

# **Food preferences of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*)**

Yvette Joanne Fenning

B.Mus

The University of Queensland

A thesis submitted in total fulfilment of the requirements of the degree  
of Master of Applied Science

March 2017

School of Medical and Applied Sciences

Centre for Environmental Management

CQ University

Rockhampton, Queensland, Australia

## Abstract

This program of research investigates food preferences of captive wombats when offered a selection of familiar, unfamiliar, natural and unnatural captive wombat diets. These diets reflect both current and future diet options for zoos and wildlife carers Australia wide.

A review of the research highlights a lack of knowledge of wombat food preferences and that feeding regimens for captive wombats may be inadequate. This was the first study of food preferences in captive southern hairy-nosed wombats.

The research was based at the Rockhampton Zoo Wombat Research Centre and involved a series of four food choice experiments. The foods offered included familiar and unfamiliar foods as well as natural and unnatural foods. All foods used throughout this program of research are routinely fed to captive wombats throughout Australian zoos.

The results showed this population of captive wombats, when offered a choice between their daily, pelleted feed, preferred pellets overall followed by natural grasses regardless of familiarity. The test food items had an impact on the amount of pellets consumed with carrot proving to be a disincentive to eating. Variables that may have influenced food choice were also considered, such as activity levels during feed, weather conditions and time of feeding trial. The results showed that active wombats consumed more food overall than sedentary wombats and were heavier in body weight. Temperature did not impact the amount of food consumed or the food choices made by this group of wombats. The time of the feeding trial did impact on the food choices with the wombats choosing to consume more pellets and less of the test foods the later the evening progressed.

These findings can be applied to captive management of wombats in zoos. Grass is recommended to form an essential part of all captive wombat diets due to the positive correlation it had with total food consumed. Carrot should be reconsidered as a popular food for wombats due to its negative correlation to total food consumed.

## Acknowledgements

This research higher degree candidature was supported by a Scholarship from the Australian Government's Research Training Program / Research Training Scheme. I gratefully acknowledge the financial support provided by the Australian Government.

This project was only made possible by the support of Rockhampton Zoo. Without wombats, a research facility and a management team supportive of the project, nothing would have happened. For this I am very grateful, as it is not me that has benefitted from this project, but the wombats.

I am enormously grateful to Prof. Dave Swain for taking me on board despite his better judgement. To my associate supervisor, Dr Tina McAdie I offer my thanks for saying the things that needed to be said, and perhaps for not saying some of the other things she may have been thinking. Dr Andrew Fenning has provided extensive and very timely support reviewing this document, among others. I also acknowledge the efforts of Prof. James Kinder who provided review and support in the early stage of thesis writing. I also acknowledge the contribution of Dr Brett Heath and Dr Rob Dixon who were involved in the early conception of this work.

I had assistance along the way from fellow wombat enthusiasts – all of whom deserve their name in lights: Lauren Thompson, Heather Campitell, Alexis Dupont and Tiffany Palmer. These amazing young women endured hours of wombat observations and all wore the scars of various misadventures with ill-contented wombats. Bruce in particular gave some harsh lessons, but I'm sure we are all richer for the experience.

Judy Couper and her assistant, Mackenzie Hansler, provided their expertise in drying and weighing all my samples. Thank you!

To my husband, Andrew – a medal of honour – for sticking with me especially through those dark days that were statistics coursework back in my first year and during the final death throws of final submission.

To my team and fellow keepers at the zoo - you guys are awesome. Thank you for all the times you picked up my slack.

To Graeme Strachan – thank you for allowing me this diversion over these last couple of years. I hope that I have done you, the wombats and the Council proud.

## Declaration of authorship and originality of thesis

I, the undersigned author, declare that all of the research and discussion presented in this thesis is original work performed by the author. No content of this thesis has been submitted or considered either in whole or in part, at any tertiary institute or university for a degree or any other category of award. I also declare that any material presented in this thesis performed by another person or institute has been referenced and listed in the reference section.

13/07/2017

Signature

Date

## Copyright statement

I, the undersigned author of the thesis, state that this thesis may be copied and distributed for private use and study, however, no chapter or materials of this thesis, in whole or in part, can be copied, cited or reprinted without the prior permission of the author and /or any reference fully acknowledged.

13/07/2017

Signature

Date

## Table of Contents

<b>Abstract</b> .....	<b>ii</b>
<b>Acknowledgements</b> .....	<b>iii</b>
<b>Declaration of authorship and originality of thesis</b> .....	<b>v</b>
<b>Copyright statement</b> .....	<b>vi</b>
<b>List of Tables</b> .....	<b>viii</b>
<b>List of Figures</b> .....	<b>viii</b>
<b>Introduction and Overview</b> .....	<b>1</b>
<b>Chapter 1</b> .....	<b>4</b>
<b>Literature Review</b> .....	<b>4</b>
Physiological adaptations.....	6
Behavioural adaptations.....	7
<b>Chapter 2</b> .....	<b>29</b>
<b>Methodology</b> .....	<b>29</b>
Experiment 1 – Unnatural/Familiar .....	34
Experiment 2 – Natural/Unfamiliar.....	41
Experiment 3 – Natural/Familiar.....	43
Experiment 4 – Unnatural/Unfamiliar food .....	43
<b>Chapter 3</b> .....	<b>46</b>
<b>Magnitude and frequency of food preferences of captive southern hairy-nosed wombats and the implications for captive management of this species</b> .....	<b>46</b>
Abstract .....	46
Introduction.....	47
Methods.....	49
Results.....	55
Discussion.....	70
Acknowledgements .....	79
References.....	80
<b>Chapter 4</b> .....	<b>82</b>
<b>Food intake and activity in captive southern hairy-nosed wombats</b> .....	<b>82</b>
Abstract .....	82
Introduction.....	83
Methods.....	87
Results.....	92
Discussion.....	98
Acknowledgements .....	100
References.....	101
<b>Chapter 5</b> .....	<b>103</b>
<b>Food intake relative to time of day in captive southern hairy-nosed wombats</b> .....	<b>103</b>
Abstract .....	103
Introduction.....	104
Methods.....	104
Results.....	105
Discussion.....	115

<b>Chapter 6 .....</b>	<b>117</b>
<b>Summary conclusions.....</b>	<b>117</b>
<b>References .....</b>	<b>125</b>

Andrew Fer

Deleted: 1

Andrew Fer

Deleted: 1

Andrew Fer

Deleted: 1

## List of Tables

Table 2.1: Experimental food choices .....	37
Table 3.1: Percentage of trials wombats selected to eat the test foods .....	55
Table 3.2: Percentage of trials consumption of test food exceeded consumption of pellets .....	55
Table 3.3 – Grams per minute consumed .....	65
Table 5.1: Correlation coefficient values for graphs presented in Figure 5.3.....	104

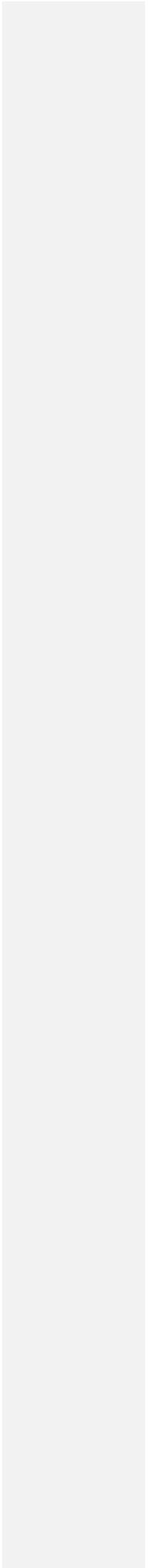
## List of Figures

Figure 1.1: Previous and current distribution of three wombat species.....	4
Figure 2.1: Layout of Rockhampton Zoo Wombat Research Facility.....	29
Figure 2.2: Wombats 3, 4 and 5 in the feeding pen during the 2-week pre-study acclimatisation period .....	31
Figure 2.3: Demonstrating the view from the elevated observation point, the Excel spread sheet being used to record data and the weather station used to collect temperatures and humidity values at the beginning of each feeding period .....	33
Figure 2.4: Wombat 4 lying down in the feeding pen during an early evening feeding period. This image also shows the positioning of the food bowls .....	33
Figure 2.5: Kangaroo grass sprouting .....	38
Figure 2.6: Kangaroo grass at growth stage used for feeding to wombats .....	39
Figure 2.7: Guinea grass clumps growing in the regular enclosures.....	40

Figure 2.8: Guinea grass freshly picked and fed out to wombat during a feeding period .....	40
Figure 2.9: Experiment 4 feeding period showing sweet potato in the feed bowl .....	41
Figure 3.1: Placement of feed bowls within feeding pen .....	50
Figure 3.2: Combined total foods consumed .....	52
Figure 3.3: Mean total combined foods .....	53
Figure 3.4: Mean grams consumed per experiment .....	54
Figure 3.5: Total consumed per feeding trial (Experiment 1) .....	56
Figure 3.6: Ratio of carrot consumed relative to pellets .....	57
Figure 3.7: Total foods consumed per feeding trial (Experiment 2) .....	58
Figure 3.8: Ratio of kangaroo grass consumed relative to pellets .....	58
Figure 3.9: Total foods consumed per feeding trial (Experiment 3) .....	59
Figure 3.10: Ratio of guinea grass consumed relative to pellets .....	60
Figure 3.11: Total foods consumed per feeding trial (Experiment 4) .....	61
Figure 3.12: Ratio of sweet potato consumed relative to pellets .....	61
Figure 3.13: Total test foods and average pellets consumed by each wombat...	62
Figure 3.14: Total foods consumed by all wombats for all experiments .....	63
Figure 3.15: Mean grams per minute consumed .....	65
Figure 3.16: Average foods consumed and average temperature .....	66
Figure 3.17: Average foods consumed and average humidity .....	66
Figure 4.1: Location of feeding bowls within the feeding pen .....	86
Figure 4.2: Percentage of time all wombats spent performing each behaviour ...	90

Figure 4.3: Time spent performing active vs sedentary behaviours for all wombats during all experiments.....	91
Figure 4.4: Ratio of active compared to sedentary behaviours per wombat (values greater than one indicate the wombat was more active than sedentary) .....	92
Figure 4.5: Correlation between average combined total foods consumed and ratio of activity.....	92
Figure 4.6: Correlation between average combined total foods consumed and ratio of activity for 30% most active wombats and 30% most sedentary wombats ( $r=0.65$ ) .....	93
Figure 4.7: Average amount of food consumed by the 30% most active and 30% most sedentary wombats.....	94
Figure 4.8: Mean body weights of active and sedentary wombats.....	94
Figure 5.1: Average pellets consumed correlated with time of day .....	103
Figure 5.2: Average test foods consumed correlated with time of day .....	103
Figure 5.3: Pellets and test food consumption correlated to time of day per experiment .....	104
Figure 5.4: Average pellet intake correlated to time of day for each wombat for each experiment.....	106
Figure 5.5: Average test food intake correlated to time of day for each wombat for each experiment.....	107
Figure 5.6: Wombat 2, average amount of pellets consumed correlated with time of day .....	108
Figure 5.7: Wombat 2, average amount of test foods consumed correlated with time of day .....	109
Figure 5.8: Wombat 5, average amount of pellets consumed correlated with time of day .....	110

Figure 5.9: Wombat 5, Total amount of test foods consumed correlated with time of day .....	110
Figure 5.10: Wombat 6, average amount of pellets consumed correlated with time of day .....	111
Figure 5.11: Wombat 6, average test foods consume correlated with time of day .....	111
Figure 5.12: Wombat 9, average pellets consumed correlated with time of day.....	113
Figure 5.13: Wombat 9, average test foods consumed correlated with time of day .....	113



## Introduction and Overview

Rockhampton Zoo houses 12 wombats (11 southern hairy-nosed wombats and one common wombat). These wombats are fed a diet of macropod pellets, lucerne chaff and root vegetables on a daily basis and observations of their feeding choices indicates the wombats have food preferences. The dietary intake as a result of available food options and subsequent food choices may create health issues and may contribute to poor reproductive rates (Woolnough 1998). To provide a platform to begin to understand the above phenomena in captive SHNWs this study was conceived to develop a basic understanding of the food preferences of a captive population of SHNWs.

Initial informal management records of daily food intake weights and fortnightly wombat weights provided important background information on what is common or normal for this captive population. In addition to the dietary records, historical notes on behaviours and group dynamics as well as records of animal movements from one enclosure to another have been kept since the wombats arrived at Rockhampton Zoo in 2001. This behavioural information helped inform management decisions to predict how each individual wombat may cope with different stressors. These data inform husbandry and animal handling techniques to enhance the overall welfare of the animals.

The aim for this thesis is to establish a research understanding of the dietary preferences of captive southern hairy-nosed wombats and to establish a methodology for studying such. The research results will provide a more formal and detailed quantitative assessment of the behavioural food preferences associated with the dietary intake of the captive wombats.

Nine captive southern hairy-nosed wombats (*Lasiorchinus latifrons*) housed at the Rockhampton Zoo Wombat Research Centre were used as the primary focus of this research. Two southern hairy-nosed wombats were excluded from this study as they were housed in a separate area of the zoo, away from the Wombat Research Centre. These two wombats were the zoo's public display animals whose living conditions differed from the Research Centre wombats. Including

these two animals would have introduced uncontrollable variables, such as the effects of disturbance by zoo visitors and differences in enclosure/den design, to this research, thus they were excluded.

The Rockhampton Zoo Wombat Research Centre was established in 2001 as a joint venture between Rockhampton Zoo and CQ University, QLD Parks and Wildlife Service and CQ Fertility Centre. Since its inception the centre has been the focus of two PhD studies, both through The University of Queensland and one masters project through CQ University. The last research project to be conducted at the facility concluded in January 2010. All three of these studies focused on reproduction and/or behaviour (Druery 2004; Hogan 2010; Descovich 2012). None of the studies assessed diet, nutrition or food preferences.

Chapters three and four of this thesis are presented as papers for submission for publication in scientific journals. As such there is a some repetition between the methods described in Chapter Two and the introduction sections of Chapters Three and Four. Chapter 5 presents data collected but not discussed in Chapters 3 and 4 of the dissertation.

The overall goals of this project were

1. To determine how much of the test food item was consumed
2. To determine how many times the test food item was selected
3. To determine how many times the consumption of the test food item exceeded consumption of pellets.
4. To establish if the test food items influenced the consumption of pellets
5. To determine if food preference is affected by variables such as rate of consumption and environmental conditions
6. To assess differences in physical activity levels between wombats.
7. To establish if activity level alters feeding behaviour.

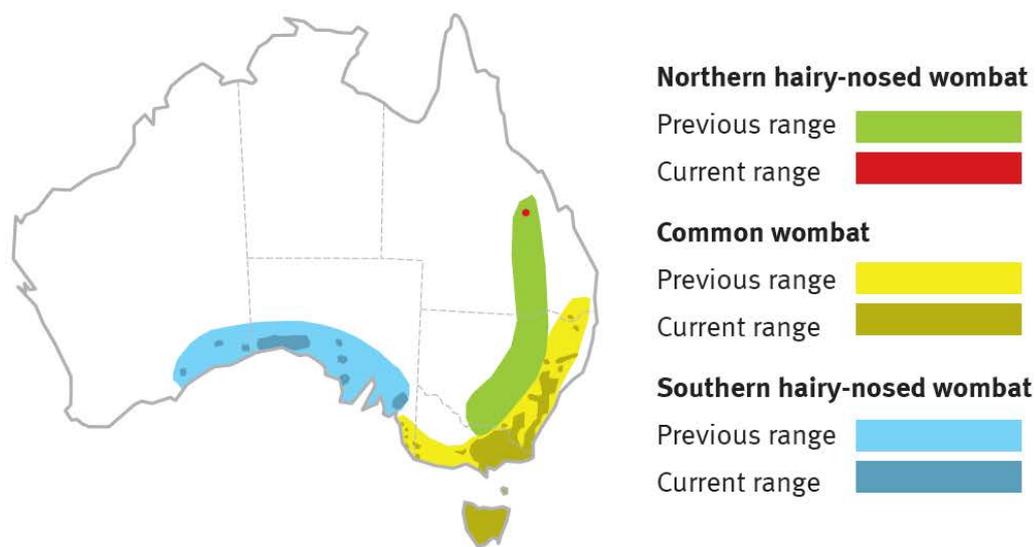
8. Does food choice make a wombat behave differently.
9. To determine if total food intake was affected by time of day
10. To determine if time of day affected food choice

## Chapter 1

### Literature Review

#### *Wombats of Australia*

There are three extant species of wombat in two genera, the common wombat (*Vombatus ursinus*), the southern hairy-nosed wombat (*Lasiorhinus latifrons*) and northern hairy-nosed wombat (*Lasiorhinus krefftii*). Figure 1 depicts the distribution of all three species.



**Figure 1.1: Previous and current distribution of three wombat species**

The northern hairy-nosed wombat (NHNW) is classified as a critically endangered species (IUCN 2017). It is among the 10 most endangered mammals on Earth (Taggart and Robinson 2008; Horsup 2012). The total population of near 200 NHNWs is being managed under a recovery program designed by Dr Alan Horsup and animals have been managed so that they reside in two separate physical locations to safeguard the species from natural disasters (Horsup 2004). As the population remains small, it is not considered ethical or safe to bring NHNWs into a captive environment for closer study.

The survival of the NHNW will depend upon breeding success and habitat protection. As wombats have a very slow rate of reproduction (approximately one joey every 3 years (Triggs 2009)), it is difficult to predict how long it will be before any NHNWs can be used for captive breeding programs. Due to the fragility of the NHNW population most captive research has been conducted on the NHNWs' closest genetic relative – the southern hairy-nosed wombat (SHNW).

The research conducted for this thesis will focus on captive southern hairy-nosed wombats (*Lasiorhinus latifrons*, hereafter referred to as SHNW).

Australia has a harsh environment with greater ambient temperatures than exists on other continents of the world (Thom 2002). It is the second driest continent on the planet. At least two thirds of it is classified as arid, or semiarid and receives less than 500 mm of average yearly rainfall (Thom 2002). The native flora and fauna of Australia has developed both behavioural and physiological adaptations to survive in these conditions. Wombats are no exception to other plants and animals in this regard. All 3 wombat species have developed extensive adaptive traits, which allows them to survive in some of the most arid environments in Australia (Triggs 2009).

Wombats are large, burrowing, hindgut fermenting, grazing herbivores (Hume 1999). Hume (1999) suggested the low number of large burrowing mammal species could be due to the large amount of energy required to construct a burrow and the potential problems of burrow stability as the size and diameter of the animal increases. Most burrowing mammals are small (5-8 kg) omnivores/carnivores that consume a more nutrient dense diet, and consequently have sufficient energy to dig and maintain burrows (Hume 1999). The sheer size of the wombat (up to 40 kg) requires a large sized burrow for entry, which would require large amounts of bodily energy to construct. The wombat survives, however, on a diet with very little energy content (Hume 1999).

Wombats are grazing herbivores. Their large size indicates that they should devote large amounts of time grazing to consume enough food to provide them the bodily energy reserves they need to survive in their natural habitat.

Herbivorous grazers of a similar weight to wombats, eastern grey kangaroos, devote 10 to 18 hours per day grazing compared with 2 to 4 hours per day for wombats (Hume 1999).

Wombats have developed unique behavioural, anatomical and physiological traits to allow them to overcome the challenges created by such a low energy diet (Hume 1999). These adaptations can be broadly explained through four main features of wombat ecology which significantly impact upon cellular metabolism.

### **Physiological adaptations**

Firstly, wombats have the lowest basal metabolic rate (BMR) and maintenance energy requirements recorded of any marsupial, (Barboza 1989; Barboza and Hume 1992; Barboza and Hume 1992; Barboza 1993; Barboza 1993; Barboza, Hume et al. 1993). Secondly, they have an efficient masticatory system, which includes permanently growing teeth – a trait unique to wombats among the marsupials (Hume 1999). Thirdly, wombats have a digestive strategy of colonic fermentation and slow digesta passage (Hume 1999). The fourth ecological feature is the wombat's sedentary and semi-fossorial lifestyle (Finlayson, Shimmin et al. 2005).

### ***Low basal metabolic rate and low maintenance energy requirements***

Wombats have the lowest recorded basal metabolic rates (BMR) for any marsupial and, consequently, have one of the lowest recorded maintenance energy requirements of the marsupials (Hume 1999). Barboza, Hume and Nolan (1993) found plasma concentrations of thyroid hormones in wombats were the least recorded for any mammal and maintenance requirements for nitrogen were among the least of the marsupial herbivores, significantly less than eutherian herbivores (Barboza, Hume et al. 1993). This can be partially explained by the wombats' ability to recycle urea to the hindgut, thus reducing nitrogen excretion in the urine (Hume 1999).

### ***Mastication and dentition***

Wombats have a split in their upper lip, which allows their incisors efficient access to cut grass close to the ground. They also have continuously growing

teeth that wear in such a way as to maintain sharp edges that are capable of reducing the fibrous grasses consumed to a remarkably small particle size (Barboza 1989). Wells (1973) compared particle size in faeces of wombats and grey kangaroos both grazing in the same semi-arid area and found that the wombat faeces contained mostly small particles, while the kangaroo faeces contained mostly large particles. This efficient mastication of fibrous grasses greatly increases the surface area of the plant matter that passes into the fermentation chamber component of the colon, which allows for more efficient use of the fermentation process (Hume 1999). As wombats have such adaptations to digesting grasses, then this would suggest that grass is a better food option to be offered in a captive environment than artificial foods such as commercially made pellets or other foods that wombats in their natural habitat would not usually come into contact with (such as root vegetables).

### **Hindgut fermentation**

Hindgut fermentation is a physiological adaptation developed by large grazing herbivores. It allows for a slow rate of passage of the digesta, allowing more time for microbial fermentation in the colon. This facilitates herbivores to extract maximum nutritional benefit from a diet that is generally fibrous and of low nutritional value (Hume 1989). Typically, hindgut fermenters rely on efficient mastication of food to break the fibrous leaves and stems of the plants in their diet into small enough sizes to be digested.

With all these physiological adaptations to cope with the harsh climate and poor quality of the diet in their home range, wombats still need to have a lifestyle of extreme energy conservation to survive.

### **Behavioural adaptations**

Wombats are sedentary grazers. That is, they remain in a single home range for their lifetime. They do not migrate with the seasons or as a result of changes in rainfall in the region in which they reside. This is possibly due to the fact that they are dependent on burrows for thermal regulation (Finlayson, Shimmin et al. 2005). The bodily energy expended in building a burrow can be greater than the energy consumed in their diet of low quality grasses, so they cannot be building

new burrows regularly. Instead they devote their time into maintaining existing burrows, only building new ones should there be a need to vacate an existing burrow (Barboza 1989).

Due to their sedentary way of functioning, for a large part of the year wombats only have low quality and a limited quantity of grass available for their dietary intake. This suggests that they may have a mechanism for storing fat during the wet season to be utilised in the dry season when food is sparse (Evans 1999).

