology*, **37**, 575-589.

- Root, R. B. (1967) The niche exploitation pattern of the Blue-gray gnatcatcher. *Ecological Monographs*, **37**, 317-350.
- Rosenzweig, M. L. (1966) Community structure in sympatric Carnivora. *Journal of Mammalogy*, **47**, 602-612.
- Ross, C. F. (2005) Finite element analysis in vertebrate biomechanics. *The Anatomical Record Part A: Discoveries in Molecular, Cellular, and Evolutionary Biology*, **283A**, 253-258.
- Roth, J. D. and Hobson, K. A. (2000) Stable carbon and nitrogen isotopic fractionation between diet and tissue of captive red fox: implications for dietary reconstruction. *Canadian Journal of Zoology*, **78**, 848-852.
- Roth, J. D., Marshall, J. D., Murray, D. L., Nickerson, D. M. and Steury, T. D. (2007) Geographic gradients in diet affect population dynamics of Canada lynx. *Ecology*, **88**, 2736-2743.
- Roy, D. M., Hall, R., Mix, A. C. and Bonnicksen, R. (2005) Using stable isotope analysis to obtain dietary profiles from old hair: A case study from Plains Indians. *American Journal of Physical Anthropology*, **128**, 444-452.
- Rucklidge, G. J., Milne, G., McGaw, B. A., Milne, E. and Robins, S. P. (1992) Turnover rates of different collagen types measured by isotope ratio mass spectrometry. *Biochimica et Biophysica Acta (BBA) - General Subjects*, **1156**, 57-61.
- Ruggiero, R. (1991) Prey selection of the lion (*Panthera leo* L.) in the Manovo-Gounda-St. Floris National Park, Central African Republic. *Mammalia*, **55**, 23-34.
- Ruxton, G. D. and Moody, A. L. (1997) The ideal free distribution with kleptoparasitism. *Journal of Theoretical Biology*, **186**, 449-458.
- Salmi, T., Maata, A., Antilla, P., Ruoho-Airola, T. and Amnell, T. (2002) *Detecting trends of annual values of atmospheric pollutants by the Mann-Kendall Test and Sen's slope estimates - The Excel template application Makesens*. Finnish Meteorological Institute. Helsinki, Finland.
- Sarich, V., Lowenstein, J. M. and Richardson, B. J., (1982) Phylogenetic relationships of *Thylacinus cynocephalus* (Marsupialia) as reflected in comparative serology.

REFERENCES

- In: *Carnivorous Marsupials*: 445-476. M. Archer (Ed.). Royal Zoological Society of New South Wales, Sydney.
- Savolainen, P., Leitner, T., Wilton, A. N., Matisoo-Smith, E. and Lundeberg, J. (2004) A detailed picture of the origin of the Australian dingo, obtained from the study of mitochondrial DNA. *Proceedings of the National Academy of Sciences of the United States of America*, **101**, 12387-12390.
- Schaller, G. B. (1967) *The deer and the tiger. A study of wildlife in India*. University of Chicago Press, Chicago.
- Schleser, G. H. and Jayasekera, R. (1985) $\delta^{13}\text{C}$ variations of leaves in forests as an indication of reassimilated CO_2 from the soil. *Oecologia*, **65**, 536-542.
- Schneider, U., Pedroni, E. and Lomax, A. (1996) The calibration of CT Hounsfield units for radiotherapy treatment planning. *Physics in Medicine and Biology*, **41**, 111-124.
- Schoener, T. W. (1969) Models of optimal size for solitary predators. *American Naturalist*, **103**, 277-313.
- Schoener, T. W. (1971) Theory of feeding strategies. *Annual review of ecology and systematics*, **2**, 369-404.
- Schwartz, M. W., Iverson, L. R., Prasad, A. M., Matthews, S. N. and O'Connor, R. J. (2006) Predicting extinctions as a result of climate change. *Ecology*, **87**, 1611-1615.
- Seamster, V. A. (2010) Consequences of woody plant encroachment for mammalian predators PhD, University of Virginia, Charlottesville.
- Sechrest, W. (2003) Global diversity, endemism, and conservation of mammals. PhD, University of Virginia, Charlottesville.
- Sen, P. K. (1968) Estimates of the regression coefficient based on Kendall's tau. *Journal of the American Statistical Association*, **63**, 1379-1389.
- Sergio, F., Caro, T., Brown, D., Clucas, B., Hunter, J., Ketchum, J., McHugh, K. and Hiraldo, F. (2008) Top predators as conservation tools: ecological rationale, assumptions, and efficacy. *Annual review of ecology, evolution, and systematics*, **39**, 1-19.