Finlayson et al (2005) reported that to cope with their poor diet, wombats have developed behavioural adaptations that are different to most other herbivorous grazers – the most significant being their sedentary lifestyle and fossorial nature (Finlayson, Shimmin et al. 2003; Finlayson, Shimmin et al. 2005). This previous study (Finlayson, Shimmin et al. 2005) paired with an earlier study by the same investigators (Finlayson, Shimmin et al. 2003) was important for delineating differences between the SHNW and other two wombat species.

The 2005 study site in South Australia supported a large population of wombats (72 individual SHNWs were caught during the 8 month study), which was surprising given the arid nature of the country and the large body size of the wombats (22-32 kg; Finlayson, Shimmin et al. 2005). This differed considerably from a study of common wombats, in which only five common wombats were located in a NSW forest of similar size but with considerably more forage available (Evans 1999). The SHNWs also had very little variation in the body weight over the course of the study. The wombats' ability to persist at these population densities in this semi-arid environment is evidence of their adaptation to this environment.

The SHNWs forage in similar areas to arid zone kangaroos. There could, therefore, be competition for food in these arid zones and one would expect both species should exhibit large home ranges during times of drought to meet their demands for energy and nutrition. The Finlayson, Shimmin et al. 2005 study, however, revealed SHNWs had a home range 10 times smaller than these kangaroos. They estimated the average home range of a SHNW at the study site to be about 4 ha and that it did not fluctuate seasonally or differ between males

and females. There was substantial overlap of home range between all the wombats studied, which indicates there is little competition for food resources among the wombats in their home range (Finlayson, Shimmin et al. 2005).

The wombats in the Finlayson, Shimmin et al. (2005) study also had minimal body energy expenditure. These animals resided for more than three quarters of their time underground. Nocturnal above-ground activity periods were of short duration (2-8 hours; Finlayson, Shimmin et al., 2005).

The mean total distance travelled per night by all wombats was 221 m. The greatest distance travelled in one night by a wombat was 872 m. “There was no significant difference in distance travelled between sex or season.” (Finlayson, Shimmin et al. 2005).

Home range estimates of SHNWs were smaller in the Finlayson et al 2005, than estimates of home range in NHNWs (Woolnough 1998). Males did not increase their home range during the breeding season, nor did lactating females expand their home range in search of greater quality food resources so as to adapt to the need for the greater energy requirements of lactation (Finlayson, Shimmin et al. 2005).

The focus of wombat social organisation in the wild seems to be centred around the warren (network of burrows), which can have one to 30 burrows (Treby 2005). Ten or more wombats often use the same warren simultaneously. Typically a wombat colony uses 10 to 20 warrens in a cluster, which can be spread over an area up to 1 km<sup>2</sup> (Treby 2005). While females have a greater preference for particular burrows than males, there has been no evidence of burrow ownership (Treby 2005).

Warren sharing is another behavioural adaptation that contributes to wombat's bodily energy conservation. If wombats were territorial in protecting their warren, then they would not only have to defend their warrens, but would also need to build new warrens each time they lost a warren defending the territory in which they reside.

Warren sharing has been found in previous studies of NHNWs and common wombats (Woolnough 1998; Evans 1999). The NHNW females were more likely to share territory with other females, while males of this species rarely shared territory in which they reside. In common wombats it seems that relatedness between individuals may influence the likelihood of sharing territory (Favreau, Jarman et al. 2009). In the 2005 study, involving SHNWs, Finlayson, Shimmin et al. (2005), found that both males and females shared warrens.

This previous study (Finlayson, Shimmin et al. 2005) also provided evidence that there were marked differences in individual animal habits. For example, one female used one warren only for the entire duration of the study (8 months) while another female used ten warrens at differing intervals throughout the study.

Wombats live in geographic areas where summer temperatures are often 35 to 45 °C with humidity of 2% to 5%, however, the corresponding temperatures in the burrows during the summer months are 10 to 27 °C with 60% to 70% humidity (Finlayson, Shimmin et al. 2005). Interestingly, Finlayson et al. (2005) found there was a noticeable preferred range in ambient temperature at which animals emerged from their warrens. This was between 6 and 18 °C (Finlayson, Shimmin et al. 2005). More needs to be learned about the impacts of temperature and humidity on SHNWs. For this reason, temperature and humidity recordings were included as part of this present study.

Grazing behaviour is also centred around the warren. Wombats in their natural habitat graze in a circular pattern around the burrow complex to produce a lawn or grazing halo of green plant shoots (Treby 2005).

### *Water use efficiency*

Another physiological adaptation the wombat has developed in the harsh climatic conditions of Australia is its ability to survive with very little water intake. Wombats have one of the least requirements for water among mammals, requiring just 25% of the water intake of the kangaroo (Triggs 2009). Residing for large amounts of time below ground in burrows helps the wombat with water retention and thermoregulation. Underground, the ambient temperature

of the dens remains low and relatively constant compared to the fluctuations of temperatures above ground, not only day to day, but also season to season (Triggs 2009).

### *Population distribution*

The SHNW populations are not evenly distributed in the areas in which they reside but rather reside in patchy distributions across their home range. Estimates indicate between 50,000 and 100,000 individuals reside in the South Australian portion of the Nullarbor Plain, but there are no estimates for the Western Australian portion of the Nullarbor Plain (Nicolson 2012). The Murray Lands population has experienced a 70% decrease since 2002 possibly due to drought and sarcoptic mange. The population estimate for this population is 10,000 to 15,000. Other small isolated populations throughout South Australia range in population size from 50 to 100 individuals (Nicolson 2012). Some wombat populations are at great risk of inbreeding, particularly the York Peninsula population (Nicolson 2012). Major threats to the wild SHNW include reduced range through conversion of suitable habitat to agricultural land, competition for grazing by domestic stock and rabbits (Taggart and Temple-Smith 2008), sarcoptic mange (kills 80%-90% of affected populations/groups; (Nicolson 2012)) and drought. Drought is a significant threat to successful reproduction as several studies have shown that wombats need a minimum of 3 years without drought to increase in population numbers and reproduction ceases during drought years (Triggs 2009; Nicolson 2012).

### *Nutrition and diet in the natural habitat*

Evans, Macgregor et al. (2006) studied the diet and feeding selectivity of wild common wombats (*Vombatus ursinus*). These investigators identified 35 plant species in the faecal pellets of wild common wombats. They determined the common wombats' diet consisted of 95% grass species that were eaten in similar proportions to the abundance in the habitat area, with a few species of grass being preferred by wombats and other plants being rejected from their diet. Twenty species of grass were eaten by wombats and the most abundant grasses in faecal pellets were great lignin content snow grasses (*Poa sieberiana* and *Poa labillardieri*). During the summer and autumn months *Microlaena stipoides* made

up a significant portion of the diet. The most favoured part of the plant consumed was the leaf (81%) with grass stem and sheath comprising 11% of the diet. Grass seed heads were also found in significant quantities (21%) in the faecal pellets during the summer months. Forbs formed less than 1% of the pellet material and there was no evidence of browse (trees or shrubs) detected in the diet. Evans et al. (2006) found feeding selectivity (and hence dietary niche breadth) varied seasonally. That is, wombats became less selective as plant diversity and abundance decreased seasonally. Conspicuous in the absence from faecal pellets were clover, subterranean plant parts (such as roots and tubers) and fungus, despite the presence of all these food items at the study site. This is of particular interest to the current study as root vegetables form a significant part of captive wombat diets, even though data from the wild suggests they avoid subterranean plants. The great proportion of leaf to sheath and stem parts consumed by the wombats more closely resembles the diet of macropodid grazers than that of eutherian grazers (Hume 1999). Also the larger than expected intake of seed heads during summer is suggestive of a smaller sized mammal with a greater metabolic rate and again indicative of the diet of macropodidae (Evans, Macgregor et al., 2006). Evans, Macgregor et al., (2006) also suggested that the ability of wombats to be selective with food items despite their broad facial morphology, may be due to their narrow incisor arcade when compared to similarly sized herbivores such as kangaroos and sheep (Evans, Macgregor et al. 2006).

Barboza and Hume (1992) undertook a study of wombats in their natural habitat consuming their natural winter diets, and captive wombats fed a high-fibre pelleted straw diet to determine digestion and digestive tract morphology in wombats. Two species of wombats were studied (*V. ursinus* and *L. latifrons*) as these two species come from contrasting habitats. They found *L. latifrons* (SHNW) grazed mainly on *Stipa nitida* in their natural habitat, which is a grass that exhibits a seasonal pattern of growth, increasing with the winter and spring rainfall and decreasing during the dry summer months (Barboza and Hume 1992). This differs from the diet of *L. krefftii* (NHNW) as the grasses consumed by NHNWs also have seasonal patterns of growth, but at different times of the

year – spring/summer – as that is when the wet season occurs. Tropical grasses tend to have less moisture content in the winter months (Woolnough 1998).

### *Reproductive physiology*

The reproductive physiology of the SHNW has been studied and reported in four published manuscripts - Taggart, Finlayson et al. 2007; Hogan 2010; Hogan, Phillips et al. 2010; Hogan, Phillips et al. 2010. Knowledge from previous research supports the following reproductive strategies:

- Wombats are monovular and polyoestrus and have an oestrous cycle of approximately 35 days and a gestation of 20 to 22 days (Taggart, Finlayson et al. 2007).
- Males are polygamous, females are monogamous (Taggart et al. 1998).
- The reproductive season is in the winter and spring months, however, breeding only occurs in years in which rainfall is plentiful and by implication when food is abundant (Hume 1999; Taggart, Finlayson et al. 2007; Triggs 2009; Hogan 2010; Hogan, Phillips et al. 2010; Hogan, Phillips et al. 2010).
- Reproduction seems to be both seasonal and opportunistic from the perspective that they mate when the conditions of their environment become favourable (Treby 2005).
- Weaning occurs in spring or early summer, as increased vegetation growth periods at this time are associated with the winter rainfall patterns of the arid and semi-arid zones of SA and south-eastern parts of WA.

### *The history of captive southern hairy-nosed wombats*

Historically, wombats have a very poor captive breeding rate. The Zoo and Aquarium Association of Australasia (ZAA) records show that Rockhampton Zoo has recorded the greatest success for breeding SHNWs in captivity, but even its results are modest. For the 12 adult wombats (four males and eight females) in 8 years the breeding program at Rockhampton Zoo has produced seven SHNW

joeys – only three of which have survived to maturity (Unpublished Rockhampton Zoo records, 2012). The previous three research projects conducted at Rockhampton Zoo Wombat Research Centre focused on reproduction and/or behaviour. None of those studies have resulted in successfully increasing fecundity after the knowledge gained in this research was utilized.

As fecundity is likely linked to food resources (Woolnough and Foley, 2002), perhaps it is time to take a more holistic approach, examining all aspects of captive wombat welfare, including diet preferences, nutritional information and behavioural adaptations to captivity. It would therefore follow that better food resources (quality, quantity, food type) would allow for better health outcomes, a measure of which could be improved breeding success.

The SHNWs in captivity in Australia are managed by a national studbook, which is held and updated by the national species coordinator, currently Mr Gert Skipper of Adelaide Zoo. The previous species coordinator, Dr Nicolson, listed in the 2012 annual report that the only concern/challenge to this population is continued lack of breeding success (Nicolson 2012).

According to this report SHNWs seldom breed in captivity and despite careful comparisons of successful and unsuccessful husbandry regimens there seems to be no easy modifications to husbandry which may improve this species poor reproductive performance (Nicolson 2012).

Many zoo wombats have a diurnal lifestyle with dens that are not only above ground but also have no temperature regulation (personal observation). This diurnal lifestyle is very different from the lifestyle they would have in a natural environment. Dr Nicolson stated that while these factors are not proven to affect reproductive performance, environmental conditions in which captive wombats are managed cannot be discounted as contributing to the relatively poor reproductive rate compared with wombats that reside in their natural environment (Nicolson 2012).

The captive SHNW population in Australia is 55 animals: 18 males and 36 females. Since 2015 only two joeys have been born. In that same time frame

three captive animals have died. The species coordinator stated that this population needs a breakthrough in reproductive husbandry and/or continued supplementation with captured wild animals (Skipper 2015). The decreasing population of captive wombats raises some ethical questions with regard to animal welfare. Some animal welfare groups contend, that if animals are reproductively impaired, then welfare is compromised (Nicolson 2012).

#### *Research projects conducted at Rockhampton Zoo*

Druery (2004) performed a series of experiments that focused on assisted reproductive techniques. These studies do not have great relevance to the current project, so will not be elaborated on in this thesis. It should, however, be noted that no live offspring resulted from the assisted reproductive techniques that were used in this research (Druery 2004). While the reasons for this require further investigation, effects of captivity such as confined living conditions, frequent and invasive human contact and unnatural diet cannot be disregarded as possible contributing factors to this failure to produce young.

Hogan (2010) measured physical activity of captive SHNWs through video surveillance and radio telemetry. Twelve months of continuous monitoring yielded the following results; 69.9% sleeping, 8.8% lying resting, 5.2% feeding, 5.2% exploring, 4.3% stereotyping, 2.5% sitting at rest, 1.7% digging, 1.4% foraging, 0.4% being handled, 0.3% sun-basking, 0.2% grooming and 0.1% courtship/mating. Daily patterns of activity showed a strong circadian cycle with greater nocturnal and lesser diurnal activity (Hogan, Phillips et al. 2009; Hogan, Johnston et al. 2010).

Hogan (2010) reported that the time devoted to feeding, sleeping and stereotyping had a seasonal variation. “Feeding and stereotyping were negatively correlated with ambient temperature and humidity, whilst being positively associated with night-time length; the inverse was true for sleeping. Ambient temperature exerted its largest effect on time spent feeding; feeding times decreased by 3.1 minutes per 1°C above 13°C and compared to spring, feeding times were reduced by 41% in summer (Hogan, Phillips et al., 2009; Hogan, Johnston et al., 2010).”

Hogan (2010) found that male reproductive function was influenced seasonally, being maximal in winter (June-August). Female receptivity occurred at night and oestrus lasted 13 hours. Hogan recorded elevated progesterone concentrations in six of eight females. From these six, 23 luteal phases, and 12 follicular phases of oestrous cycles as well as 12 complete oestrous cycles were assessed and there was a mean length of 20.9, 11.6 and 31.8 days, respectively. (Hogan, Phillips et al. 2010; Hogan, Phillips et al. 2010).

Wild wombats have greater freedom to explore and forage in new areas within their local environment compared to the limited opportunities offered for captive wombats. This lack of freedom for captive wombats may result in a reduction of expression of normal behaviours and the development of stereotypies. Stereotypies are associated with poor animal welfare and most animal welfare standards require the captive animal to have access to or the opportunity to express natural behaviours. Hogan (2010) investigated the use of environmental enrichment using food treats and olfactory stimulants. Using 24-hour video surveillance Hogan recorded a 333% increase in foraging behaviour and a 13% increase in exploratory behaviour for wombats that were kept in an enriched environment. This enrichment did not have any effect on the amount of time devoted stereotyping, but Hogan believes that animal welfare was still enhanced citing the increase in wombat-specific behavioural expression and diversity (Hogan, Johnston et al. 2010).

Hogan et al. (2011) assessed 42 food treats as a means of administering glitter as a feed ingredient to be later used as a faecal pellet marker to identify individual wombats housed in a group setting. Of these feed ingredients, 6 of the 42 were accepted by the wombats as palatable and thus consistently consumed as part of the diet. These were, golden syrup with horse pellets, golden syrup with weet-bix, pitted dates, honey with kangaroo pellets, Nutrigel with rolled oats and strawberry sauce with rolled oats (Hogan et al., 2011). There were marked individual differences in food treat preferences between wombats with each wombat preferring 2 or more of the 42 food choices offered. The significance of these differences were not closely examined during that study, though it's possible there was an energy density driver for these choices. Hogan et al.

(2011) described the need to interchange the food treat used due to the wombats losing their appetite for the items.

In a PhD project conducted at the Rockhampton Zoo Wombat Research Centre by Descovich (2012), the following variables were assessed: auditory laterality, response to the presence of faeces from conspecifics and different species, group size effects on vigilance and amount of physical activity, space allowance behavioural effects, and seasonal and environmental influences on body temperature and glucocorticoids in this captive population of SHNWs.

The results of Descovich's (2012) study have made significant improvements to the knowledge of captive wombat management. For example, Descovich found that enclosure size had significant effects on wombat behaviour. As enclosure size decreased, escape attempts and social conflict between animals increased (Descovich, Lisle et al. 2012). Group size also affected behaviour. Vigilance behaviour decreased as group size increased (Descovich, Lisle et al. 2012). These findings are consistent with those of studies with other social species such as the Tibetan antelope, *Pantholops hodgsoni* (Lian, Zhang et al. 2010), but differs from studies of the common wombat (*V. ursinus*) where vigilance behaviour increased when conspecifics were grazing in close proximity (Favreau, Jarman et al. 2009).

All three of these studies have made significant contributions to the knowledge about captive wombats, and it is the purpose of this study to further build the knowledge base. In summary, it has been established that the poor breeding rate of wombats in captivity is the result of one or more limitations in management or insufficient animal welfare standards. Druery (2004) attempted to improve this breeding rate using assisted reproductive techniques, but had no success. This strengthens the understanding that captive management, and thus animal welfare standards are insufficient for SHNWs. Hogan (2010) and Descovich (2012) investigated environmental and behavioural influences of the captive environment. Both their studies have provided evidence that there are aspects of captive animal management that have had negative effects on animal welfare, such as enclosure size, group size and human interaction. Previous research

indicated that there are both deficiencies and excesses of certain nutrients in diets that can lead to decreased fecundity in many animals and that poor fecundity is correlated with poor animal welfare standards (Johnston, Grune et al. 2006). In no studies have there been investigations of associations between food preferences, nutritional requirements and animal welfare in the captive SHNW.

Fecundity is likely associated with food resources in wombats (Johnson, 1991; Woolnough, 1998). Native grasses, once abundant in the habitat of NHNWs, are being displaced by introduced pasture grasses (Woolnough and Foley, 2002). These authors suggest that nutritional analysis of the pasture resources available to free-ranging NHNWs is crucial to the management of this critically endangered grazing herbivore.

In the Woolnough and Foley (2002) study, it was found that even though sedges and forbs were more nutritious than the native grasses, the NHNWs did not consume these plant components. This may be due to plant secondary compounds/defences that make the plant unpalatable or toxic when consumed by wombats. The wombats were sustained on a combination of native grasses and an introduced grass, *Cenchrus ciliaris*, the latter being greater in nutritional value compared to the former.

The nutritional requirements of wombats have been studied and reported in several studies (Hume, 1989; Barboza and Hume, 1992; Barboza and Hume, 1992; Barboza, 1993; Barboza, 1993; Barboza, Hume et al., 1993; Hume, 1999). A few reports indicate the preferred species of grasses consumed by wild wombats (Barboza, 1993; Barboza, Hume et al., 1993; Woolnough, 1998), but a close examination of diets fed to captive wombats in zoos have little resemblance to that of their counterparts in their natural environments. For example, Barboza (1993), Triggs (2009) and Hume (1989) report that wild wombats choose a high fibre/low protein diet that is also low in copper and nitrogen. Information gathered from zoos Australia-wide indicates that captive diets for wombats are greater energy/greater protein feeds such as carrot, corn and lucerne chaff/hay compared to the native grasses consumed in the natural habitat (personal observation). An emailed survey provided to Australian Zoos

that hold SHNWs asked keepers to report observed food preferences in their SHNS. The zookeepers reported that wombats showed a preference for these food items when offered lower quality feeds, such as oaten hay, but that there was also a prevalence of obesity in captive wombat populations. The zoos also identified that captive wombats were offered very little opportunity to graze on fresh growing grasses. Many zoos housed wombats in enclosures that had a dirt, sand or concrete substrate floors, which allowed for easy cleaning of the enclosures, but restricted the wombats' ability to perform the natural behaviour of selective grazing. Some zoos allowed the wombats' partial access to grazing, while others provided grass in the form of fresh forage while other zoo animal caretakers fed no grasses at all.

There are a couple of factors that have lead to the tradition of feeding captive wombats a pelleted diet with constituents similar to that of the forages in pastures rather than their natural diet of grass.

Firstly, wombats in captivity are usually housed in enclosures much smaller than their usual home range would be in their natural habitat. This means that it can be difficult to maintain a grass substrate in their enclosures due to over grazing and trampling. Secondly, a pelleted diet is a convenient substitute to grass as it stores easily for long periods of time and it is easy to present to the wombats in a bowl or trough. While the bulk of most captive wombat diets are pellets, many zoos also supplement this with fruits and vegetables such as carrots, corn, apples, sweet potato and peanuts. There is no evidence to suggest that wombats in their natural habitat would feed on these items, yet zoos continue to offer these dietary components to wombats.

The low basal metabolic rate (BMR) and low energy requirements of SHNWs has lead to a tendency towards obesity in captive situations (Treby 2005). Therefore a low energy, low protein diet based on grass and/or low quality food is recommended (Treby 2005). Food items such as fruits and vegetables ferment more rapidly than grasses. Given the wombats' low metabolic rate and slow passage of digestion, such food items cannot be economically utilized and may

lead to health problems such as obesity and systematic calcification (Booth, 1999).

### *Nutrition and animal welfare*

Zoo accreditation boards such as the Zoo and Aquarium Association (ZAA) require that high standards of husbandry and animal welfare be maintained and requires zoos to take an active role in conservation. Zoos can fulfil this obligation through participation in research that has a conservation benefit to the species (Rees 2005).

Animal welfare is defined through three states; physical state, mental state and naturalness. Physical state is measured by the animal's physiology and can include body condition, injury, illness or any symptom that impairs survival or reproduction. Mental state is measured by behavioural observations and the presence or absence of stereotypies, increased heart rate, or cortisol concentrations in the blood, among others. Naturalness refers to the animal's ability to fulfil its natural needs and desires (Unknown 2012).

Nutrition is increasingly becoming an important consideration of animal welfare. Researchers studying predominantly large grazing herbivores, have been asking the questions "Is feeding a diet that meets nutritional needs sufficient? What is adequate nutrition, and what is the standard for adequate animal nutrition and consequently adequate animal welfare?" (Vellalba et al., 2010). Vellalba et al., 2010, have studied intensive livestock production systems (mainly sheep, cattle and goats), which historically have feeding regimens based on a monotonous "nutritionally balanced" premixed feed, or a pasture monoculture. Such feeding regimens, using uniformly formulated foods, are based on the assumption that if nutrient requirements are met for the average animal of the herd considering their stage of productivity (lactating, growing, etc.), then adequate nutrition is achieved for the herd as a whole. This method, however, does not allow for individual differences in morphology and physiology. Gordon et al. (1996) found that sheep and goats foraged the same pastures with differing efficiencies due to variations in their dental arcade. Konarzewski and Diamond (1994) found foraging could be affected by individual differences in organ mass and

macronutrient metabolization (Konarzewski and Diamond 1995). Even sheep and cattle of the same sex, breed and age will vary in their preferences for food, with some preferring foods high in energy, others preferring low energy foods, and individuals varying diet selections day by day (Provenza et al. 1996: Early and Provenza, 1998: Scott and Provenza 1998: Atwood et al. 2001). Villalba et al. (2010) proposed that offering a diverse diet offered many benefits to the animals including increased resistance to disease, ability to self-medicate using the natural medicinal properties of many plants, allowing for individual differences in morphology and physiology, and a decrease in fear and stress. This theory is supported by earlier studies by Bechara et al. (1995) who found that the amygdala and hippocampus contained neuronal networks that were involved in diet selection and fear. Bechara, Tranel et al. (1995) and Carlstead and Shepherdson (2000) studying both wild and farm animals found that chronic stress was decreased with environmental enrichment that allowed animals access to flexible foraging behaviour (Carlstead and Shepherdson 2000).

Diet choice has been shown to improve the welfare of livestock through the facilitation of natural foraging behaviour. The same may be true for non-livestock captive animals.

There has been a trend recently to study food preferences and animal-plant interactions with regard to the livestock industry. Studies abound in which there has been research on sheep, goats, cattle and chickens and their food preferences/choices (Arnold et al. 1980, Everitt et al. 1981, Hutson and van Mourik, 1981, Forbes and Kyriazakis, 1995, Villalba and Provenza, 1999,). This has led to a general consensus that variety in diet leads to increased animal welfare standards and to economic benefits for farmers of these animals (Atwood, Provenza et al. 2001). But how does this relate to the zoo industry and could the same benefits be found in captive zoo animals? A brief Scopus search for the key words “nutrition and livestock” yielded 1067 results, whereas the search for “nutrition and zoo” yielded only 198 results.

Consumers and customers are becoming increasingly aware of animal welfare. This is evidenced by the recent increase in demand for humane rearing,

transport and slaughter of farm animals. The term animal welfare has been subject to much debate and scrutiny, but is generally agreed to mean the meeting of physical, mental and natural states of animals. One of the first requirements of animal welfare is adequate nutrition (Manteca, Villalba et al. 2008). Manteca et al. (2008) “submit that in many cases domestic animals are provided with diets that, even when abundant and nutritious, are not necessarily adequate to foster the welfare of animals.” Total mixed rations, or monoculture pastures often contain an excess of nutrients, have nutrient imbalances and/ or contain toxins that adversely influence animal welfare (Manteca, Villalba et al., 2008). Many of these premixed diets are also designed to be palatable and able to be stored for long periods (i.e. contain many preservatives) and are not necessarily designed with the best nutritional needs of the animal in mind (Manteca, Villalba et al., 2008). The diets are a relatively convenient, economically inexpensive and easily stored way to feed large numbers of stock for food production (e.g. beef, lamb, pork, chicken, eggs) or animal performance (e.g. dog/horse racing) while still providing adequate nutrition to the animals. Food intake and preferences, however, are dependent on individual differences between animals. Even closely related animals have a marked variation in relative needs for nutrients and tolerance to toxins (Manteca, Villalba et al., 2008).

It has been determined through various scientific studies that animals possess nutritional wisdom (Villalba and Provenza, 2007). That is, they can achieve homeostasis through diet selection as a function of their physiological state across space and time (Villalba and Provenza, 2007). Additionally, studies also provide evidence that feeding varied diets had an economic advantage (Atwood, Provenza et al. 2001). Animals fed a premixed diet consumed more food than animals offered the same foods as an individual choice, but did not gain weight faster (Atwood, Provenza et al., 2001). Food costs per day, therefore, were greater for animals fed the premixed ration than for those offered the choice of foods (Kyriazakis and Oldham, 1993; Manteca, Villalba et al., 2008).

The Manteca, Villalba et al. (2008) study drew the following three conclusions – (1) animals can more efficiently meet their individual needs for macronutrients when offered a choice among dietary ingredients than when constrained to a

single diet, even if it is nutritionally balanced; (2) transient food aversions compound the inefficiencies of a single mixed diet by depressing intake, even among animals suited to that nutritional profile; and (3) alternative feeding practices may allow producers to efficiently capitalize on the agency of animals, thus reducing illness and improving performance.

What was once considered proper grazing management – rotational grazing at low stock densities – may have trained generations of livestock to “eat the best and leave the rest” thus inadvertently accelerating a decrease in biodiversity and an increase in the abundance of less desirable plant species (Provenza, Villalba et al. 2003). Provenza, Villalba et al. 2003, therefore suggest that offering a variety of dietary choices allows the uniqueness of each plant and animal to be expressed and manifest.

Rees (2005) stated that much of the research conducted by zoos is not suitable for publication in mainstream academic journals as it is oftentimes conducted in unnatural conditions with small sample sizes. But Provenza et al. (2003) provided a counter argument that 20<sup>th</sup> century statistics have placed too much emphasis on assessing the response of the “average” animal to a treatment. Statistics may have advanced our ability to conduct experiments, but it has caused variation among individuals to be treated as errors rather than an opportunity to explore these differences in more detail. It could be erroneous to emphasize means and populations over individuals and variation. Nature and evolutionary processes rarely conform to means (Provenza et al. 2003).

#### *Assessing food preferences in small sample sizes*

Aside from environmental survey information on the diet of wild wombats, no study has assessed any combination of food choice or preference in wild or captive wombats. The lack of any baseline data provides some significant challenges even in determining if wombats will be curious enough to engage with and consume a variety of food items during a choice-based experiment. Food choice and preference experiments for most mammalian grazing herbivores typically assess large herd based eating patterns in paddock environments. Whilst this shows a basic similarity to the captive aspect of most

zoo enclosures, the much smaller enclosure size found in zoos makes this a very different comparison. The unique animals and their diet preferences, enclosure size and number of individuals create the need for compromises in study design when dealing with experiments in zoo settings.

A successful repetitive food choice experiment was conducted in a zoo housed population of moose (*Alces alces*) (Clauss et al, 2013). Moose are a grazing herbivore with specific dietary/nutritional needs, which are similar to the dietary/nutritional needs of wombats. This study (Clauss et al, 2013) fed a baseline zoo pelleted diet with access to enclosure browse followed by a sequence of five (in two Moose) or four (in the remaining two Moose) experimental diets (Clauss et al, 2013). By measuring food intake in grams, digestible energy and calculation of dry matter consumed, a repeated measures ANOVA assessment of the animals' intake and choice for the experimental diets showed that the animals fed freshly cut grass preferred this as a feed compared to other forms of grass hay and ultimately maintained a better weight (Clauss et al, 2013). The experimental design showed that the animals avoided grass hay compared to freshly cut grass and browse in the one week on and off feeding choice experiment (Clauss et al, 2013). This was an interesting finding and shows that even short periods of different feeding regimes under unusual experimental design constraints, produced significant results.

A captive koala (*Phascolarctos cinereus*) food choice experiment used a behavioural scan sampling based assessment of food preference and consumption of different types of eucalypt browse (Higgins et al, 2011). A Latin Square Design utilizing four different feeding canisters with nine different Eucalypt choices were presented over two four-week blocks with trials conducted on four consecutive days per week (Higgins et al, 2011). A random mix of non-study eucalypt browse was provided for the other three days in the week for the duration of the four-week study blocks (Higgins et al, 2011). The koalas were observed under three broad behaviour categories; eating, sitting and other (all other activities such as sleeping, walking and climbing). Immediately following feeding of the food choice, the koalas were observed for the first hour with observations recorded on the minute. This experiment

produced significant choice preferences (percentage of time spent eating and magnitude of consumption of the chosen eucalypt species) in all of the koalas with some individual differences observed (Higgins et al, 2011). Whilst this study provides support for a small study design of a food choice experiment with repeated sampling of individual subjects, the data acquisition could have been improved by recording more specific and defined behaviours and by presenting a binary choice of only two *Eucalyptus* species at a time. Another strength of this experiment was the noted individual presentation of each Koala's food preference with significant global trends observed across all of the subjects' yet also significant individual food preferences (Higgins et al, 2011). Thus, the study was able to clearly infer both group and individual behavioural food preferences and maintained a level of unfamiliar and novel eucalypt choices. Unfortunately, only male koalas were used which provides a level of limitation of the potential outcomes. Koalas are currently the closest living relative to the wombat and potentially provide some insight on designing a similar food choice experimental design.

Repetitive, two-choice and multiple-choice preference feeding have also been conducted successfully in cotton-top tamarins (Fernandez et al, 2004) and goats (De et al, 2002). A paired food choice experiment of seven different food items showed both individual variation and generalized group trends in food preference of captive tamarins (Fernandez et al, 2004). Repetitive trials of three two-choice presentations of flavoured straw pellets in goats, showed selectivity and increased preferences for pellets flavoured with grassy aromatic extracts (Fernandez et al, 2004). This was maintained even after repetitive exposure to the choice items showing that repetition and lack of novelty failed to impact upon the significant outcomes. These repeated choice trials were utilized to improve statistical validity of the observed behaviours and food consumption related to preference.

A limitation of many studies conducted in zoos and similar captive environments are the small population sizes that don't exist in agricultural or bench science based research. Most zoos only maintain relatively small population groups of animals showing significant heterogeneity (age and gender) which can impact

upon the number of replicates and degree of significance in comparative experimental studies. As long as the lower number of subjects is accounted for by combining both quantitative and qualitative study design, basic and descriptive statistical analyses and individual and group observations, then valid outcomes can be obtained. A recent study examining the nutritional drivers of moose food intake using a food choice experimental design, published significant findings using only six animals of different ages and gender (two adult males of differing ages, two pregnant females of similar age and two juveniles of equivalent age) (Felton et al, 2016). In this case, statistical power was maximized by using a mixed model design with the dietary treatment assessed as a within-subjects effect and the controlling factor (order of presentation of diets) was the between-subjects effect. Similar small subject numbers were utilized in food preference experiments in tamarins (five animals) (Fernandez et al, 2004), koalas (four adult male animals) (Higgins et al, 2011) moose (four adult animals) (Clauss et al, 2013) and goats (twelve animals) (De et al, 2002). Statistical significance and individual variation was determined through the use of repeated exposure trials to more quantitatively establish food consumption and preferences. If the limitations of small samples sizes are clearly established at the beginning of the study and enough repetitive trials conducted, together with individual subject observations then groups of as little as four animals can generate valid results. The study by Felton et al (2016) even established that captive moose (six animals) preferentially selected food items to maintain a nutritionally balanced diet showing that preference equates to good nutrition. Therefore, experiments using a diverse group of captive individuals, can effectively determine food choice of familiar and unfamiliar food items with four to ten subjects.

At present, zoos are bound by a commitment to four pillars – research, education, conservation and entertainment. Where once zoos were an entertaining family outing, modern culture demands high standards in animal welfare, a commitment to conservation, education programs and research. Australian Zoos who are members of the Australasian Zoo and Aquarium Association (ZAA) must

demonstrate their commitment to animal welfare through meeting strict accreditation standards which are based on ZAAs animal welfare position statement (Zoo and Aquarium Association, 2018). Within this position statement ZAA acknowledges that zoos face challenges in providing nutritionally balanced food for non-domestic species whose nutritional requirements are not yet fully understood, and that the physical form of the food items needs to be considered to aid in meeting the animal's psychological needs. For example, providing variable foraging opportunities for grazing herbivores may improve welfare states for these animals when compared to grazing herbivores fed a concentrated high energy food source presented in a trough once a day (Mellor and Beausoleil, 2015). The ZAA animal welfare position statement points out the Zoos can contribute to the building of knowledge regarding non-domestic animal nutritional requirements by participating in research focused on nutrition, taste and preference (Zoo and Aquarium Association, 2018). Rockhampton Zoo is an accredited institution under ZAAs governance, and thus must continually demonstrate a commitment to animal welfare. This research will benefit the Rockhampton Zoo, but will have larger implications for the zoo industry as the information gathered from this research is shared both within the zoo industry and with other animal industries.

#### *The need for zoo based research*

More zoos are becoming actively involved in recovery programs for endangered animals. Understanding and developing captive management techniques is crucial to the future survival of many species, including the SHNW. Zoos are in the unique position of being able to contribute significantly to this knowledge given with the zoo facilities there is already the infrastructure in place to house and care for non-domestic animals. Collaborations between zoos and the scientific community are a vital aspect of conservation research.

This study will contribute to the collective knowledge on captive SHNWs. It will aim to add significant knowledge to the understanding of dietary needs and choices of captive SHNWs and make further advances towards understanding adequate nutrition for wombats and, therefore, what role does diet choice has in animal welfare for this species.

There is a total of 58 SHNWs in captivity, though the scope of this research only allows for the close examination of nine of individuals at one institution. This may not be sufficient numbers for conventional statistics, so, rather than treat these animals as a population, this project aims to embrace their individuality in food preferences with the ultimate goal of improving animal welfare standards for wombats in captivity. Secondary to the main aims of this project is the goal of developing innovative methods and techniques for the close study of individuals within the population.

## Chapter 2

### Methodology

This methods section (Chapter 2) covers the full methods completed for the whole study. There are some aspects of this chapter which are repeated in the results chapters (Chapters 3, 4 and 5) with these chapters being presented as discreet papers. This series of experiments was designed to assess the food preferences of captive southern hairy-nosed wombats when offered a choice between their daily, pelleted feed and four experimental dietary options - familiar foods compared with unfamiliar and natural foods compared with unnatural foods. The Central Queensland University animal ethics committee approved the protocols of animal management in all four experiments. This is the first study of food preferences of captive southern hairy-nosed wombats.

For the purposes of this research project, the terms familiar, unfamiliar, natural and unnatural when used in the context of food choices are defined in the following ways -

- Familiar – foods that are routinely fed to the SHNWs at Rockhampton Zoo
- Unfamiliar – foods that the Rockhampton Zoo wombats have not been fed in the past 13 years of their recorded history in captivity
- Natural – foods that wombats in their natural habitat have been recorded as selecting, eating and, therefore, surviving on in their natural environment
- Unnatural – foods that wombats would not routinely have access to in their natural habitat and, therefore, do not naturally survive on in their natural habitat

The familiar foods chosen for this group of captive wombats were pellets (Riverina Macropod Pellets), carrots and the grass that grows naturally in their enclosures, *Megathyrsus maximus* (guinea grass). The unfamiliar foods were

sweet potato, and a native grass, *Themeda triandra* (Kangaroo grass). *Themeda triandra* is one of the preferred grass species of SHNWs in their natural habitat; hence it's selection for this study. While in their natural habitat SHNWs' most preferred grass species is *Stipa nitida*; this particular grass was not chosen, as seeds could not be sourced to propagate for the present study.

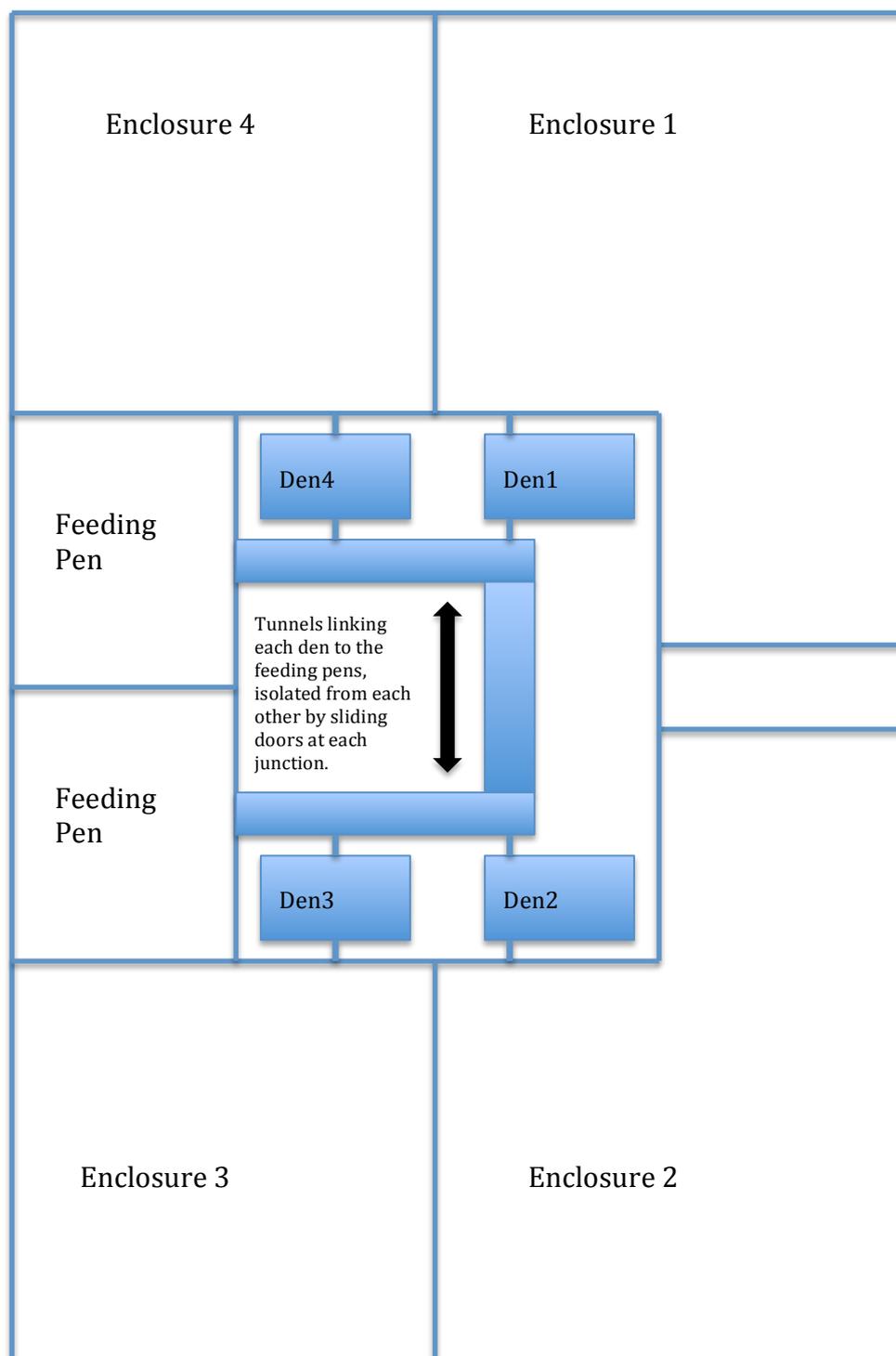
The wombats, with the exception of two that were born at the Rockhampton Zoo, were all captured as adults in South Australia and were bought to the Rockhampton Zoo in 2001. This group of wombats has not had access to *Themeda triandra* for at least 13 years. While it is not possible to ascertain the level of exposure these wombats had to *Themeda triandra* prior to capture, it is assumed they were unfamiliar with it due to the fact they had spent the bulk of their life experience away from this particular grass type. The two captive bred wombats had no prior exposure to *Themeda triandra*. The remaining seven wombats were captured as young adults (approximately 4yrs of age) and transferred to Rockhampton Zoo in 2001 and thus had only had the possibility of being exposed to *Themeda triandra* in their natural habitat for a short period of their earlier life.

At the Rockhampton Zoo the wombats are fed a daily diet of pellets and carrots. On every second day the wombats are offered a third food item, either corn on the cob, peanuts, apples or alfalfa sprouts. The wombats were maintained with this zoo-approved diet throughout the course of the study; however, the additional food items not being examined in the present study were fed to the wombats after the feeding periods were completed for the day.

The regular daily pelleted feed ration was used as control for all four food choice experiments. This allowed for the wombats' relative food preferences to be examined in the presence of a known preferred food choice option. The wombats could choose to ignore the experimental food items and consume only the pellets, or, they could make deliberate choices to consume the experimental food options.

The wombats are housed in a purpose built wombat research centre, which comprises four enclosures (Figure 2.1). Three enclosures house two wombats

each and the other houses a group of three wombats. Three enclosures house mixed sex groups, and one a single sex pair of females.



**Figure 2.1: Layout of Rockhampton Zoo Wombat Research Facility**

The effects that group feeding may have on individual food choices needed to be eliminated for this study. The experiments were designed to examine individual wombat's food preferences, therefore, by removing each wombat from its group for 30 minutes within one 24 hour period and offering that wombat the choice of

two food types; pellets and the experimental food item. The same experimental design was used to complete four separate food preference experiments. Pellets were used as the control food choice for all four experiments. The other food items studied were as follows-

1. Experiment 1 – carrots (unnatural/familiar)
2. Experiment 2 – Kangaroo grass (natural/unfamiliar)
3. Experiment 3 – Guinea grass (natural/familiar)
4. Experiment 4 – Sweet potato (unnatural/unfamiliar)

Prior to the commencement of the four experiments the wombats were allowed a 2-week acclimatisation period to explore the feeding pens. This methodology was used to familiarise the wombats with the feeding pen area, the sugar cane mulch substrate that was being used throughout the experiments, and to ensure that the wombats were unstressed when entering and exiting the feeding pens through the tunnels. During this acclimatisation period the wombats were fed in the feeding pen area their usual zoo diet, which included pellets and carrots. It was therefore part of the regular daily routine to move through the tunnels, into the feeding pens in order to consume their daily diet.



**Figure 2.2: Wombats 3, 4 and 5 in the feeding pen during the 2-week pre-study acclimatisation period**

**Experiment 1 – Unnatural/Familiar**

Pellets and carrots were chosen for the first experiment, as both were familiar to this group of wombats. The results from this experiment provided evidence for the decision to continue to use pellets as the control food choice for the three subsequent food preference experiments.

The experiments commenced in the evening at the time when the wombats were beginning to exit their dens to begin their nocturnal activities. In winter, this could be as early as 15:30, and in summer it could be as late as 18:00. On days when ambient temperatures were great, the experiments commenced later due to the wombats not exiting the dens until the temperature had decreased to the extent they were tolerant of the heat. The wombats were not forced into the feeding pen, but rather encouraged to enter by restricting access to their normal enclosure and opening access to the feeding pen. The wombats' curiosity drew them to explore the newly opened tunnel, so there was never a need to manually manoeuvre the wombats through the tunnels.

Each wombat was selected at random to be the first wombat allowed to enter the feeding pen. This random selection process was continued until all five wombats for study on the specific evening had completed their feeding period. No more than five wombats could be studied on any given night due to the length of time of the feeding periods and the associated time involved in moving the wombats to and from the feeding pen. The first wombat would enter the feeding pen anytime between 16:30 and 18:00. It could take as long as 20 minutes to move one wombat through the tunnels into the feeding pen, depending on the disposition of the individual. Usually, when a wombat has first awakened from its day's sleep, it will not initially be interested in eating. It will usually exit the den and lie down outside the den before beginning to graze. An example of this behaviour is shown in Figure 2.3. By the time the last wombat for the evening was ready to enter the feeding pen it could be 20:30. By this stage that particular wombat may be quite hungry, and would, therefore, move through the tunnels

into the feeding pen very quickly. The random selection of each wombat every evening controlled for the effects of hunger on the food choices made. Random selection was carried out by drawing a number from a container. Each wombat was assigned a number at the commencement of this project. Once that wombat had performed its feeding trial for that evening, their number was excluded from the future draws on that same day until all wombats had been randomly chosen. All numbers were placed back in the draw at the commencement of the next days feeding trials.

The two food items were placed in similar bowls 1 metre apart in the centre of the feeding pen. The placement of each food item was randomised so that sometimes the pellets were on the right and sometimes on the left (Figure 2.3). This controlled for preferences relating to the position of the food in relation to the entrance/exit point of the feeding pen. Figure 2.4 depicts the position of the bowls



within the pen.