- Sergio, F., Newton, I. A. N., Marchesi, L. and Pedrini, P. (2006) Ecologically justified charisma: preservation of top predators delivers biodiversity conservation. *Journal of Applied Ecology*, **43**, 1049-1055.
- Service, S. E., (1990) History of emergency events issue I. July 1990. State Emergency Service, Hobart.
- Settle, G. A. (1978) The quiddity of Tiger Quolls. *Australian Natural History*, **19**, 165-169.
- Shaffer, L. C. (1971) Specializations in the feeding behaviour of gulls and other birds. PhD thesis, Oxford University, Oxford.
- Sharland, M. (1963) *Tasmanian Wildlife*. Melbourne University Press. Victoria.
- Sih, A., Englund, G. and Wooster, D. (1998) Emergent impacts of multiple predators on prey. *Trends in ecology & evolution*, **13**, 350-355.
- Skerratt, L. F., Skerratt, J. H., Banks, S., Martin, R. and Handasyde, K. (2004) Aspects of the ecology of common wombats (*Vombatus ursinus*) at high density on pastoral land in Victoria. *Australian Journal of Zoology*, **52**, 303-330.
- Sleightholme, S., (2011) International Thylacine Specimen Database. DVD. Stephen Sleightholme, London.
- Smith, B. N. and Brown, W. V. (1973) The Kranz syndrome in the Gramineae as indicated by carbon isotopic ratios. *American Journal of Botany*, **60**, 505-513.
- Smith, B. N. and Epstein, S. (1971) Two categories of $^{13}\text{C}/^{12}\text{C}$ ratios for higher plants. *Plant physiology*, **47**, 380-384.
- Smith, M., (1982) Review of the Thylacine (Marsupialia, Thylacinidae). In: *Carnivorous Marsupials*: 237-253. M. Archer (Ed.). Sydney: Royal Zoological Society of New South Wales.
- Sotiropoulos, M., Tonn, W. and Wassenaar, L. (2004) Effects of lipid extraction on stable carbon and nitrogen isotope analyses of fish tissues: potential consequences for food web studies. *Ecology of Freshwater Fish*, **13**, 155-160.
- Stefen, C. (1999) Tooth enamel structure of some Australian carnivorous marsupials. *Alcheringa*, **23**, 111-132.
- Steffen, W. and Scientific, C. (2009) Australia's biodiversity and climate change.

REFERENCES

- Stenhouse, M. J. and Baxter, M. S., (1979) The uptake of bomb ^{14}C in humans. In: *Radiocarbon dating*: 324-341. R. Beger and H. E. Suess (Eds.). Berkeley: University of California Press.
- Strahan, R. (1995) *The mammals of Australia*. Reed Books. Sydney.
- Strait, D. S., Grosse, I. R., Dechow, P. C., Smith, A. L., Wang, Q., Weber, G. W., Neubauer, S., Slice, D. E., Chalk, J., Richmond, B. G., Lucas, P. W., Spencer, M. A., Schrein, C., Wright, B. W., Byron, C. and Ross, C. F. (2010) The structural rigidity of the cranium of *Australopithecus africanus*: Implications for diet, dietary adaptations, and the allometry of feeding biomechanics. *The Anatomical Record: Advances in Integrative Anatomy and Evolutionary Biology*, **293**, 583-593.
- Strait, D. S., Weber, G. W., Neubauer, S., Chalk, J., Richmond, B. G., Lucas, P. W., Spencer, M. A., Schrein, C., Dechow, P. C., Ross, C. F., Grosse, I. R., Wright, B. W., Constantino, P., Wood, B. A., Lawn, B., Hylander, W. L., Wang, Q., Byron, C., Slice, D. E. and Smith, A. L. (2009) The feeding biomechanics and dietary ecology of *Australopithecus africanus*. *Proceedings of the National Academy of Sciences USA*, **106**, 2124-2129.
- Suess, H. E. (1955) Radiocarbon concentration in modern wood. *Science*, **122**, 415-417.
- Switalski, T. A. (2003) Coyote foraging ecology and vigilance in response to gray wolf reintroduction in Yellowstone National Park. *Canadian Journal of Zoology*, **81**, 985-993.
- Szalay, E. S., (1982) A new appraisal of marsupial phylogeny and classification. In: *Carnivorous Marsupials*: 621-640. M. Archer (Ed.). Royal Zoological Society of New South Wales, Sydney.
- Tabari, H., Somee, B. S. and Zadeh, M. R. (2011) Testing for long-term trends in climatic variables in Iran. *Atmospheric Research*, **100**, 132-140.
- Tanner, J. B., Durmont, J. B., Sakai, S. T., Lundrigan, B. L. and Holekamp, K. A. Y. E. (2008) Of arcs and vaults: the biomechanics of bone-cracking in spotted hyenas (*Crocuta crocuta*). *Biological Journal of the Linnean Society*, **95**, 246-255.