**Figure 2.3: Demonstrating the view from the elevated observation point, the Excel spread sheet being used to record data and the weather station used to collect temperatures and humidity values at the beginning of each feeding period**



**Figure 2.4: Wombat 4 lying down in the feeding pen during an early evening feeding period. This image also shows the positioning of the food bowls**

Throughout each 30 minute feeding period the wombat was observed from an elevated position and behaviours recorded on an excel spreadsheet in a continuous fashion. The excel spread sheet was automated such that the hitting of one key recorded the exact time the previous behaviour ended and the new behaviour begun. At the end of the 30 minute feeding period the data collected displayed the time devoted to performing each observed behaviour – i.e a start and end time of each behaviour had been recorded and totals for each behaviour calculated automatically for each 30 minute feeding trial. The food that was not consumed by the wombat during the feeding period was weighed, disposed of and the weights then entered into the spread sheet to calculate total amounts of pellets and test foods consumed during that feeding trial. Values for temperature and humidity were also recorded at the time of the feeding period.

Feeding trials were not conducted in rainy weather. The wombats were very reluctant to exit the dens if it was raining, even though the feeding pens were undercover and the wombats did not need to travel through the areas where rain was falling to the ground to get to the feeding pen.

Inter-observer reliability was controlled by having one primary observer who oversaw all of the data recording on one spread sheet template. On occasions

other students/research assistants did participate in recording of observational data. Before performing any observations on their own, the assistants completed two full observation sessions with the primary observer. The additional observers results were compared to that of the primary observers. That is, the total times for each behaviour recorded by the additional observer were compared to the primary observers total times for each behaviour. Once the primary observer was satisfied that they assistants behavioural observations were sufficiently similar, the assistants were able to perform the observations. The primary observer was present for every feeding trial for the full duration of the research. Assistants were able to ask clarification about any behaviour at any time from the primary observer.

The amount of food offered to each wombat did not exceed the recommended daily dietary intake. For each feeding period 300 g of pellets were provided and up to 300 g of the other food item. Following the completion of each study period each evening, the wombats were fed the balance of their daily diet in the usual feeding location. That is, if the wombat consumed 200 g of pellets during the feeding trial, then it was fed 100 g when it returned to its usual enclosure. This ensured all wombats had access to adequate nutrition for the duration of the study. It was important that excess food was not offered to avoid excessive weight gain during the study.

The feeding pens were located at the back of the Research Centre. The pens are connected to the dens via a series of tunnels such that each wombat could exit its den and follow a tunnel system through to the feeding pen.

Once a wombat had entered the feeding pen, a sliding door was closed within the tunnel so that the wombat still had access to part of a tunnel, but could not return to its normal enclosure until released at the end of the 30 minute feeding period.

It was quickly discovered that the wombats became distressed if they did not have a retreat option into the tunnel during the feeding period. If tunnel access was blocked, the wombat would frantically dig at the door trying to get back into the tunnel rather than make a food selection. If one door to the tunnel was,

however, left open, but shut off further within the tunnel, the wombats were content to stay in the feeding pen and did not enter the tunnel even though they could.

As well as the food choices being recorded, 10 other behaviours were recorded on the observation checklist. Start and end times for each behaviour were recorded through the use of an automated excel spreadsheet. Coded keyboard keys for each behaviour were allocated a formula such that when one key was pressed it automatically recorded the exact end time of the previous behaviour and the start time of the next behaviour and calculated the totals of each behaviour. Most of the behaviours recorded were standard wombat behaviours such as walking, digging, sitting, lying, and scratching. Behaviours indicative of a stress response in wombats such as vocalizing, pacing and escape attempts were also included in the observational data spreadsheets. If a wombat exhibited stressed behaviour for more than 5 minutes within the 30 minute assessment period, then that period of assessment ceased and the wombat returned to its regular enclosure. During a total of 288 feeding trials this only occurred on three occasions. These three assessment periods were repeated at a later time when the wombats were demonstrating unusual or stressed behaviour. During pre-assessment periods data for two other behaviours were included on the spreadsheet, these being pacing and drinking. These two behaviours were not detected during any of the feeding periods over the course of the four experiments and so were not included in experimental analyses. Pacing is generally considered a negative welfare marker, so its absence from these observations is an indication that the wombats were not stressed by the experimental procedures to the extent that they were pacing during the period of experimental assessments. It is not surprising that drinking was not detected during the assessment periods in any of the experiments, as wombats rarely drink. It is likely that the wombats drank at times outside of their period of feeding assessment when they had access to their normal enclosures.

Following is a list of the classifications of wombat behaviours as defined for the series of experiments in this thesis.

1. Eating pellets –positioned at the pellet bowl and/or is actively chewing on pellets
2. Eating other –positioned at the other (grass, sweet potato, carrot) bowl and/or actively chewing on the other food item
3. Walking – walking around the feeding pen
4. Sitting – positioned with posterior body part on the ground with a stationary posture
5. Lying –lying down but awake (eyes open)
6. Sleeping –lying down not awake (eyes closed)
7. Digging –digging in the soil
8. Scratching –pawing self
9. Vocalising –making a typical noise
10. Sniffing –smelling the ground
11. Standing –standing upright in a stationary posture
12. Escape attempt – digging at an exit point or attempting to climb out of feeding pen

Carrots, sweet potato, kangaroo grass and guinea grass were selected as the food choices (Table 2.1) to be compared with pellets (the unnatural/familiar control food item).

**Table 2.2; Experimental food choices**

	<b>Familiar</b>	<b>Unfamiliar</b>
<b>Natural</b>	Guinea Grass ( <i>Megathyrsus maximus</i> )	Kangaroo Grass ( <i>Themeda triandra</i> )
<b>Unnatural</b>	Carrot	Sweet Potato

During the course of each experiment the wombats were housed in their normal groups in their normal enclosures. The Wombat Research Centre is designed in such a way that any wombat can be given access to any pen at any time by opening a combination of upward sliding doors. The wombats were never actively forced in to the feeding pen. The observers waited for the wombats to enter the pen in own time. The wombat's own curiosity lead them to explore the newly opened door. This method was used to move one wombat at a time into the feeding pen. At the end of the 30 minute feeding period the wombat was moved back through the tunnel to its regular den and the next wombat was moved into the feeding pen. This method of moving wombats between enclosures was familiar to the wombats, as it has been part of their daily husbandry since they arrived at the zoo in 2001 (or later in the case of the two captive bred wombats). When considering the ethical implications of removing the wombats' choice to leave the den system until after their feeding trial, it was decided that moving the wombats in this fashion was less stressful than manually handling the animals. In this regard the wombats still has some choice available to them (ie leave the den to enter the feeding pen, or stay in the den). The wombats were not ushered out of the dens, but were allowed to exit the den in their own time. This lengthened the duration of the feeding trails, but maintained some level of control by the wombats.

The wombats were selected at random (by drawing numbers from a container which corresponded with the names of the wombats) for the sequence of feeding so that the same wombat was not entering the feeding pen in the same sequence each day. In this way the effects of time were controlled so that data were not confounded with regard to feeding sequence. A maximum of five feeding periods were conducted per feeding session. This reduced the amount of time the wombats had to wait before being given access to their food sources. Once the feeding period for each wombat was completed and the individual moved through the tunnel back into its usual enclosure it was given full access to its outdoor area as well as den system. Once all wombats from any one group had finished their daily feeding period, they were fed the balance of their usual zoo diet.

At the beginning of each feeding period the food items were weighed and any remaining food was weighed at the conclusion of the 30 minute feeding periods. This allowed for quantity of food to be recorded as well as the time devoted to eating each of the food items. Eight replicates were performed for each wombat. That is, each wombat had eight 30 minute feeding periods in which feeding behaviours were assessed for each of the experimental food choices.

### Experiment 2 – Natural/Unfamiliar

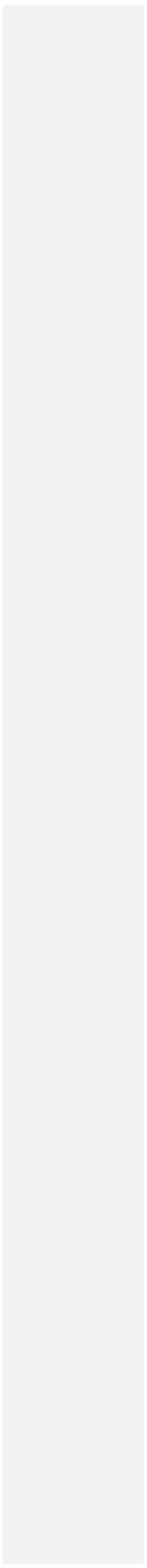
The same experimental design was used in Experiment 2 as in Experiment 1 only the grass replaced carrots as the food source for which feeding behaviour was being assessed. As kangaroo grass could not be locally sourced, it was propagated on site. A local high school's wildlife action group volunteered to propagate this grass for the project. Seeds were purchased and the school children came to the zoo to plant the seeds in potting mix in trays. The trays were monitored and watered every second day to ensure a desirable growth rate for the kangaroo grass.



Figure 2.5: Kangaroo grass sprouting



Figure 2.6: Kangaroo grass at growth stage used for feeding to wombats



Fourteen weeks after planting the grass was of a length suitable to be fed in the trays to the wombats. As for Experiment 1, samples of the grass were collected and stored after each evening feeding period so that dry weights of the grass could be subsequently calculated.

### Experiment 3 – Natural/Familiar

Guinea grass grows in all of the wombat enclosures at Rockhampton Zoo (Figure 2.7), so this grass was picked fresh on site and fed in trays during Experiment 3 (Figure 2.8). Enough grass for five feeding periods was collected prior to the commencement of the feeding period each day and was stored in a cooler box until ready to be used. Any grass that had not been fed at the end of the evening was disposed of and fresh grass was collected again the next day for the next feeding period.



**Figure 2.7:** Guinea grass clumps growing in the regular enclosures



**Figure 2.8:** Guinea grass freshly picked and fed out to wombat during a feeding period

### Experiment 4 – Unnatural/Unfamiliar food

The fourth experiment was conducted to compare an unnatural/unfamiliar food, sweet potato, to the staple diet of pellets. All methods were the same as for the previous three experiments.



**Figure 2.9: Experiment 4 feeding period showing sweet potato in the feed bowl**

The wombats were weighed by placing scales inside their den area at the beginning and end of each of the four experiments. There were no significant changes in wombat weights during the period during which these experiments were conducted.

Wet weight measurements (rather than dry weight measures) were used for all food items to standardise the mass of food consumed. Whether or not the foods were chosen because of their moisture content was not examined in this research. The aim of this research was to examine the food choices of the wombats. Wet weights would have yielded the same results for the chosen experimental question – can wombats make clear choices regarding different food items.

The average amount of time spent observing each individual wombat over all four experiments was 69 hours  $\pm$  3 hours.

The following two chapters are presented as research papers for peer refereed journals. As such, there is some repetition of the methods described in this chapter.

Chapter 3 (p40-75) has been **formatted and referenced according to the requirements of *Zoo Biology*, the journal to which this paper has been submitted.**

Chapter 4 (p76-96) has been formatted and referenced according to the requirements of *Journal of Applied Animal Behaviour Science*, the journal to

Chapter 3 – Magnitude and frequency of food preferences of captive southern 45  
hairy-nosed wombats

**which this paper is due to be submitted. As such, the referencing styles  
used within these two chapters differ.**

## Chapter 3

### Magnitude and frequency of food preferences of captive southern hairy-nosed wombats and the implications for captive management of this species

#### Abstract

This research examined food preferences of captive wombats when offered a selection of common captive wombat diets used by zoos and wildlife carers Australia wide. Although there has been research on the food preferences of many domestic and captive wild animals, there has been *no study* on the food preferences of captive southern hairy-nosed wombats. A review of the research has highlighted a lack of knowledge of wombat food preferences and that feeding regimens for captive wombats may be inadequate.

The hypothesis for this study was that captive southern hairy-nosed wombats would prefer natural familiar foods to unnatural unfamiliar foods. The research was conducted at the Rockhampton Zoo Wombat Research Centre and involved a series of four food preference experiments (carrot, kangaroo grass, guinea grass and sweet potato). All of the foods selected for this project are routinely fed to captive wombats in all Australian zoos.

The results showed that this group of captive wombats preferred natural foods regardless of familiarity and that given the choice of pellets (control test food item) and one other food item, grasses were preferred even if they were unfamiliar. The various test food items also significantly influenced the average and total consumption of the control food item (pellets) with carrot decreasing food intake and eating behaviour while the grasses and sweet potato enhanced this. Wombats also showed important individual differences in food preference which advocates for more tailored dietary options with fresh grasses as an essential component.

#### Keywords

Food choice, Food preference, Wombat, Zoo

### Introduction

Southern hairy-nosed wombats (*Lasiorhinus latifrons*; SHNWs) are grazing herbivores that feed predominantly on the low nutrient content grasses of the arid regions of South Australia and southern parts of Western Australia (Triggs, 2009). To cope with this poor quality diet, wombats have developed unique behavioural and physiological adaptations that can be broadly explained through four main features. Firstly, they have the lowest basal metabolic rate of any marsupial (Barboza, 1989; Barboza and Hume, 1992). Secondly, they have an efficient masticatory system, which includes permanently growing teeth – a trait unique to wombats among the marsupials (Hume, 1999). Thirdly, wombats have a digestive system that facilitates colonic fermentation and slow passage of digestion (Hume, 1999). The fourth feature is the wombat's sedentary and burrowing lifestyle (Finlayson, Shimmin et al., 2005).

The nutritional requirements of wombats have been studied and previously reported (Hume, 1989; Barboza and Hume, 1992; Barboza and Hume, 1992; Barboza, 1993; Hume, 1999). The preferred species of grasses consumed by wild wombats are not often fed by zoos (Barboza, 1993; Barboza, 1993; Barboza, Hume et al., 1993; Woolnough, 1998). Close examination of diets fed to captive SHNWs in zoos reveals there is little resemblance to the diet of their wild counterparts. For example, Barboza (1993), Triggs (2009) and Hume (1989) report that wild SHNWs choose a high fibre/low protein diet that is also low in copper and nitrogen. Information gathered from personal correspondence with Australian zookeepers indicates that captive wombat diets are greater in energy and protein (unpublished survey 2012) when compared with the diets of wombats in their natural habitat. This may be due to the inclusion of items such as carrots, corn and lucerne chaff/hay in captive diets when compared to the lesser energy and protein content in the grasses SHNWs graze in their natural habitat. The zookeepers reported that wombats had a preference for these food items when offered a choice of lesser quality feeds, such as oaten hay. In most zoos the captive wombats were offered very little opportunity to graze fresh growing grasses. Many zoos house wombats in enclosures with a dirt, sand or concrete substrate, which allows for easy cleaning of the enclosures, however,

this restricts the wombats' ability to perform the natural behaviour of selective grazing. Some zookeepers allowed wombats partial access to grazing, while others provided grass in the form of fresh pickings or no grasses at all.

Rate of consumption was another area of interest to this study. The relationship between time spent eating and the amount of food ingested has not been studied in this species, though studies in other species have demonstrated substantial variation both between- and within- subjects for feeding times and ingestion rates (Zinner 1999).

The present research was conducted to study the food preferences of a group of captive SHNWs housed at the Rockhampton Zoo Wombat Research Centre and at present is the initial and only study of food preferences in captive SHNWs. These experiments were designed to assess the food preferences of captive SHNWs when offered familiar foods compared with unfamiliar foods and natural foods compared with unnatural foods. The foods options were categorised as follows:-

- Pellets (unnatural/familiar) control test food (provided during all trials/experiments)
- Carrots (unnatural/familiar) – Experiment 1
- Kangaroo grass (natural/unfamiliar) – Experiment 2
- Guinea grass (natural/familiar) – Experiment 3
- Sweet potato (unnatural/unfamiliar) – Experiment 4

The primary hypothesis states that natural, familiar food items will be preferred over unnatural, unfamiliar items.

The aims of these experiments were as follows:-

1. To determine how much of the test food item was consumed
2. To determine how many times the test food item was selected
3. To determine how many times the consumption of the test food item exceeded consumption of pellets.

4. To establish if the test food items influenced the consumption of pellets
5. To determine if food preference is affected by variables such as rate of consumption and environmental conditions
6. To design an experiment that would provide results, be repeatable and provide a model for future experiments in this field.

## Methods

### *Animals*

The wombats had been in residence at the Rockhampton Zoo Wombat Research Centre for several years prior to this study. Two of the wombats were born at the zoo in 2004 and 2010, respectively, while the remainder of the wombats were originally born in their natural habitat. These wombats were trapped and transported to the zoo in 2001 for the zoo's research program. Nine of the zoo's eleven southern hairy-nosed wombats were included in the present study. The two animals excluded from this study were the zoo's display wombats housed in an area separate from the Wombat Research Centre. All nine experimental animals were long-term residents of the Research Centre and were in group housing arrangements where individuals of the groups had been in cohabitation within the group for extended periods. The group comprised of three males and six females. Apart from the two wombats born at the zoo, ages of the wombats were unknown.

All of the wombats were in good health and body condition, as declared by the zoo veterinarian, with a mean weight of  $30.5 \pm 4.4$  kg. The wombats were weighed fortnightly in accordance with standard zoo practice to ensure they were maintaining a healthy weight.

The procedures employed with this research project were approved by the CQUniversity Animal Ethics Committee (approval number A12/11-290).

### *Experimental facilities*

The wombats were housed in Rockhampton Zoo's Wombat Research Centre, which comprises four enclosures. This facility has been described in previous

publications by Descovich et al. (Descovich, Lisle et al. 2012 (a); Descovich, Lisle et al. 2012 (b); Descovich, Lisle et al. 2012 (c)).

Three enclosures housed two wombats each and the other housed a group of three wombats. The wombats in three enclosures were of mixed sex, and one enclosure housed only two female wombats.

Prior to the commencement of this study, the experimental group of wombats had been maintained on a diet of Riverina Macropod Pellets (unnatural/familiar) and carrots (unnatural/familiar) fed daily, as well having 24 hr access each day to grazing grass that was growing in the enclosures where they were housed (*Megathyrus maximus* and *Paspalum spp.*). This diet was approved by the zoo's veterinarian and had been used for many years (unpublished zoo records). The normal feeding routine included feeding of the pellets and carrots late in the afternoon in a trough large enough for three wombats to feed at one time.

The effects that group feeding had on individual food choices were negated for this study by removing each wombat from its group for 30 minutes per day. During this 30 minute period each wombat was individually offered the choice of two food items; pellets and one of the four experimental food items. Pellets (unnatural/familiar) were used as the control for all four food choice experiments as this was the staple diet for this group of wombats and all wombats reliably consumed pellets daily.

Prior to the commencement of the four experiments, there was a 2-week acclimatisation period to familiarise the wombats with the feeding pen area, sugar cane mulch substrate that was used for floor cover throughout the experiments, and to ensure that the wombats were adapted to the regimen of entering and exiting the feeding pens through the tunnels. Including the acclimation activities, the wombats were routinely managed for husbandry purposes (body weight, enclosure switching) hence the interaction in the tunnels and feeding pens was a familiar activity with minimal welfare impact.

The experiments commenced in the evening at the time when the wombats were beginning to exit their dens to begin their nocturnal activities. In winter, this was as early as 1530, and in summer it was as late as 1800. The wombats were

not forced into the feeding pen, but rather encouraged to enter by restricting access to their normal enclosure and opening access to the feeding pen. The wombats' curiosity drew them to explore the newly opened tunnel, so there was never a need to manually manoeuvre the wombats through the tunnels.

The feeding pens were located at the back of the Research Centre. The feeding pens were connected to the dens via a series of tunnels such that each wombat could exit its den and follow a tunnel system to the feeding pen.

Once a wombat had entered the feeding pen, a sliding door was closed within the tunnel, so that the wombat still had access to part of a tunnel, but could not return to its normal enclosure until released at the end of the 30 minute feeding period.

Each wombat was selected at random by drawing a number from a container. The numbers were allocated to each wombat at the commencement of this project. This selection process was continued until all five wombats that were to be studied on a specific evening had participated in the feeding period. No more than five wombats could be studied on any specific night due to the length of time of the feeding and associated time involved in moving the wombats to and from the feeding pen. The random selection of each wombat every evening controlled for the effects of hunger as influenced by order of feeding on the food preference choices made by the wombats.

### *Food choices*

For the purposes of this research project, the terms familiar, unfamiliar, natural and unnatural are defined in the following ways-

- Familiar – foods that are routinely fed to the SHNWs at Rockhampton Zoo
- Unfamiliar – foods that the Rockhampton Zoo wombats have not been exposed to in the past 13 years of their recorded history in captivity

- Natural – foods that wombats in their natural habitat have been recorded as selecting, eating and therefore surviving on in their natural environment
- Unnatural – foods that wombats would not routinely have access to in their natural habitat and therefore do not naturally eat in their natural environment

The familiar foods chosen were pellets (Riverina Macropod Pellets), carrots and a grass that grows naturally in the enclosures housing the wombats, *Megathyrsus maximus* (Guinea grass). The unfamiliar foods were sweet potato, and a native grass, *Themeda triandra* (Kangaroo grass). *Themeda triandra* is one of the preferred grass species of wild SHNWs (Triggs 2009), hence its selection for the present study. Sweet potato was chosen as the unfamiliar, unnatural food as it is commonly fed to captive wombats as reported by the zoo keeping staff throughout Australia, but was not part of the routine diet for the Rockhampton Zoo SHNWs.

The experimental food items studied were as follows:-

5. Experiment 1 – carrots (unnatural/familiar) vs. pellets (unnatural/familiar)
6. Experiment 2 – Kangaroo grass (natural/unfamiliar) vs. pellets (unnatural/familiar)
7. Experiment 3 – Guinea grass (natural/familiar) vs. pellets (unnatural/familiar)
8. Experiment 4 – Sweet potato (unnatural/unfamiliar) vs. pellets (unnatural/familiar)

The two food items (pellets and experimental) were placed in similar bowls one metre apart in the centre of the feeding pen (Figure 3.1). The placement of each food item was randomised to eliminate placement bias. The random placement of the bowls controlled for preferences relating to the position of the food in relation to the entrance/exit point of the feeding pen. Feeding periods were not

conducted in rainy weather due to the reluctance of the wombats to exit the dens if it was raining.



**Figure 3.1: Placement of feed bowls within feeding pen**

The amount of food offered to each wombat did not exceed the recommended daily dietary intake as stated in the *SHNW Husbandry Manual* (Treby 2005). During each feeding period, 300 g of pellets were fed and approximately 300 g of the other food item. On some occasions more of the test food was added during the feeding trial to prevent the wombat from running out of food choices. The wombats' were not left without food during the 30 minute feeding period. Following the completion of each feeding period, the wombats were fed the remaining portion of their daily diet in the usual feeding location. That is, if the wombat consumed 200 g of pellets during the feeding period, it was fed 100 g when it returned to its usual enclosure. This ensured all wombats had access to adequate nutrition for the duration of the study. It was important that excess food was not offered to avoid excessive weight gain during the study.