- Tanner, Z. and Hocking, G. J. (2001) *Status and Management of the Forester Kangaroo in Tasmania, 2000*. Nature Conservation Branch, Department of Primary Industries, Water and Environment, Hobart.
- Taylor, R. J. (1986) Notes on the diet of the carnivorous mammals of the Upper Henty River Region, western Tasmania. *Papers and Proceedings of the Royal Society of Tasmania*, **120**, 7-10.
- Temple, S. A. (1987) Do predators always capture substandard individuals disproportionately from prey populations? *Ecology*, **68**, 669-674.
- Templer, P. H., Arthur, M. A., Lovett, G. M. and Weathers, K. C. (2007) Plant and soil natural abundance $\delta^{15}\text{N}$: indicators of relative rates of nitrogen cycling in temperate forest ecosystems. *Oecologia*, **153**, 399-406.
- Terborgh, J., Lopez, L., Nunez, P., Rao, M., Shahabuddin, G., Orihuela, G., Riveros, M., Ascanio, R., Adler, G. H. and Lambert, T. D. (2001) Ecological meltdown in predator-free forest fragments. *Science*, **294**, 1923-1926.
- The Animals and Birds Protection Act Amendment Act 1928 (19 Geo V, No. 51). Tasmanian Numbers Acts, Tasmania.
- Theil, H. (1950) A rank-invariant method of linear and polynomial regression analysis.
- Thexton, A. J. and Hiimae, K. M. (1977) A radiographic and electromyographic study of snapping and biting in the opossum. *Archives of Oral Biology*, **22**, 303-308.
- Thiemann, G. W., Iverson, S. J., Stirling, I. and Obbard, M. E. (2011) Individual patterns of prey selection and dietary specialization in an Arctic marine carnivore. *Oikos*, **120**, 1469-1478.
- Thomas, C. D., Cameron, A., Green, R. E., Bakkenes, M., Beaumont, L. J., Collingham, Y. C., Erasmus, B. F. N., De Siqueira, M. F., Grainger, A. and Hannah, L. (2004) Extinction risk from climate change. *Nature*, **427**, 145-148.
- Thomas, R. H., Schaffner, W., Wilson, A. C. and Pääbo, S. (1989) DNA phylogeny of the extinct marsupial wolf. *Nature*, **340**, 465-467.
- Thomason, J. J. (1991) Cranial strength in relation to estimated biting forces in some mammals. *Canadian Journal of Zoology*, **69**, 2326-2333.