If a wombat exhibited stressed behaviour for more than 5 minutes within the 30-minute feeding period, the feeding period was discontinued and the wombat was allowed to return to its regular enclosure. This occurred on only three occasions. These three feeding periods were completed at a later time when the wombats were not stressed

Outside of the 30-minute feeding periods the wombats were housed with their cohabitants in their normal enclosures. Once the feeding period was completed for each wombat, and the wombat had moved through the tunnel back into its usual enclosure it was given full access to its outdoor area as well as den system.

As this was the first organised study of food preferences in captive SHNWs, the methods used allowed for addressing one final experimental aim: That the experimental design used in the present study would provide results, be repeatable, and be a model for future experiments in the field of wombat study.

### *Data Collection*

At the beginning of each individual feeding period the food items were weighed and any remaining food was weighed at the conclusion of the feeding period. This allowed quantity of food to be recorded as well as the time that elapsed while eating each of the food items. Eight feeding period replicates were performed for each wombat. That is, for each wombat there were eight 30-minute feeding periods for each of the four experimental food choices: a total of 288 feeding trials (72 feeding trials for each of the four test food items).

Throughout each 30 minute feeding period the wombats were observed from an elevated position and behaviours recorded on a Microsoft Excel spread sheet in a continuous fashion. At the end of the 30 minute feeding period, the data collected provided numeric documentation of the amount of time that had elapsed while the wombat was eating. The food that was not eaten was weighed, disposed of and the data for weights of this food were then entered into the spread sheet along with numeric values for temperature and humidity at the time of the period. The majority of the data collection experiments were conducted by the primary study investigator, with several other trained individuals used during different experiments to observe the wombats. These individuals were continually supervised by the primary study investigator at all times and there were no differences found in the recording/observation of the data by the various observers.

### Statistical analyses

Graphical representations of the data collected were created using Microsoft Excel. All data was presented as mean  $\pm$  SEM with analysis of variance and tests evaluating preferences for naturalness compared with familiarity conducted using the Graphpad Prism statistical program. T-tests for significance was used at  $P \leq 0.05$ .

### Results

#### Total, average consumption and group preference data for test food items

There were a total of 288 feeding trials (8 x 30minute observations periods per wombat for four food choice experiments). Each food choice, therefore, had 72 feeding trials (8 feeding trials per wombat). The wombats showed a clear difference in the magnitude of food consumed (by weight in grams) during the four experiments (Figure 3.2). When kangaroo grass, guinea grass or sweet potato were provided, the wombats cumulatively consumed a larger quantity of food by weight. Experiments 2, 3 and 4 showed significantly higher average combined food consumption than Experiment 1 (Figure 3.3). When carrot was offered as the test food item, the wombats consumed 50% less total food. There were no changes observed in the average of the combined foods eaten during Experiments 2, 3 and 4.

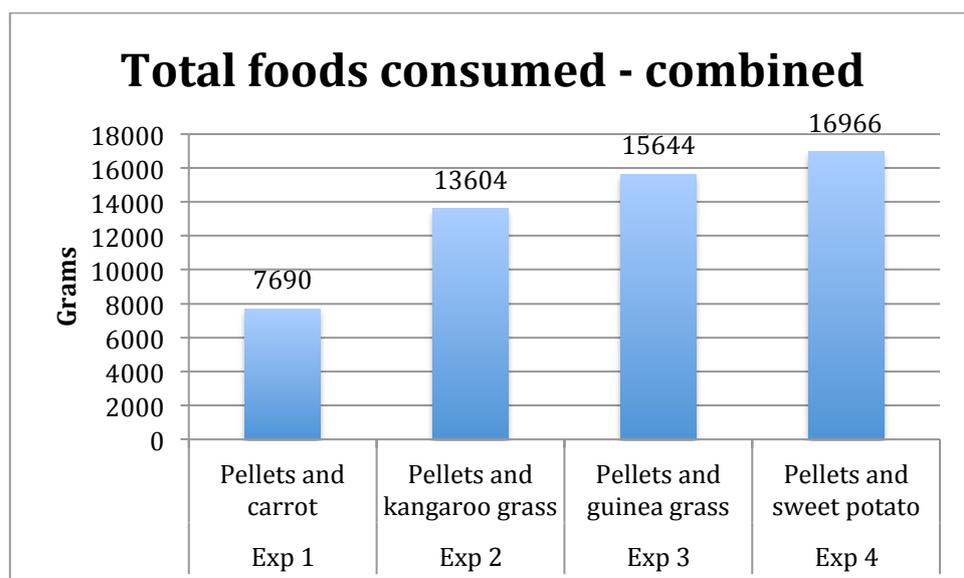


Figure 3.2: Total foods consumed - combined

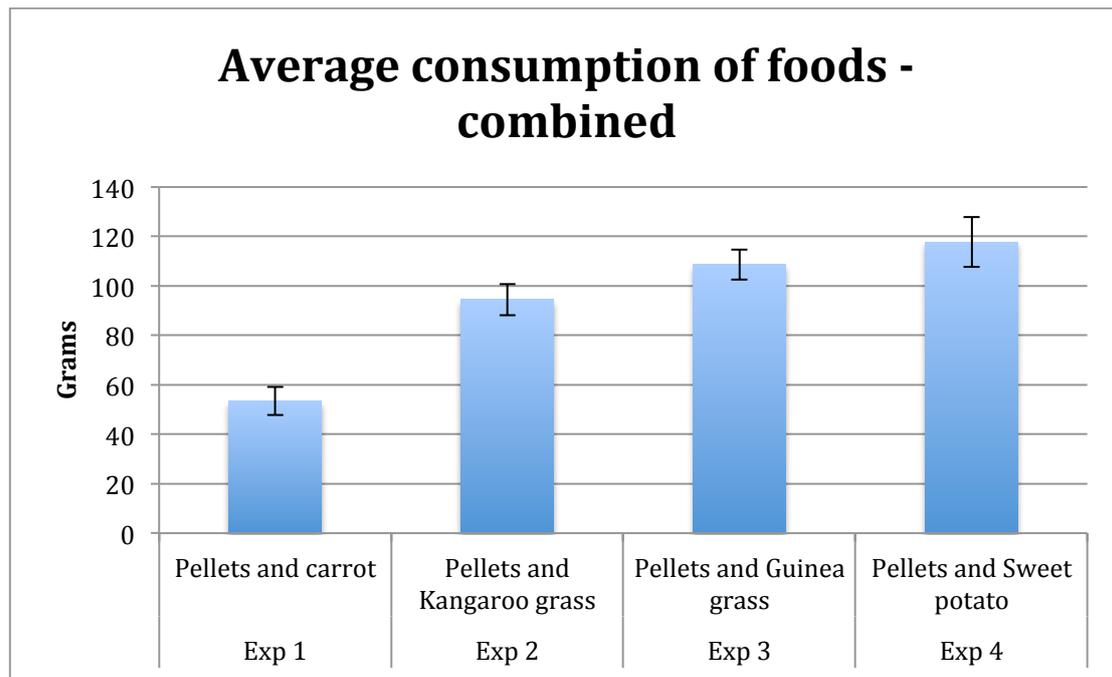
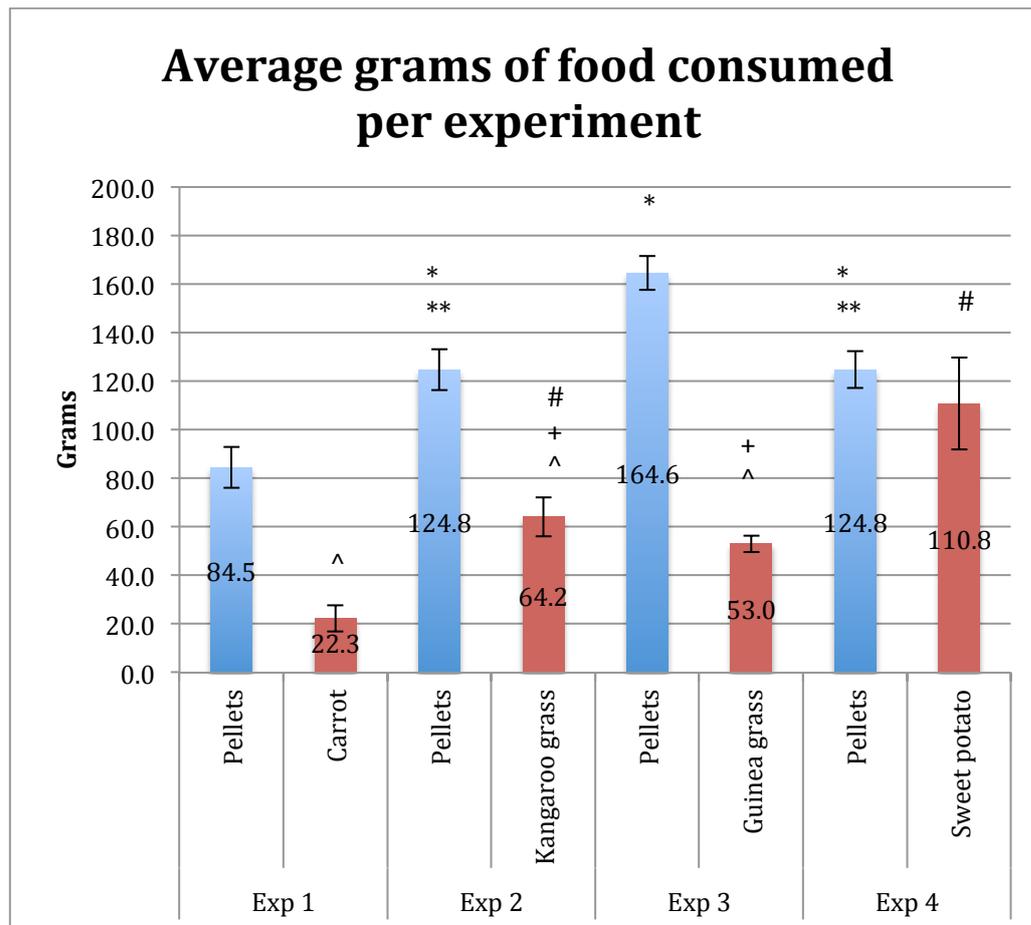


Figure 3.3: Mean total combined foods

These data (Figures 3.2 and 3.3) are supported by the calculation of mean grams of each food item consumed during each of the food choice experiments (Figure 3.4). Following a similar trend to the total and average total food consumed, when carrot was the test food, significantly less pellets were consumed compared to when kangaroo grass, guinea grass or sweet potato were offered. On average wombats consumed the most pellets when guinea grass was offered with a similar quantity of pellets consumed when kangaroo grass or sweet potato was provided. Both kangaroo grass ( $64.2 \pm 8$  g) and guinea grass ( $53 \pm 3.3$  g) were consumed on average to a similar level during Experiments 2 and 3, however, this was significantly less than the control food item (pellets) for their respective trials. The average consumption of carrot ( $22.3 \pm 5.5$  g) was significantly lower than all other food items and significantly less than pellets. Sweet potato was, on average, the most consumed test food item and showed an equal average consumption compared to pellets during Experiment 4.



**Figure 3.4: Mean grams consumed per experiment. Data represented as mean  $\pm$  SEM,  $n=72$  for all groups; \*  $p<0.05$  vs pellets Experiment 1; \*\*  $p<0.05$  vs pellets Experiment 3; #  $p<0.05$  vs carrots; +  $p<0.05$  vs sweet potato; ^  $p<0.05$  vs pellets.**

The number of times the wombats chose to eat some of the test food items is another indicator of choice, regardless of the amount of the test food consumed. That is, the percentage of trials (i.e. 30 minute feeding periods) each wombat chose the test food item (Number of times test food chosen divided by the total number of trials) (Table 3.1). These results have some similarity to the average of the total amounts consumed (Figure 3.4), but also show important differences. The wombats demonstrated a strong preference to consume pellets (95.1%), guinea grass (95.8%) or kangaroo grass (81.9%) during the feeding trials (Table 3.1). In contrast, carrot and sweet potato were consumed during only 43% and 50% of the feeding trials respectively (Table 3.1). This demonstrated a strong

preference for trying natural food items regardless of familiarity. However, the wombats still maintained a high preference for the control food item (pellets – unnatural/familiar).

The unnatural familiar control food item (pellets) was consumed on average the most in quantity and frequency during the trial experiments. Therefore, if a test food item was consumed in greater quantity than pellets during any of the feeding trials, this was considered a compelling decision by the wombat. These results show that the unfamiliar food items (kangaroo grass 22.6%; sweet potato 33.3%), when chosen, were consumed more frequently and in greater quantity than the familiar food items (Guinea grass 7%; Carrot 6.9%) (Table 3.2). Whilst the summary data of average and frequency of preference of food item consumption and consumption of the test items relative to pellets demonstrates strong differences across the wombats, the individual wombat trial preference data are equally important.

**Table 3.1: Percentage of trials wombats chose to eat the test foods**

	<b>Familiar</b>	<b>Unfamiliar</b>
<b>Natural</b>	Guinea Grass 95.8%	Kangaroo Grass 81.9%
<b>Unnatural</b>	Carrot 43%	Sweet Potato 50%
<b>Control food item</b>	Pellets 95.1%	

**Table 3.2: Percentage of trials consumption of test food exceeded consumption of pellets**

	<b>Familiar</b>	<b>Unfamiliar</b>
<b>Natural</b>	Guinea grass 7%	Kangaroo grass 22.6%
<b>Unnatural</b>	Carrot 6.9%	Sweet potato 33.3%

Experiment 1 – Carrot (unnatural/familiar)

The general trend for all wombats in Experiment 1 was that they consumed significantly more pellets than carrots by weight (Figures 3.4, 3.5) and that carrot consumption by weight was consistently low relative to pellets (Figure 3.6). To calculate the ratio of carrot consumed relative to pellets, the total amount of carrot eaten was divided by the total amount of pellets consumed for each wombat. A value less than one indicates that less carrot was eaten than pellets. A value equal to 1 means that equal amounts of carrot and pellets were consumed. A value greater than one indicates that more carrot than pellets was consumed. Wombat 6 consumed more carrot than pellets on three separate occasions. Across all other wombats and feeding trials there were only two other occasions of wombats consuming more carrot than pellets (wombats 4 on feeding trial 4 and wombat 8 on feeding trial 6). Overall, only three out of nine wombats ever ate more carrot than pellets. From this information it could be determined that wombat 6 demonstrated a greater liking of carrot than the other wombats and that eight of the nine wombats consistently preferred pellets to carrots.

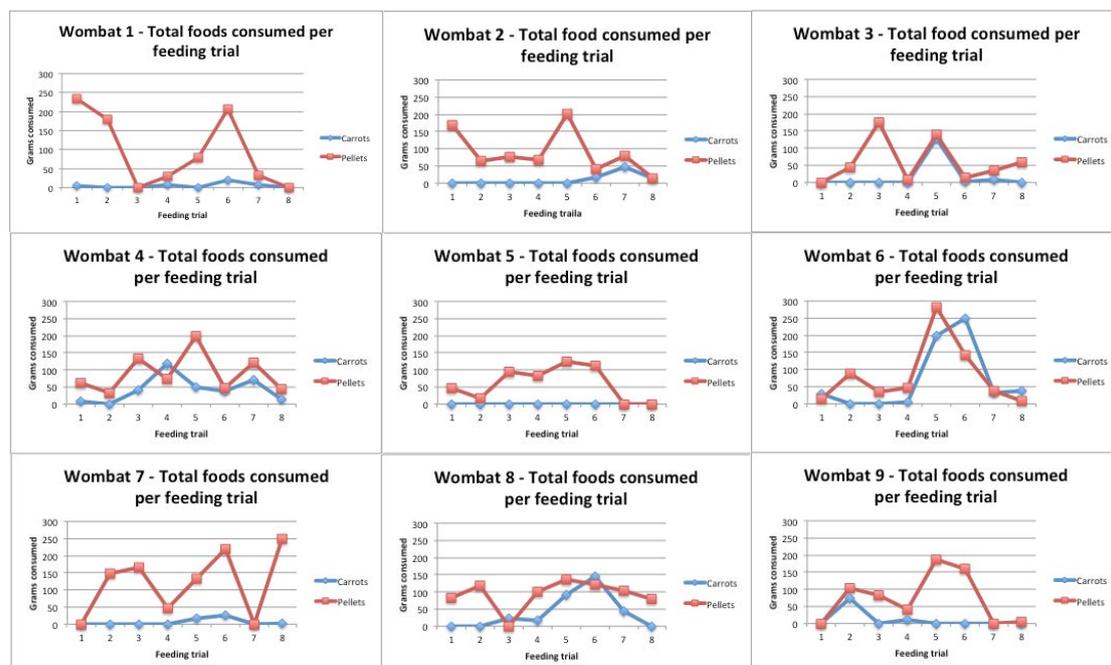
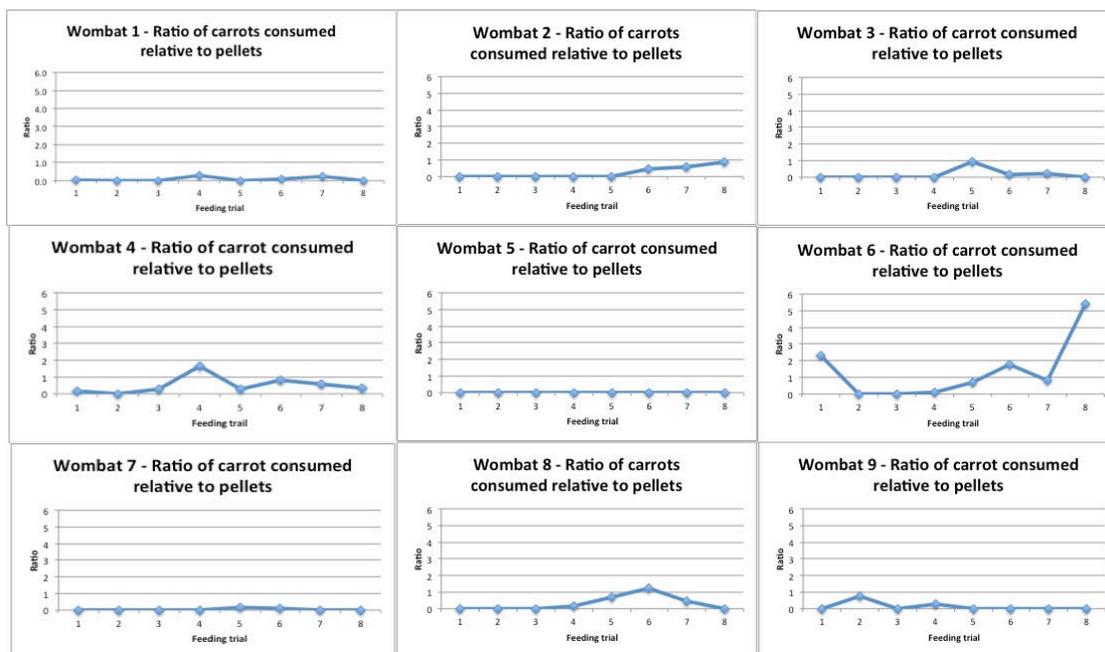


Figure 3.5: Total foods consumed per feeding trial (Experiment 1)

## Chapter 3 – Magnitude and frequency of food preferences of captive southern 60 hairy-nosed wombats



**Figure 3.6: Ratio of carrot consumed relative to pellets**

### Experiment 2 – Kangaroo grass (natural/unfamiliar)

Pellet consumption was more variable (i.e. intake fluctuated between each feeding trial) for all wombats in this experiment (Figure 3.7) when compared to Experiment 1, however the overall mean consumption of pellets was similar to the other three experiments (Figure 3.4). The test food (kangaroo grass) consumption was again relatively low compared to pellets except for a few occasions when some wombats consumed greater quantities of grass (Figure 3.8). The average consumption of kangaroo grass was still three times the average consumption of carrots (Figure 3.4). The wombats consumed more kangaroo grass than pellets during 17 of the 72 feeding trials. This equates to 23.6%. Wombat 1 consumed more grass than pellets 62% of the time (five out of eight trials). Six out of nine wombats ate more kangaroo grass than pellets at least once, which was greater than the choices during Experiment 1. These results indicate the wombats generally preferred kangaroo grass over the carrot offered in Experiment 1.

# Chapter 3 – Magnitude and frequency of food preferences of captive southern hairy-nosed wombats

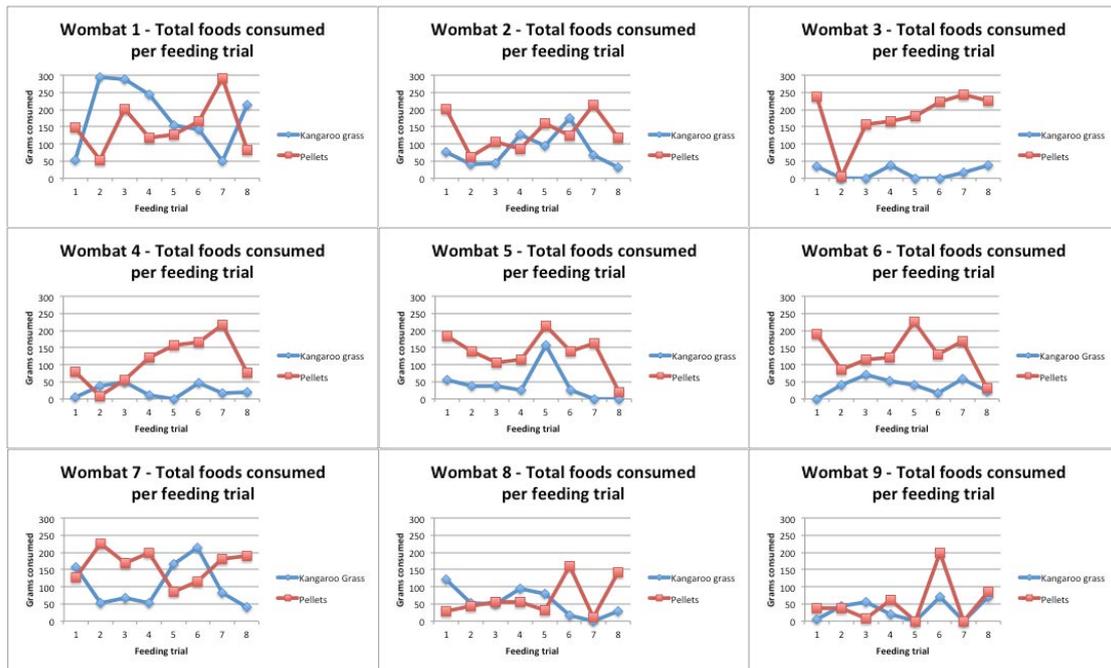


Figure 3.7: Total foods consumed per feeding trial (Experiment 2)

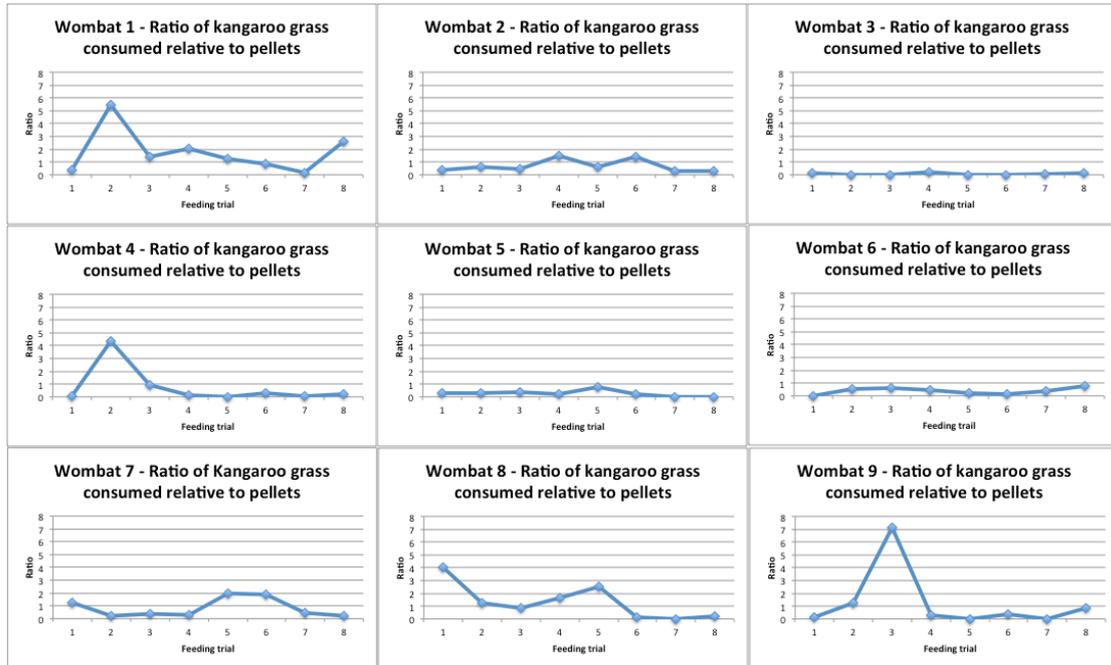


Figure 3.8: Ratio of kangaroo grass consumed relative to pellets

## Chapter 3 – Magnitude and frequency of food preferences of captive southern 62 hairy-nosed wombats

### Experiment 3 – Guinea grass (natural/unfamiliar)

The graphical representation of these data show a more even consumption of both food items offered during these feeding trials (Figures 3.9, 3.10). That is, the individual graphs are less varied than for the previous two food choices. This means that there were variations in the amount consumed of each food each day and that consumption was similar across days in this experiment – Experiment 3. Wombats consumed more grass than pellets 7% of the time. Wombat 9, on one occasion, ate no pellets at all, and consumed only guinea grass. Three out of nine wombats ate more guinea grass than pellets on at least one occasion (Wombats 3, 8 and 9). The amount of guinea grass consumed was not significantly more than this wombat had consumed in previous or later trials, but the data stands out due to the wombat's decision to not eat pellets on this occasion. When being fed both the control food (pellets – unnatural/familiar) and guinea grass (natural/familiar) the wombats consumed the greatest amount of pellets (Figure 3.4). In comparison to experiment 2 (Kangaroo grass) the guinea grass was consumed equally (Figure 3.4).

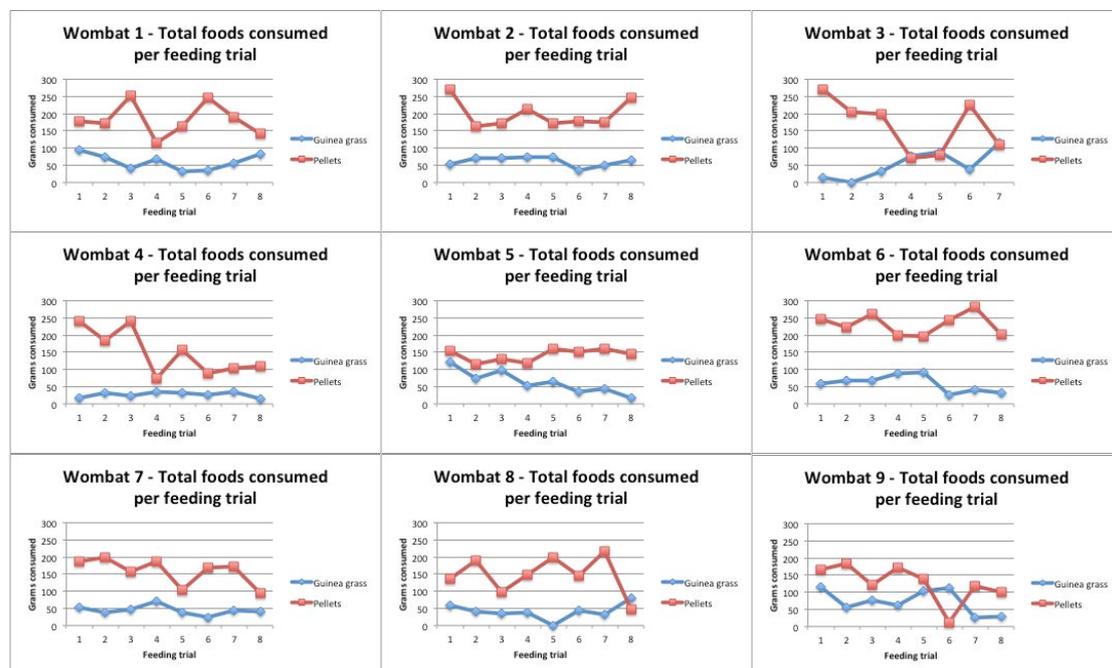
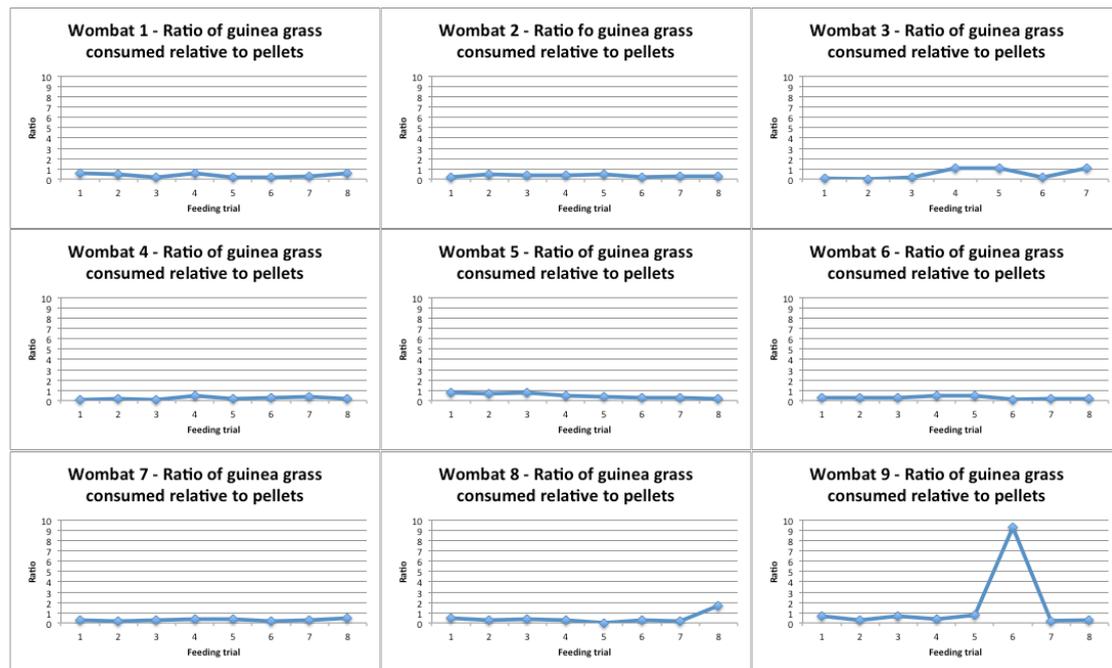


Figure 3.9: Total foods consumed per feeding trial (Experiment 3)



**Figure 3.10: Ratio of guinea grass consumed relative to pellets**

#### Experiment 4 – Sweet potato (unnatural/unfamiliar)

The results for the sweet potato feeding trials are the most polarising of the four food choice experiments. The wombats consumed more sweet potato by weight than pellets during 33.3% of the feeding trials. Pellets were still preferred during 66.6% of the trials when measured by weight of food consumed. During these trials it was necessary on some occasions to add more sweet potato to the food bowls before the 30 minute trial ended to prevent the wombats from running out of food choices. As such, there were some instances where more than 300 grams of sweet potato were consumed. For this reason the scale of the “total foods consumed” graphs have been increased to accommodate the larger amounts consumed (Figure 3.11). Four of the nine wombats did not consume any sweet potato (Wombat 4, 5, 6 and 9). The five wombats that consumed sweet potato ate so much of it that the ratio graphs (Figure 3.12) have been scaled individually in some instances (Wombats 1 and 7). Two wombats produced data different from the other wombats in this food choice experiment – wombats 1 and 7. Wombat 1 consumed up to 20 times more sweet potato than pellets during one feeding trial. Wombat 7 consumed more than 70 times more sweet potato than pellets on two separate occasions. During this experiment the average pellet consumption was identical to that observed when kangaroo

Chapter 3 – Magnitude and frequency of food preferences of captive southern 64 hairy-nosed wombats

grass was the test food item but significantly less than when guinea grass was provided (Figure 3.4). Sweet potato was the only item with an average consumption not significantly different to the average pellets consumed during the various food choice trials (Figure 3.4).

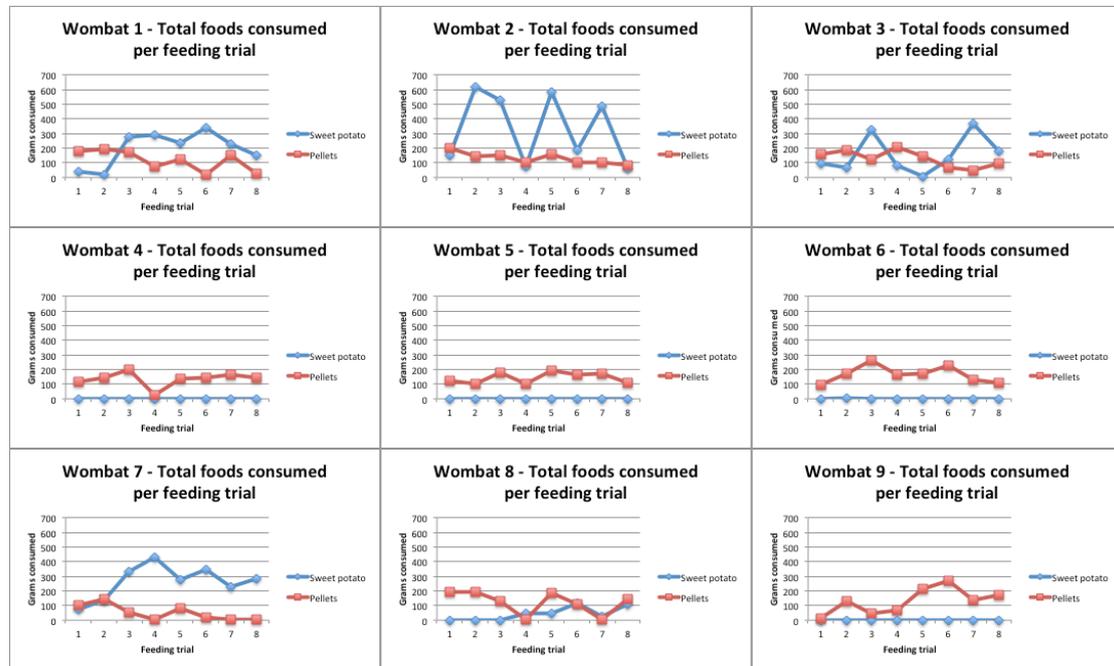


Figure 3.11: Total foods consumed per feeding trial (Experiment 4)

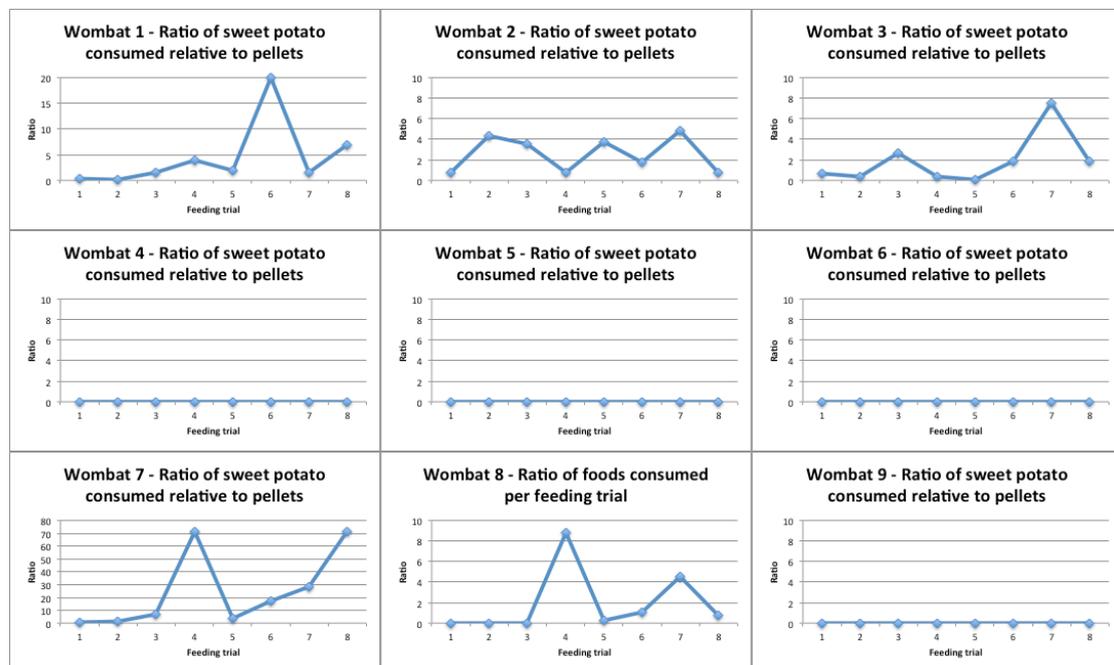


Figure 3.12: Ratio of sweet potato consumed relative to pellets

## Chapter 3 – Magnitude and frequency of food preferences of captive southern 65 hairy-nosed wombats

All wombats demonstrated a similar consumption of pellets across all feeding trials (Figure 3.13). This indicates that the wombats were accustomed with pellets as a food item and that they enjoyed eating it enough to routinely chose it even when offered alternatives.

The test food items showed marked variety among the wombats. In order to present the data in an easy to read format, the scale for wombats 1, 2, 3 and 7 have been adjusted to account for their increased intake of certain food items.

From these graphs (Figure 3.13) it can be observed that wombats 1 and 7 preferred sweet potato and kangaroo grass over the other food items. That is, they both preferred the unfamiliar foods over the familiar foods.

Wombats 2 and 3 showed strong preference towards sweet potato. Wombats 5, 6, 8 and 9 all demonstrated a preference to consume more pellets than any other food item. Wombat 5, while preferring pellets over all other items offered, did show a preference for the grasses over carrot and sweet potato.

Each set of feeding trials produced at least one wombat whose food choices differed from the other wombats within that feeding trial.

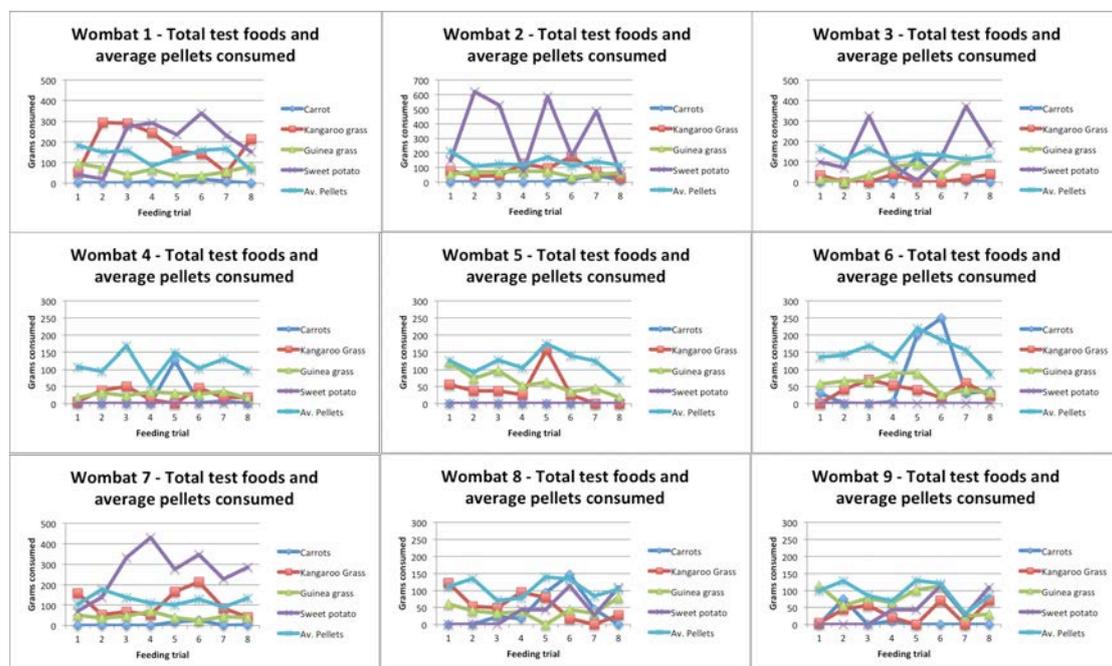
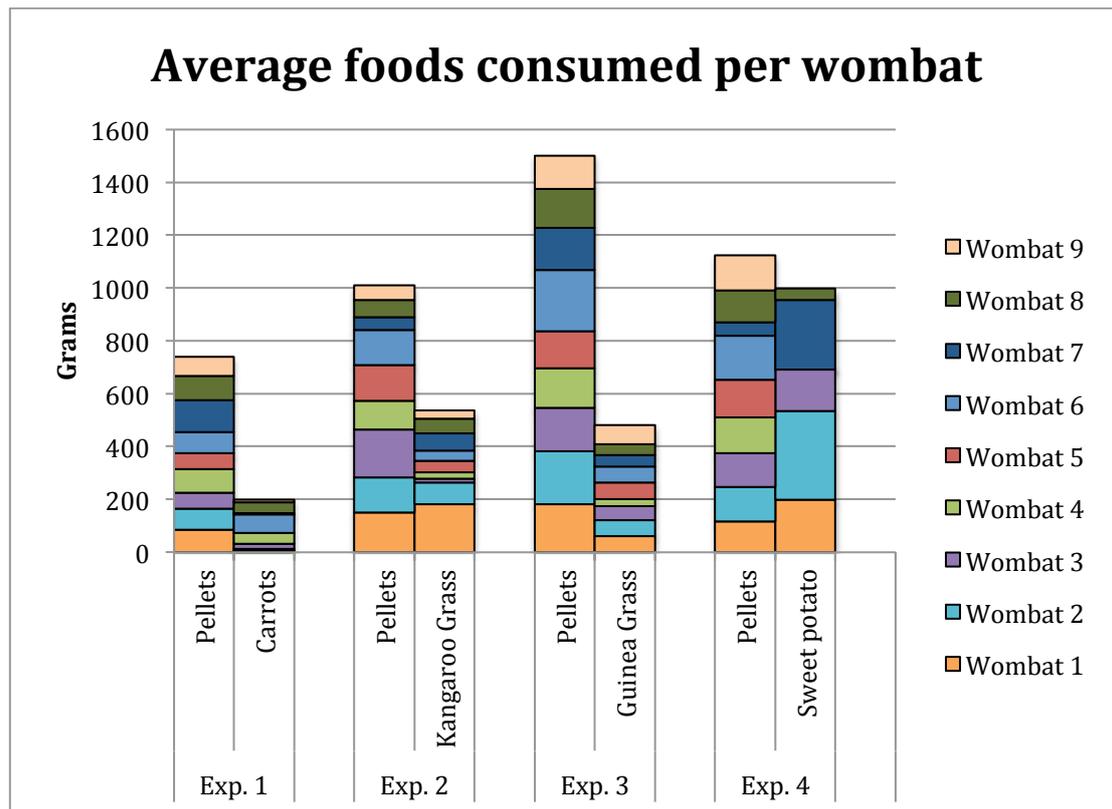


Figure 3.13: Total test foods and average pellets consumed by each wombat



**Figure 3.14: Average foods consumed by all wombats for all experiments**

There were significant differences between wombats for preferred food items (Figure 3.14). In Experiment 1, Wombat 5 did not consume any carrot during all eight feeding periods. Wombat 6 consumed a large amount of carrot, 1, 554 g during all eight feeding periods. In Experiment 1, the unnatural/familiar food (carrot) was selected as the preferred food item by eight of the nine wombats with an average consumption of carrots for all feeding periods being 205.9 g ± 61.93 g (Max. 554.0 g, Min 0.0 g).

During Experiment 2 (Kangaroo grass - natural/unfamiliar), all nine wombats consumed some grass during the eight feeding periods. The average consumed was 593.4 g ± 180.64 g (Max. 1729.0 g, Min. 129.0 g). In Experiment 2, Wombat 1 consumed the most grass, while Wombat 3 consumed the least. The average amount of the alternative food item to pellets consumed during each feeding period in Experiment 2 is greater than twice the average amount of the alternative food item consumed in Experiment 1.

The guinea grass (natural/familiar) food in Experiment 3 was again selected by all nine wombats over the course of eight feeding periods with an average consumption of  $421.6 \text{ g} \pm 36.22 \text{ g}$ . Wombat 5 consumed the most guinea grass with 506.0 g consumed while Wombat 4 consumed the least with 217.0 g during the eight feeding periods.