REFERENCES

- Thomason, J. J., (1995) To what extent can the mechanical environment of a bone be inferred from its internal architecture. In: *Functional morphology in vertebrate paleontology*: 249-263. J. J. Thomason (Ed.). Cambridge University Press, Cambridge.
- Thompson, C. M. and Gese, E. M. (2007) Food webs and intraguild predation: community interactions of a native mesocarnivore. *Ecology*, **88**, 334-346.
- Thresher, R. W. and Saito, G. E. (1973) The stress analysis of human teeth. *Journal of biomechanics*, **6**, 443-449.
- Tieszen, L. L., Boutton, T. W., Tesdahl, K. G. and Slade, N. A. (1983) Fractionation and turnover of stable carbon isotopes in animal tissues: Implications for $\delta^{13}\text{C}$ analysis of diet. *Oecologia*, **57**, 32-37.
- Todd, S., Ostrom, P., Lien, J. and Abrajano, J. (1997) Use of biopsy samples of humpback whale (*Megaptera novaeangliae*) skin for stable isotope ($\delta^{13}\text{C}$) determination. *Journal of Northwest Atlantic Fishery Science*, **22**, 71-76.
- Travouillon, K., Legendre, S., Archer, M. and Hand, S. (2009) Palaeoecological analyses of Riversleigh's Oligo-Miocene sites: implications for Oligo-Miocene climate change in Australia. *Palaeogeography, Palaeoclimatology, Palaeoecology*, **276**, 24-37.
- Triggs, B. (1996). The Wombat: Common Wombats in Australia. University of New South Wales Press, Sydney.
- Tsafnat, N. and Wroe, S. (2010) An experimentally validated micromechanical model of a rat vertebra under compressive loading. *Journal of anatomy*, **218**, 40-60.
- Tseng, Z. J. (2008) Cranial function in a late Miocene *Dinocrocuta gigantea* (Mammalia: Carnivora) revealed by comparative finite element analysis. *Biological Journal of the Linnean Society*, **96**, 51-67.
- Turnbull, W. D. (1970) Mammalian masticatory apparatus. *Fieldiana: Geology*, **18**, 149-356.
- Van Auken, O. W. (2009) Causes and consequences of woody plant encroachment into western North American grasslands. *Journal of Environmental Management*, **90**, 2931-2942.

- van der Merwe, N. J. and Medina, E. (1989) Photosynthesis and $^{13}\text{C}/^{12}\text{C}$ ratios in Amazonian rain forests. *Geochimica et Cosmochimica Acta*, **53**, 1091-1094.
- Van Valkenburgh, B. (2007) Deja vu: the evolution of feeding morphologies in the Carnivora. **47**, 147-163.
- Van Valkenburgh, B. and Hertel, F. (1998) The decline of North American predators during the Late Pleistocene. *Quaternary paleozoology in the Northern Hemisphere. Illinois State Museum Scientific Papers*, **27**, 357-374.
- Van Valkenburgh, B., Wang, X.M. & Damuth, J. (2004) Cope's rule, hypercarnivory, and extinction in North American canids. *Science*, **306**, 101-104.
- Vander Zanden, M. J., Cabana, G. and Rasmussen, J. B. (1997) Comparing trophic position of freshwater fish calculated using stable nitrogen isotope ratios ($\delta^{15}\text{N}$) and literature dietary data. *Canadian Journal of Fisheries and Aquatic Sciences*, **54**, 1142-1158.
- Vander Zanden, M. J., Chandra, S., Allen, B. C., Reuter, J. E. and Goldman, C. R. (2003) Historical food web structure and restoration of native aquatic communities in the Lake Tahoe (California–Nevada) basin. *Ecosystems*, **6**, 274-288.
- Vanderklift, M. A. and Ponsard, S. (2003) Sources of variation in consumer-diet $\delta^{15}\text{N}$ enrichment: a meta-analysis. *Oecologia*, **136**, 169-182.
- Vézina, A. F. (1985) Empirical relationships between predator and prey size among terrestrial vertebrate predators. *Oecologia*, **67**, 555-565.
- Vogel, J. C. and Van der Merwe, N. J. (1977) Isotopic evidence for early maize cultivation in New York State. *American Antiquity*, **42**, 238-242.
- Wake, D. B. and Vredenburg, V. T. (2008) Are we in the midst of the sixth mass extinction? A view from the world of amphibians. *Proceedings of the National Academy of Sciences*, **105**, 11466-11473.
- Wallach, A. D., Murray, B. R. and O'Neill, A. J. (2009) Can threatened species survive where the top predator is absent? *Biological Conservation*, **142**, 43-52.
- Walmsley, C. W., Smits, P. D., Quayle, M. R., McCurry, M. R., Richards, H. S., Oldfield, C. C., Wroe, S., Clausen, P. D. and McHenry, C. R. (2013) Why the

REFERENCES

- long face? The mechanics of mandibular symphysis proportions in crocodiles. *PLoS ONE*, **8**, e53873.
- Warwick, F., (1998) European Farming, Australian Pests: Agricultural Settlement and Environmental Disruption in Australia, 1800-1920. In: *Environment and History*: 129-143. The White Horse Press, United Kingdom.
- Watt, A. (1993) *Conservation status and draft management plan for Dasyurus maculatus and D. hallucatus in Southern Queensland*. Department of Environment and Heritage: 1–132. Sports and Territories, Queensland.
- Watts, D. (1993) *Tasmanian mammals: a field guide*. Peregrine Press, Kettering.
- Weijjs, W. A. and Hillen, B. (1985) Cross-sectional areas and estimated intrinsic strength of the human jaw muscles. *Acta Morphology Neerlandiae-Scandinavica*, **23**, 267-274.
- Werdelin, L. (1986) Comparison of skull shape in marsupial and placental carnivores. *Australian Journal of Zoology*, **34**, 109-117.
- Werner, E. E. and Peacor, S. D. (2003) A review of trait-mediated indirect interactions in ecological communities. *Ecology*, **84**, 1083-1100.
- West, J. B., Bowen, G. J., Cerling, T. E. and Ehleringer, J. R. (2006) Stable isotopes as one of nature's ecological recorders. *Trends in ecology & evolution*, **21**, 408-414.
- Wheless, C. R., (2009) *Wheless' Textbook of Orthopaedics*. Duke Orthopaedics, Durham.
- White, C. D., Longstaffe, F. J. and Law, K. R. (1999) Seasonal stability and variation in diet as reflected in human mummy tissues from the Kharga Oasis and the Nile Valley. *Palaeogeography, Palaeoclimatology, Palaeoecology*, **147**, 209-222.
- White, P. and Garrott, R. A. (1997) Factors regulating kit fox populations. *Canadian Journal of Zoology*, **75**, 1982-1988.
- Wiens, J. A. (1993) Fat times, lean times and competition among predators. *Trends in ecology & evolution*, **8**, 348-349.
- Wilmers, C. C., Crabtree, R. L., Smith, D. W., Murphy, K. M. and Getz, W. M. (2003a) Trophic facilitation by introduced top predators: grey wolf subsidies to

- scavengers in Yellowstone National Park. *Journal of Animal Ecology*, **72**, 909-916.
- Wilmers, C. C., Stahler, D. R., Crabtree, R. L., Smith, D. W. and Getz, W. M. (2003b) Resource dispersion and consumer dominance: scavenging at wolf-and hunter-killed carcasses in Greater Yellowstone, USA. *Ecology Letters*, **6**, 996-1003.
- Wilson, D. S. (1975) The adequacy of body size as a niche difference. *The American Naturalist*, **109**, 769-784.
- Wolff, J. (1892) *Das Gesetz der Transformation der Knochen*. Springer-Verlag. Berlin.
- Woodroffe, R. (2000) Predators and people: using human densities to interpret declines of large carnivores. *Animal Conservation*, **3**, 165-173.
- Woodroffe, R. and Ginsberg, J. R. (1998) Edge effects and the extinction of populations inside protected areas. *Science*, **280**, 2126-2128.
- Woodward, G., Ebenman, B., Emmerson, M., Montoya, J. M., Olesen, J. M., Valido, A. and Warren, P. H. (2005) Body size in ecological networks. *Trends in ecology & evolution*, **20**, 402-409.
- Wroe, S. (1996) *Muribacinus gadiyuli*, (thylacinidae: marsupialia), a very plesiomorphic thylacinid from the Miocene of Riversleigh, northwestern Queensland, and the problem of paraphyly for the Dasyuridae (marsupialia). *Journal of Paleontology*, **70**, 1032-1044.
- Wroe, S. (2001) *Maximucinus muirheadae*, gen. et sp. nov. (Thylacinidae: Marsupialia), from the Miocene of Riversleigh, north-western Queensland, with estimates of body weights for fossil thylacinids. *Australian Journal of Zoology*, **49**, 603-614.
- Wroe, S. (2002) A review of terrestrial mammalian and reptilian carnivore ecology in Australian fossil faunas and factors influencing their diversity: The myth of reptilian domination and its broader ramifications. *Australian Journal of Zoology*, **49**, 603-614.
- Wroe, S., (2003) Australian marsupial carnivores: an overview of recent advances in palaeontology. In: *Predators with pouches: the biology of carnivorous*