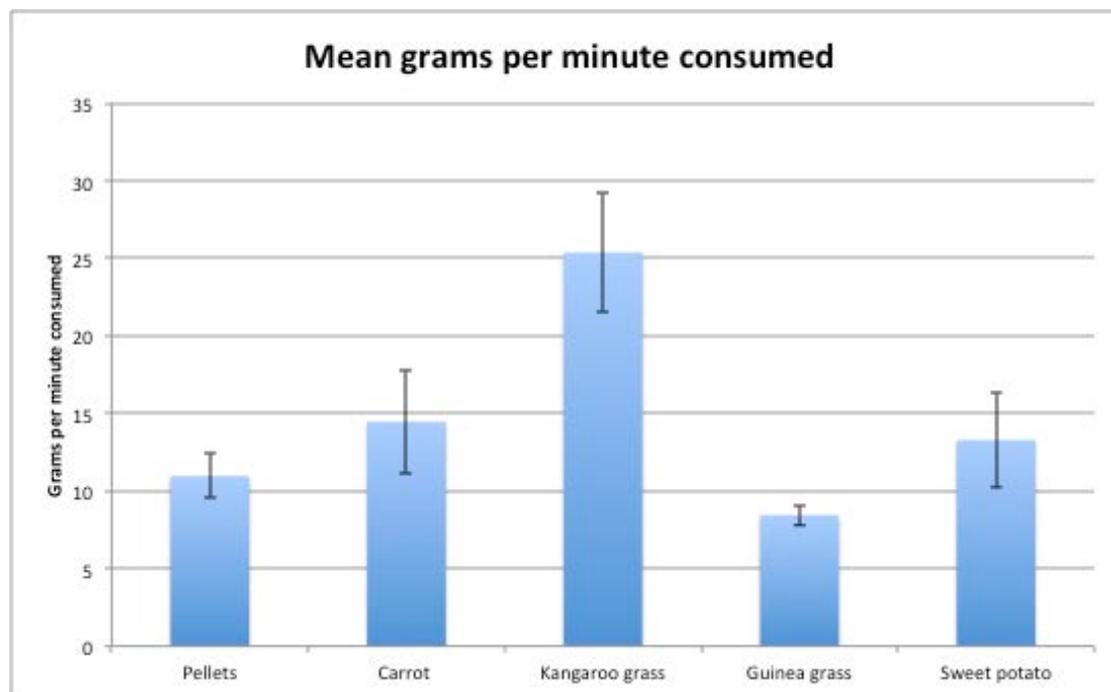
Data for Experiment 4 were highly variable among wombats. Four of the nine wombats (Wombat 2, 6, 7 and 8) did not consume any sweet potato during the eight feeding periods. The other five wombats, however, consumed very large amounts of sweet potato. Wombat 3 consumed the most sweet potato with 2687.0 g consumed during the eight feeding periods. This value is 400.0 g greater than the maximum values for the consumption by this wombat of the three other food items combined. All nine wombats chose both types of grass when offered (natural/familiar and natural/unfamiliar) but still ate more pellets.

*Time taken and test foods consumed relative to pellets.*

Table 3.3 presents the speed at which each individual wombat consumed the different food items. The data are presented as grams per minute and were calculated using the total amount of food consumed divided by the total amount of time spent eating that food item. The wombats were generally faster at consuming the kangaroo grass than the other food items (Figure 3.15). The harder feeds (pellets, carrots and sweet potato) took the wombats longer to eat than kangaroo grass. The guinea grass was the slowest food consumed.

**Table 3.3 – Grams per minute consumed**

<b>1</b>	10.60	7.80	26.97	7.43	15.80
<b>2</b>	21.12	20.34	28.39	11.94	27.61
<b>3</b>	9.09	13.37	36.51	11.30	16.33
<b>4</b>	8.33	11.17	18.12	8.63	20.00
<b>5</b>	8.85	0.00	40.92	7.50	0.00
<b>6</b>	13.55	20.68	38.62	8.79	7.83
<b>7</b>	8.88	10.22	15.95	6.71	15.17
<b>8</b>	6.94	11.27	12.97	7.41	16.80
<b>9</b>	11.40	35.34	9.91	6.30	0.00



**Figure 3.15: Mean grams per minute consumed**

#### *Environmental factors*

Aside from the nature of the food choice itself, two other experimental conditions may have influenced the results - temperature and humidity. There

Chapter 3 – Magnitude and frequency of food preferences of captive southern 69 hairy-nosed wombats

was no discernable influence of temperature on food consumption or choice with similar temperatures in Experiment one and Experiment three producing contrasting results (Figure 3.16). The results indicate that higher average humidity may result in lower food consumption (Figure 3.17). However, the highest two average humidity observations produced the least (Experiment 1) and the most (Experiment 4) food consumed.

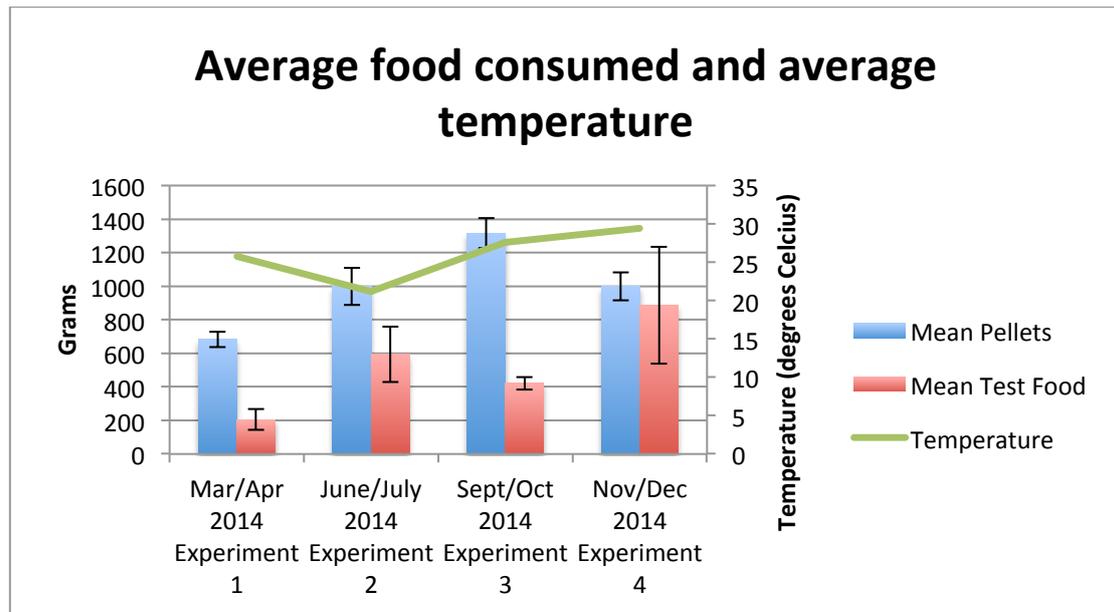


Figure 3.16: Average foods consumed and average temperature

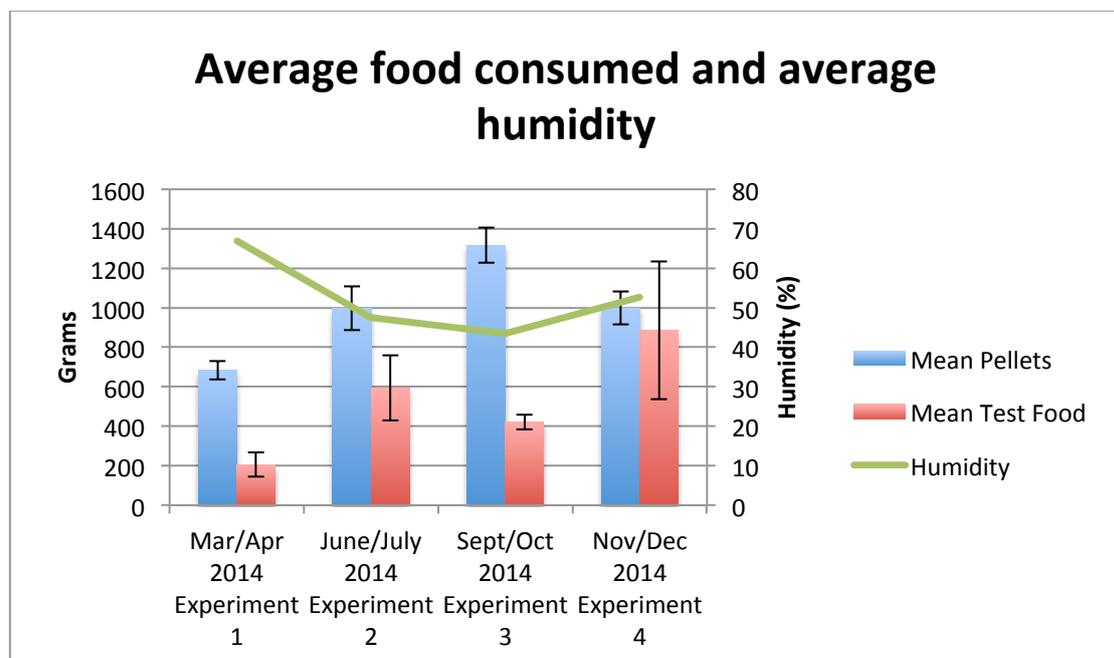


Figure 3.17: Average foods consumed and average humidity

## Discussion

The study was conducted to explore food choices in the captive SHNW. The experiment was designed to determine whether individual wombats would choose a second food item when offered in combination with their normal pelletised formulated diet. Four food choice options were compared and the choices were replicated using an experimental design that enabled nine wombats to be monitored for their food choices over eight feeding trials per food type. The use of this replicated design provided data that addressed questions of factors that determine food preferences in relation to familiarity and naturalness.

The results demonstrate that the wombats chose to eat food items other than the commercial pelleted diet, indicating that they had obvious preferences of dietary choices. Secondly, the wombats ate significant amounts of the test food items assessed by both magnitude and time spent eating, indicating that consumption of the test food items were not just random events by chance. Thirdly, there were individual differences in these choices among wombats.

An important finding of this study was that the control test food item (pellets – unnatural/familiar) was consistently chosen in preference and magnitude compared to any of the other test food items (carrot, kangaroo grass, guinea grass or sweet potato) and that the test food items seemed to influence the amount of pellets consumed. The familiar/unnatural (carrot) test food item negatively influenced feeding behaviour with little carrot and significantly less pellets consumed despite both items being part of their familiar feeding regimes. If carrot was the only other food provided to wombats, there was a marked disincentive to eating. The combination of pellets (unnatural/familiar) and guinea grass (natural/familiar) produced the highest average consumption of pellets and the most even choice of both food items across the two choices provided during Experiment 3. A similar result occurred in Experiment 2 with the combination of pellets (unnatural/familiar) and kangaroo grass (natural/unfamiliar) inducing an even choice within both food items. It would appear that if the combination of unnatural (pellets) and natural (in the form of

grasses) were provided as a food choice then familiarity/novelty was irrelevant. This group of captive wombats were balancing the unnatural pellets with natural grassy material as a priority.

This conclusion is supported by the frequency data (Table 3.1) where the majority of wombats chose to eat pellets, guinea grass or kangaroo grass across the majority of trials compared to carrot or sweet potato. Less than half of the wombats half of the time chose to consume carrot which also decreased pellet consumption and overall food intake. The other unnatural food item, sweet potato, also showed inconsistent feeding preference despite some wombats choosing to eat a lot of it. Unlike the pellets and grasses, sweet potato was only consumed during 50% of the feeding trials. Ultimately, a combination of pellets (unnatural) and either of the other unnatural items (carrot or sweet potato) regardless of familiarity, induced the most variable and polarising feeding behaviours in these wombats.

The marked individuality of the food choices made in these experiments help to demonstrate that wombats are able to make choices. This is demonstrated in two ways i) by describing the relative amount eaten and ii) by whether the alternative food was selected at all. Offering a varied diet would increase the wombats' ability to meet their individual needs and desires regarding taste, textures, moisture content and nutritional requirements.

For example, if wombat 5 lived in a facility that only offered pellets and hard foods, such as carrot and sweet potato, then this wombat may struggle to maintain a healthy body weight, or may suffer in ways not readily observable due to the limited choices of preferred food options for this individual. This would impact upon the welfare state of this individual, even though other wombats housed at the same facility on the same diet may have more positive welfare states. The lack of appropriate choice of foods for this individual presents the wombat with a dilemma rather than a choice. The facility may be perceived to be meeting the animals welfare needs with regard to providing adequate nutrition and a variety of food choices, however, this particular wombat does not particularly enjoy eating any of the foods offered. This would

have a negative impact on this individual's welfare state. By offering live growing grass as an additional food choice, the welfare state of this animal would be greatly improved. It will still receive adequate nutrition through the food options available, but now it has a choice to consume a food that it prefers over the other options. Since the wombat has more control over its decisions, welfare is improved.

Given that guinea grass was consumed as reliably as pellets by all wombats it could be argued that this was the preferred food of the four test food choices offered. This would confirm the hypothesis that the wombats prefer natural/familiar food over unnatural/unfamiliar food. However, when taking into consideration the individual preferences, guinea grass would not be preferred by all wombats all of the time. For example, Wombat 7 consumed an average of 44 grams of guinea grass and 158 grams of pellets per feeding trial during experiment 3. In contrast, the same wombat consumed an average of 263 grams of sweet potato and 51 grams of pellets per feeding trial during experiment 4. It is clear from these figures that Wombat 7 preferred sweet potato over both pellets and guineas grass.

However, the larger average consumption variance for sweet potato indicates marked differences in individual wombats' preferences for this particular food item. Only five of the nine wombats chose to eat sweet potato. These five wombats consumed enough sweet potato that the overall results were skewed to the extent sweet potato was the food item of preference, however, it is important to recognize that four of the nine wombats avoided sweet potato completely.

The time it takes to consume food may influence food choices. Wombats in their natural habitat spend time grazing on different quality grasses (Triggs, 2009). Rarely would a wombat in a natural habitat have access to a nutrient dense feed in ample quantity. It is therefore interesting to examine the time taken by this group of wombats to consume the test foods offered. The difference between the speed at which both grasses were consumed is interesting. The reason for this difference could be in the presentation of the grasses. The kangaroo grass was presented in the trays it had been grown in. This required the wombats to

physically graze the growing grass as if it were growing in the earth. The guinea grass was picked fresh and presented to the wombats on a plate lying flat. Another factor that could explain this difference in speed of consumption could be the difference in the leaf width of these two grass species. Kangaroo grass is a thin-stemmed grass, while guinea grass has a broader leaf in comparison. Perhaps the speed at which they consumed the kangaroo grass was a demonstration of their efficient grazing technique that has evolved over thousands of years. Wombats in their natural habitat need to be efficient grazers in order to consume the quantity of grass required to meet their nutritional needs. Therefore, the results in this study could simply reflect the wombats' natural ability to graze on grass.

Another interesting finding is the individual differences between the wombats in their rate of consumption. These results mirror those reported by Zinner (1999) with a group of Hamadryus baboons (*Papio hamadryus*) in a zoo environment. The baboons food intake rates deviated between 8% and 50% between the subjects (Zinner 1999).

Wombat 9 was slightly faster at eating pellets than kangaroo grass, when all of the other wombats were faster at eating the grass. Wombat 5 was 4.6 times faster at eating kangaroo grass than eating pellets, but slower at eating guinea grass. Wombat 5 did not consume any carrot or sweet potato. From this information we could draw two different conclusions. Firstly, that wombat 5 is an efficient grazer of grass presented naturally but has difficulty in consuming grass that is presented lying flat on a plate. Secondly, wombat 5 either doesn't like the harder foods (carrot and sweet potato), or finds them difficult to eat and thus avoids them.

In contrast, it is also interesting to note how slow the wombats were at consuming guinea grass. As mentioned previously, this could be due to the presentation of the grass, or the width of the leaf. Further study would be required to determine which of these factors caused the slower consumption rate. This information could help inform zoos about how to present grass as a

food choice item. For environmental enrichment purposes, it may be better to provide the wombats with picked, broad leafed, grasses presented lying flat, just as the guinea grass was presented in this series of feeding trials. Presented in this manner, the wombats take longer to eat it, so they will be kept busy for larger parts of their waking hours.

The importance of food choice for captive animals has been recognised from both dietary and welfare perspectives (Provenza, Villalba et al. 2003). Provenza, Villalba et al. (2003), found that offering food choices provided for several benefits for the animals. Individuals that are allowed to select among alternatives are better able to meet their individual nutritional needs, regulate intake of toxins and these choices account for their individual differences in morphology and physiology. The overall performance of the group is improved by allowing the uniqueness of individuals to manifest through selective food choices (Provenza, Villalba et al. 2003). This has been clearly demonstrated in this study. The wombats consumed more of the control food item during Experiments 2 and 3 when grasses were provided across all individuals. When sweet potato was administered, five wombats chose to consume a large quantity of both the test and control food item. In contrast, carrot negatively influenced feeding drive on average even for pellets.

Primary producers of domesticated farm animals and the zoo industry managers have different incentives. Primary producers need to ensure best practice animal management to improve the production outputs of the animals in their care. Research in the field of varied diets and selective grazing strategies, therefore, is becoming increasingly accepted and applied to farm animal husbandry (Provenza, Villalba et al. 2003). In the zoo industry, production outputs are not a major incentive, thus, the study of preferences of individual animals with regard to food choices and selective grazing strategies has not occurred to any great extent. A more important incentive for the zoo industry is animal welfare and conservation of threatened species through captive breeding and public education programs. While the incentives for these two industries are different, the results of offering food choices are beneficial to both. These captive wombats are shown to be negatively impacted when carrot was provided

as a food choice. In this series of experiments carrot appeared to be a disincentive to eating even the control food item.

In contrast to the findings of Villalba et al. (2009), in which it was demonstrated that the herbivores that were studied preferred familiar to novel foods and had a reluctance to eat novel foods, the wombats in this study were curious enough to explore their nutritional options even when it contained novel food items with unknown post-ingestion outcomes.

In their book, *The Nature of Nutrition*, Simpson and Raubenheimer (2012) describe the capacity of animals to determine an optimal nutrient intake during dietary manipulations and nutritional challenges (such as food choice experiments) by using three strategies for regulation of food item consumption: assessing the nutritional quality of the foods available, assessing the animal's own nutritional state, and, having the innate biological systems to ascertain nutrient needs and alter dietary intake to address these needs (Simpson and Raubenheimer 2012). The wombats in the present study had access to a nutritionally balanced food item – pellets. Having been sustained by feeding the same pellets for many years, the post-ingestion biological feedback systems when the pelleted diet was consumed would be well ingrained in these wombats. Likewise, the biological systems sensing nutritional qualities and post-ingestion consequences of carrots and guinea grass would have been ingrained in these wombats. The experiments that offered these familiar food choices demonstrated that the wombats chose to consume differing amounts of each food item, which demonstrates the ability of these wombats to regulate intake according to individual nutritional needs and/or preferences. The sensing of nutritional qualities of sweet potato and kangaroo grass through innate biological systems would not have occurred in the wombats of the present study. The wombats appear to need to sample these items before the biological sensing of the qualities of these food items could occur. Some wombats completely avoided sampling sweet potato for all eight feeding periods while others consumed considerably more sweet potato than all other food items combined. Because all wombats were in the same nutritional state (weight and health status) prior to commencing these feeding experiments, the nutritional value of

the sweet potato cannot be considered the primary factor determining the amount of sweet potato consumption. The biological sensing of wombats that chose to eat sweet potato must have been controlled by some other factor than nutritional value of this food item. Likewise, the wombats that avoided sweet potato must have done so due to biological sensing that is not related to nutrition. In applying Simpson and Raubenheimer's theory from a biological perspective of the wombats included in the present study, wombat consumption of a food item would have been regulated by three biological senses before sampling the sweet potato: (1) Perceived nutritional composition of the food item (2) Required nutrients for the optimal function of their innate biological systems and (3) Amount of food item that needed to be consumed to satisfy the specific nutritional needs of each animal (Simpson and Raubenheimer 2012). The five wombats that avoided sweet potato may have sensed there was a risk of consuming the novel food that overrode the possible benefits of consuming this food item, while the remaining four wombats' sensory systems were activated in ways that the benefits overrode the risk of consuming this food item.

There may have been other factors influencing these different preferences about food item consumption. Behavioural traits of individual wombats may have a role in food choice. The field of animal personality research is growing rapidly with many researchers describing animals within the same species as possessing different personality traits (Wemelsfelder 2007). With the present study, the wombats that avoided the novel food may have more cautious personality traits when compared with the more bold wombats that quickly explored the novel nutritional environment provided.

There is a strong focus in zoos to design habitat centric exhibits that offer the inhabitants a dynamic physical environment and a strong emphasis is placed on enrichment and training programs to further enhance the physical and mental stimulation of the captive animals (Zoo and Aquarium Association 2016). Increasing opportunities for food choice among individuals will contribute to an enriched nutritional environment.

Just as there is more to nutrition than nutrient consumption (i.e. foods vary in colour, texture, flavour, size, shape), there is more to wombats than just their taxonomic description. Individual wombats are unique in neurology, morphology, physiology and through their individually different environmental (all factors other than genetics) factors to which they have been exposed (nutrition, learning experiences, etc.) throughout life through the epigenetic regulation of their behaviour and physiology. All of the epigenetic and genetic factors combine to create individual physiology and behavioural differences in the wombats. By having uniqueness of individuals within the group, the group as a whole will be healthier and express more typical behaviours and physiology within a captive environment. Increased food choices for animals in zoos will allow for an enriched nutritional environment that enhances the capacity of individuals to express innate differences in their behaviour and physiology within the group.

Throughout this research project, all the wombats were in the same nutritional state, having been maintained on the same diet daily for the past 14 years (excluding the two wombats born at the zoo, who are aged 4 and 10 respectively). Certain aspects of the physiology of the experimental animals, however, could not be accounted for, such as age or reproductive status. Of the seven wombats of undetermined age, two were male and five were female. Four of the females were considered to be post-reproductive due to an absence of oestrous cycles in recent years. One of the older females (Wombat 9) and the 10 yr old female (Wombat 1) were both considered to be reproductively sound. However, the relative average consistency of consumption across all wombats for pellets during each of the experiments shows that these factors may have little influence on this group of wombats.

There could be a correlation between decreasing humidity levels and increased food consumption, however changes in temperature showed no impact upon food consumption or preference. For example the average temperature was the same during experiment 1 (carrots) and 3 (guinea grass) yet vastly different consumption of both the test food item and pellets occurred (Figure 3.16). Wombats were clearly being driven by factors other than temperature. The

influence of humidity cannot be completely ruled out with the highest average humidity associated with the lowest food consumption and the lowest average humidity conditions associated with the highest average food consumption (Figure 3.17). This is a limitation of this study and should be explored in the future. One method that could be considered to control for seasonal differences would be to randomise the test foods across all four experiments. However this also appears to be a novel finding with no literature to support or refute this observation even in domesticated livestock species of changes in food consumption in grazing herbivores to fluctuations in humidity.

Finally, the stress of the enclosure conditions was controlled for by allowing the wombats a two-week acclimatisation period prior to the commencement of experiment one. During this two-week period the wombats were given free access to explore the feeding pen area in their own time. This acclimatisation period also included familiarity with the presentation of the control and test food items. They were also fed their usual zoo diet in the feeding pen during this time (which included pellets and carrot). There were no discernable differences in food consumption during the pre-trial period compared to the trial period.

The variability in individual food intake could be associated with individual dietary needs, however, neurological, physiological and morphological differences between wombats cannot be discounted. Further investigation would be required to explore how these other factors might be associated with food preferences.

The present study has demonstrated a reliable methodology for studying the food preferences of captive SHNWs and has yielded significant results indicating clear food preferences of individual wombats. The factors controlling these individual differences in food preferences among wombats require further exploration to determine more fully the implications of providing dietary options for captive wombats. The original hypothesis was partly disproven with naturalness rather than familiarity established as the more potent influence on food preference. For this group of captive wombats, carrot should be reconsidered as a more infrequent feeding option given the negative impact on

overall food consumption during the test period and fresh grasses should form an essential element of captive diets. This is an obvious conclusion supported by the data for grazing herbivores. Other unnatural food options should be considered on a case by case basis providing a more tailored feeding regime.