REFERENCES

- marsupials*: 102-123. M. E. Jones, C. Dickman and M. Archer (Eds.). CSIRO, Collingwood.
- Wroe, S. (2008) Cranial mechanics compared in extinct marsupial and extant African lions using a finite-element approach. *Journal of Zoology (London)*, **274**, 332-339.
- Wroe, S. and Archer, M., (2006) Origins and early radiations of marsupials. In: *Evolution and Biogeography of Australasian Vertebrates*. J. Merrick, M. Archer, G. M. Hickey and M. S. Y. Lee (Eds.). Auscipub Pty Ltd, Sydney.
- Wroe, S., Brammall, J. and Cooke, B. (1998) The skull of *Ekaltadeta ima* (Marsupialia, Hypsiprymnodontidae): an analysis of some marsupial cranial features and a re-investigation of propleopine phylogeny, with notes on the inference of carnivory in mammals. *Journal of Paleontology*, **72**, 738-751.
- Wroe, S., Clausen, P., McHenry, C., Moreno, K. and Cunningham, E. (2007) Computer simulation of feeding behaviour in the thylacine and dingo as a novel test for convergence and niche overlap. *Proceedings of the Royal Society B: Biological Sciences*, **274**, 2819-2828.
- Wroe, S., Ferrara, T. L., McHenry, C. R., Curnoe, D., Chamoli, U., Wroe, S., Ferrara, T. L., McHenry, C. R., Curnoe, D. and Chamoli, U. (2010) The craniomandibular mechanics of being human. *Proceedings of the Royal Society B: Biological Sciences*, **277**, 3579-3586.
- Wroe, S., Lowry, M. B. and Anton, M. (2008) How to build a mammalian super-predator. *Zoology*, **111**, 196-203.
- Wroe, S., McHenry, C. and Thomason, J. (2005) Bite club: comparative bite force in big biting mammals and the prediction of predatory behaviour in fossil taxa. *Proceedings of the Royal Society B: Biological Sciences*, **272**, 619-625.
- Wroe, S. and Milne, N. (2007) Convergence and remarkable constraint in the evolution of mammalian carnivore skull shape. *Evolution*, **61**, 1251-1260.
- Wroe, S. and Musser, A. (2001) The skull of *Nimbacinus dicksoni* (Thylacinidae: Marsupialia). *Australian Journal of Zoology*, **49**, 487-514.

- Wroe, S., Myers, T. J., Wells, R. T. and Gillespie, A. (1999) Estimating the weight of the Pleistocene marsupial lion, *Thylacoleo carnifex* (Thylacoleonidae: Marsupialia): implications for the ecomorphology of a marsupial super-predator and hypotheses of impoverishment of Australian marsupial carnivore faunas. *Australian Journal of Zoology*, **47**, 489-498.
- Yoneyama, T., Ohta, Y. and Ohtani, T. (1983) Variations of natural ^{13}C and ^{15}N abundances in the rat tissues and their correlation. *Radioisotopes*, **32**, 330-332.
- Yoshioka, T., Wada, E. and Hayashi, H. (1994) A stable isotope study on seasonal food web dynamics in a eutrophic lake. *Ecology*, **75**, 835-846.
- Young, M. T., Rayfield, E. J., Holliday, C. M., Witmer, L. M., Button, D. J., Upchurch, P. and Barrett, P. M. (2012) Cranial biomechanics of *Diplodocus* (Dinosauria, Sauropoda): testing hypotheses of feeding behaviour in an extinct megaherbivore. *Naturwissenschaften*, **99**, 637-643.
- Zimen, E. (1976) On the Regulation of Pack Size in Wolves. *Zeitschrift für Tierpsychologie*, **40**, 300-341.