### **Acknowledgements**

The authors acknowledge the support of Rockhampton Regional Council and the Rockhampton Zoo for providing the facility and the animals for this study. Thanks also to Judy Couper and Mackenzie Hansler for their assistance with the dry matter data collection. The following people assisted with data collection at various times and for this the authors offer sincere thanks and gratitude; Lauren Thompson, Heather Campitell, Alexis DuPont, Tiffany Palmer and Andrew Fenning.

## References

- Barboza, P. S. 1989. The Nutritional Physiology of the Vombatidae. Doctor of Philosophy PhD Thesis, The University of New England.
- Barboza, P. S. 1993. Digestive strategies of the wombats: feed intake, fiber digestion, and digesta passage in two grazing marsupials with hindgut fermentation. Physiological Zoology **66(6)**: 983-999.
- Barboza, P. S. 1993. Effects of restricted water intake on digestion, urea recycling and renal function in wombats (Marsupialia: Vombatidae) from contrasting habitats. Australian Journal of Zoology **41**: 527-556.
- Barboza, P. S. and I. D. Hume 1992. Digestive tract morphology and digestion in the wombats (Marsupialia: Vombatidae). Journal of Comparative Physiology B **162**: 552-560.
- Barboza, P. S. and I. D. Hume 1992. Hindgut fermentation in the wombats: two marsupial grazers. Journal of Comparative Physiology B **162**: 561-566.
- Barboza, P. S., I. D. Hume, et al. 1993. Nitrogen metabolism and requirements of nitrogen and energy in the wombats (Marsupialia: Vombatidae). Physiological Zoology **66(5)**: 807-828.
- Descovich, K., A. Lisle, et al. 2012(a). Differential responses of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*) to the presence of faeces from different species and male and female conspecifics. Applied Animal Behaviour Science **138**: 110-117.
- Descovich, K., A. Lisle, et al. 2012(b). The effect of group size on vigilance and activity budgets in a captive solitary marsupial (*Lasiorhinus latifrons*). Brisbane, The University of Queensland: 73-90.
- Descovich, K., A. Lisle, et al. 2012(c). Space allowance and the behaviour of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*), The University of Queensland.
- Finlayson, G. R., G. A. Shimmin, et al. 2005. Burrow use and ranging behaviour of the southern hairy-nosed wombat (*Lasiorhinus latifrons*) in the Murraylands, South Australia. Journal of Zoology **265**: 189-200.
- Hume, I. D. 1989. Nutrition of marsupial herbivores. Proceedings of the Nutrition Society **48**: 69-79.
- Hume, I. D. 1999. Marsupial Nutrition. London, Cambridge University Press.

Chapter 3 – Magnitude and frequency of food preferences of captive southern hairy-nosed wombats 81

- Provenza, F., J. J. Villalba, et al. 2003. Linking herbivore experience, varied diets, and plant biochemical diversity. *Small Ruminant Research* **49**: 257-274.
- Simpson, S. and D. Raubenheimer 2012. The Nature of Nutrition: A unifying framework from animal adaptation to human obesity. Princeton, Princeton University Press.
- Treby, D. 2005. Southern hairy-nosed wombat (*Lasiorhinus latifrons*) husbandry manual, Australasian Zoo Keeping: 54.
- Triggs, B. 2009. Wombats: second edition. Collingwood, CSIRO Publishing.
- Wemelsfelder, F. 2007. How animals communicate quality of life: the qualitative assessment of behaviour. *Animal Welfare* **16(S)**: 25-31.
- Woolnough, A. P. 1998. The feeding ecology of the northern hairy-nosed wombat, *Lasiorhinus krefftii* (Marsulialia: Vombatidae). PhD, James Cook University.
- Zoo and Aquarium Association, A. 2016. ZAA website. Retrieved 5/5/16, 2016, from <http://www.zooaquarium.org.au/>.

## Chapter 4

### Food intake and activity in captive southern hairy-nosed wombats

#### Abstract

The research conducted in the present study examined the individual differences in activity during feeding of a group of captive wombats. Although there has been research on the daily activity budgets of captive southern hairy-nosed wombats (SHNWs), there has been no study on the effects of activity on food intake in this species. This study looked at activity during feeding time of nine captive SHNWs. The hypotheses were, 1) Captive SHNWs would exhibit individual differences in amount of physical activity during feeding, and 2) SHNWs with greater activity ratios would have different food intake rates as compared with wombats with lesser physical activity ratios.

The research was based at the Rockhampton Zoo Wombat Research Centre and involved a series of four experiments. The foods used were standard food items used in the diets of captive wombats throughout Australian zoos.

The results indicate significant differences in amounts of activity among wombats. Wombats with greater activity ratios consumed significantly more food than wombats with lesser activity ratios ( $P = 0.0082$ ). Understanding the individual differences among wombats in captivity may have implications for improving the captive management of animals of this species and may lead to more positive welfare outcomes for this species in captivity.

#### Keywords

Active, Sedentary, Physical activity and food intake

## Introduction

The SHNWs in their natural habitat are grazing herbivores. They feed on grasses that are low in nutrient value (Triggs, 2009). Wombats have developed unique behavioural and physiological adaptations that allow them to cope with this low value diet. These can be broadly explained through four main features; 1) lesser basal metabolic rate than most animals (Barboza, 1989; Barboza and Hume, 1992), 2) efficient masticatory system (Hume, 1999), 3) digestive adaptations of colonic fermentation and slow passage of digestion (Hume, 1999), and 4) a sedentary and burrowing/underground lifestyle (Finlayson, Shimmin et al., 2005). It is the adaptation to a sedentary lifestyle, which was of most interest in the present study, as it seems likely there is a link between energy requirements and activity levels.

Wombats remain in a single home range for their lifetime (Hume, 1999). They do not migrate with the seasons like other similar sized marsupial grazers such as kangaroos. This may be due to the wombats' reliance on burrows for thermal regulation (Finlayson, Shimmin et al., 2005). Building new burrows may expend greater energy than the energy available in their low quality diet, so the SHNWs rarely build new burrows relying instead on maintaining existing burrows (Barboza, 1989).

This sedentary lifestyle means that during poor seasons of the year wombats are subjected to a low quality and quantity of grass (Evans, 1999). A 2005 study site in South Australia supported 72 individual SHNWs (during the 8 month study). This was a surprising large number of wombats given the arid conditions and the large weight size of the wombats (22-32 kg; Finlayson, Shimmin et al., 2005). In contrast, a study of common wombats in New South Wales, where the forage was better in quantity and quality than the South Australian site, found that there were only five common wombats in an area of similar size (Evans, 1999). Over the duration of the study the SHNWs of South Australia showed little variation in body weight. This ability to sustain high population densities even in arid conditions is testament to the SHNWs environmental adaptations.

The SHNWs in their natural habitat slow their metabolic rate, reduce activity and remain underground for longer periods to cope with seasonal changes in food availability (Triggs, 2009). In a captive environment wombats don't experience seasonal fluctuations in food quality or availability. Thus, in captivity, there is no

driver telling the wombats to limit activity or slow their metabolism. This *ad libitum* access to good quality food year round may have unintended negative consequences for SHNWs in captivity. Some negative welfare indicators have been identified with this species in captivity. The most pressing issue for the captive SHNW is a lack of breeding success (Nicolson, 2012) which may be linked to inadequate captive husbandry techniques for this species that have not yet been discovered through science. Even though there have been careful comparisons of successful and unsuccessful husbandry regimens there is no recommended modification to husbandry which could reliably improve reproductive performance (Nicolson, 2012).

Captive SHNWs are managed by a national studbook. This is held and updated by the national species coordinator, currently Mr. Gert Skipper of Adelaide Zoo. The captive SHNW population in Australia is 58 animals: 22 males and 37 females (ISIS 2016). Only six joeys have been born in captivity in the last 10 years. Within that same time period almost three times that number of wombats have died (unpublished zoo records). This species requires a breakthrough in reproductive success or continued supplementation of wombats captured from the wild if the captive population is to be maintained (Nicolson 2012). Ethical questions are raised by this decreasing captive population size as some animal welfare groups argue that reproductively impaired animals experience compromised welfare (Nicolson 2012).

Three previous research projects conducted at Rockhampton Zoo Wombat Research Centre focused on reproduction and/or behaviour, though none of these projects resulted in increased fecundity. It is likely that fecundity is linked to food resources (Woolnough and Foley, 2002). While captive husbandry techniques have not been proven to affect reproductive performance, they cannot be discounted (Nicolson 2012). It is, therefore, time to thoroughly investigate all aspects of captive wombat husbandry. Further research is needed to examine diet preferences, behavioural adaptations to captivity and individual differences among wombats. This is what led to the development of this study on activity levels during feeding of this group of captive SHNWs.

Wombats that were in their natural habitat had minimal energy expenditure (Finlayson, Shimmin et al., 2005). More than 75% of their time was spent underground. Above-ground activity periods were short in duration and limited to

nocturnal hours (2-8 hours per day; Finlayson, Shimmin et al., 2005). The distances travelled by the wombats were small with the mean distance travelled 221 m per night. No significant difference was found in distance travelled between sexes or across seasons (Finlayson, Shimmin et al., 2005).

Hogan's 2009 study on activity budgets of SHNWs housed at Rockhampton Zoo showed very similar results to those of the Finlayson et al. (2003, 2005) study. Wombats in their natural habitat were active above ground for  $\approx 26\%$  of the day. Foraging was the dominant behaviour when the wombats were out of their burrows (Finlayson, Shimmin et al., 2003; Finlayson, Shimmin et al., 2005). The captive wombats in Hogan's (2009) study were active for 18.4% of the day. Feeding was the dominant activity during when they were out of their burrows (Hogan, Phillips et al., 2009). Hogan suggested that the lesser physical activity of captive wombats was due to the reduced need to forage in search of food compared to the wombats in a natural environment. Hogan also suggested that the lesser physical activity of the captive wombats could be due to reduced stimulus diversity in the captive environ (Hogan et al., 2009).

Mean emergence times from burrow differed between the wombats in their natural habitat (1911 hours, Finlayson et al., 2003) and the captive wombats ( $\approx 1700$  hours, Hogan et al., 2009). This difference could be attributed to the different climatic conditions between the two study sites (Murraylands, South Australia and Rockhampton, Central Queensland). Emergence times from burrows in the captive wombat study (Hogan 2009) were used to inform this project, as the animals used in the present study were the same animals as those used in the previous study.

The hypothesis states that SHNWs with higher activity ratios will consume greater amounts of food to accommodate for the added energy expenditure, while less active wombats will consume less food.

The study design allowed for a series of observations around activity during feeding and the following aims were established-

9. To examine if physical activity levels differed between wombats.
10. To examine whether there is a relationship between activity levels and feeding behaviour.

11. To examine if there were differences in activity levels for each food choice.

## Methods

### Animals

Nine SHNWs (*Lasiorchinus latifrons*) housed at the Rockhampton Zoo Wombat Research Centre were used for this research.

The wombats used in this study have resided at the Rockhampton Zoo Wombat Research Centre for several years prior to this study. Two of the wombats were born at the zoo in 2004 and 2010, respectively, while the remainder of the wombats were born in the natural habitat of SHNWs before being captured. These wombats that were born in the natural habitat were trapped and transported to the zoo in 2001 for the zoo's research program. Nine of the zoo's 11 southern hairy-nosed wombats were used for the present study. The two animals excluded were display wombats residing in a separate area of the zoo. All nine animals were long-term residents of the Research Centre and had been in group housing arrangements with other wombats for extended periods of time. The research animals comprised of three males and six females. Apart from the two captive bred wombats, the ages of the other wombats were unknown.

All of the wombats were in good health and body condition, as declared by the zoo veterinarian, with a mean weight of  $30.5 \pm 4.4\text{kg}$ . The wombats were weighed fortnightly in accordance with standard zoo practice to ensure maintenance of healthy body weights.

The procedures employed by this project were approved by the CQUniversity Animal Ethics Committee (approval number A12/11-290).

### Experimental facilities

The wombats were housed in Rockhampton Zoo's purpose built Wombat Research Centre, which comprises four enclosures. This facility has been described previously (Descovich et al., 2012a; Descovich et al., 2012b; Descovich et al., 2012c).

Three enclosures housed two wombats each and the other housed a group of three wombats. Three enclosures housed mixed sex cohabitants, and one a single sex pair of females.

The effects that group feeding had on individual behaviour were eliminated by removing each wombat from its group for 30 minutes per day and moving it through a series of tunnels into a 3 m x 3 m feeding pen. During this 30 minute period each wombat was fed and then observed from an elevated position.

Prior to the commencement of the four experiments the wombats were given a 2-week acclimatisation period to explore the feeding pens in their own time. This was to familiarise the wombats with the feeding pen area, the sugar cane mulch substrate that was being used throughout the experiments, and to ensure that the wombats were not stressed when entering and exiting the feeding pens through the tunnels.

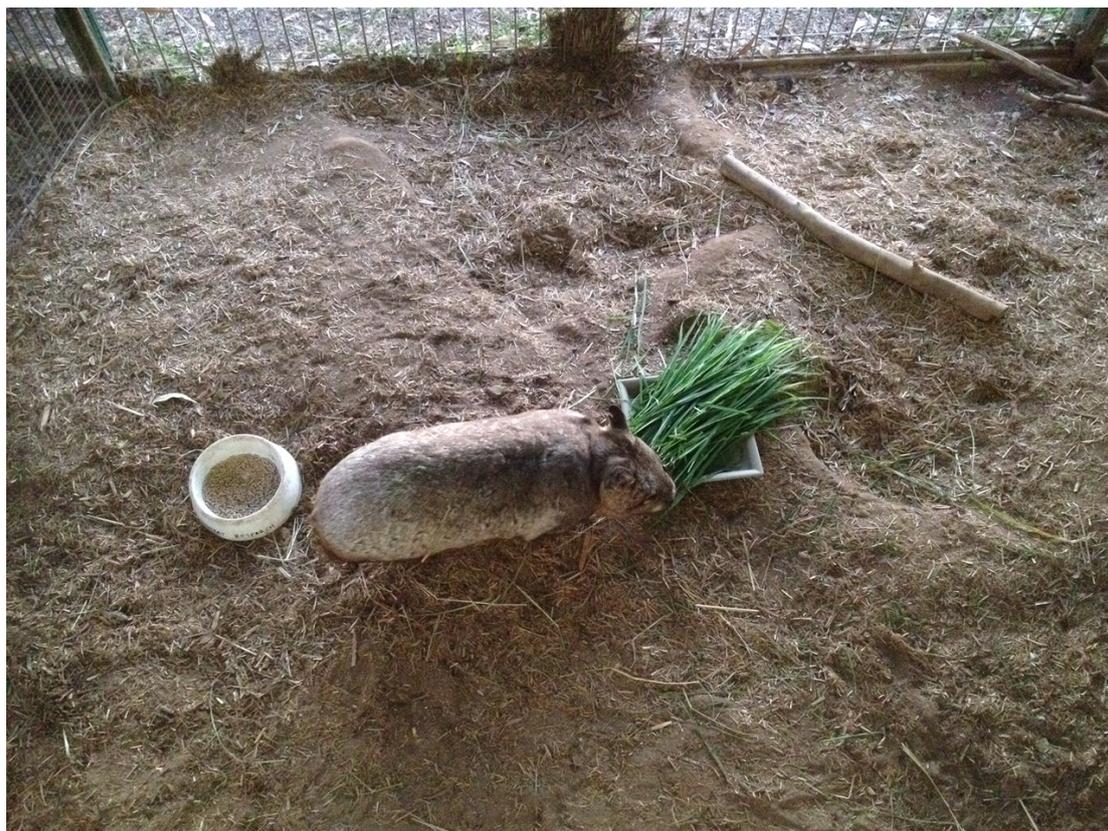
The experiments commenced in the evening at the time when the wombats were beginning to exit their dens to begin their nocturnal activities. In winter, this was as early as 1530, and in summer was as late as 1800. The wombats were not forced into the feeding pen, but rather encouraged to enter by restricting access to their normal enclosure and opening access to the feeding pen. The wombats' curiosity drew them to explore the newly opened tunnel, so there was never a need to manually manoeuvre the wombats through the tunnels.

The feeding pens were located at the back of the Research Centre. The pens were connected to the dens via a series of tunnels such that each wombat could exit its den and follow a tunnel system through to the feeding pen.

Once a wombat had entered the feeding pen, a sliding door was closed off within the tunnel, so that the wombat still had access to part of a tunnel, but could not return to its normal enclosure until released at the end of the 30 minute feeding period.

Wombats were selected at random before the commencement of each evening's feeding periods. No more than five wombats were studied on any night due to the length of time of the feeding periods and the associated time involved in moving the wombats to and from the feeding pen.

Two food items (pellets and a test food item) were placed in similar bowls 1 metre apart in the centre of the feeding pen. The test food items used were carrot, kangaroo grass, guinea grass and sweet potato. The placement of each food item was randomised so that sometimes the pellets were on the right and sometimes they were on the left (Figure 4.1).



**Figure 4.1: Location of feeding bowls within the feeding pen**

Feeding of the experimental food items did not occur in rainy weather. The wombats were reluctant to exit the dens if it was raining, even though the feeding pens were undercover and the wombats did not need to go through the rain to get to the feeding pen.

The amount of food offered to each wombat did not exceed the recommended daily dietary intake. For each feeding period, 300 g of pellets were presented and up to 300 g of the other food item. On some occasions, more than 300 g of the test food item was fed out. This was done to ensure the wombats' did not run out of food during their 30 minute feeding period. Following the completion of the feeding periods each evening, the wombats were fed the balance of their daily ration in their usual feeding location. That is, if the wombat consumed 200 g of pellets during the feeding period, it was fed 100 g when it returned to its usual enclosure. This ensured all wombats had access to adequate nutrition for the duration of the study. It was important that excess food was not offered to avoid excessive weight gain during the study.

From the elevated position above the feeding pen, observations were made in a continuous fashion for the full 30 minute feeding period. The behaviours recorded were standard wombat behaviours such as eating, walking, digging, sitting, lying, and scratching and were derived from and then refined from previous behavioural observation studies conducted on this group of wombats by another researcher (Hogan, 2010). Behaviours indicative of a stress response in wombats such as vocalizing, pacing and escape attempts were also included in the observational data spread sheets. If a wombat exhibited stressed behaviour for more than 5 minutes within the 30 minutes feeding period, then that feeding period was ceased and the wombat returned to its regular enclosure. This occurred on only three out of a total of 288 feeding trials. These three feeding periods were conducted again at a later time when the wombats were not stressed. The movement of wombats to and from the feeding pens was part of the established regular daily husbandry procedures for this group of wombats. Therefore, this was a familiar pattern of activity for this group of wombats and the stress on the animals caused by this activity was minimal. During the pre-experimental period two other behaviours were included on the observation checklist, these being pacing and drinking. These two behaviours were, however, not seen during any of the feeding periods over the course of the four experiments and so were deleted from the observational checklist. Pacing is generally considered a negative welfare marker. Its absence from these observations is an indication that the wombats were not stressed by the experiments enough to perform a pacing behaviour. It is not surprising that drinking was not seen during these experiments, as wombats rarely drink. Even though water was available in the feeding pens it is likely that the wombats drank at times outside of their feeding period when they had access to their regular enclosures.

Below is a list of the behaviours as they were defined for this series of experiments.

1. Eating pellets – positioned at the pellet bowl and/or is actively chewing on pellets
2. Eating other – positioned at the other (grass, sweet potato, carrot) bowl and/or actively chewing on the other food item
3. Walking – walking around the feeding pen

4. Sitting – positioned with posterior body part on the ground and in a stationary posture
5. Lying – lying down but awake
6. Sleeping – lying down not awake
7. Digging – digging in the soil
8. Scratching – pawing/itching itself
9. Vocalising – producing a vocal noise
10. Sniffing – smelling the ground
11. Standing – standing upright but in a stationary posture
12. Escape attempt – digging at an exit point or attempting to climb out of the feeding pen

When the 30 minute feeding periods were not being conducted, the wombats were housed in their normal groups in their normal enclosures. Once each wombat had finished its feeding period and had been moved through the tunnel back into its usual enclosure it was given full access to its outdoor area as well as den system.

#### **Data Collection**

At the beginning of each individual feeding period, the food items were weighed and then any remaining food weighed at the conclusion of the 30 minutes feeding period. This enabled the amount of food consumed to be recorded as well as the time spent eating each of the food items. Eight 30 minute replicates were performed for each wombat. That is, each wombat had eight 30 minute feeding periods during this experiment.

At the end of the feeding period the data collected was used to determine the amount of time each wombat spent performing each behaviour. The food that had not been eaten during the feeding period was weighed, disposed of and the weights recorded. Temperature and humidity were also recorded at the time of the feeding trial.

### Statistical analyses

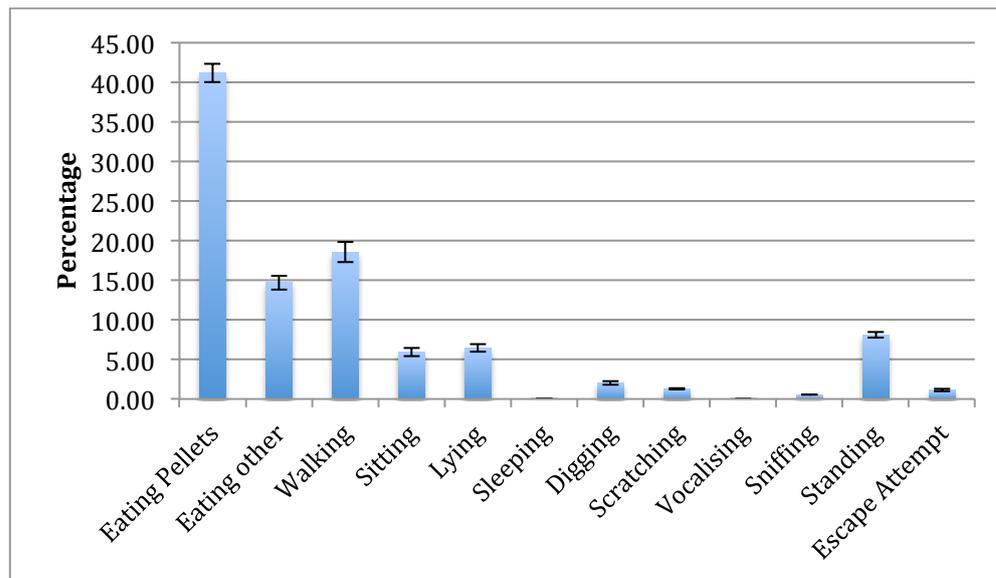
Raw data, means  $\pm$  SEM, ratios of behaviour and graphical representation of the data were created using Excel. Statistical analysis was performed in Graphpad Prism. T-tests probability significance used was  $p \leq 0.05$ .

Data were recorded for all nine wombats, though for some specific analysis, only six wombats were used – the three most active wombats and the three least active wombats. This was done to isolate the data from only those wombats whose activity levels were representative of the two extremes, so that data from active wombats could be clearly defined and separated from sedentary wombats. Graphical representation of the data has been clearly labelled to identify when reporting figures on all wombats, or on the top and bottom 30% of active wombats

For the purposes of analysing amount of activity, the time devoted to eating was excluded from this analysis. Behaviours were divided into active behaviours (walking, digging) and sedentary behaviours (sitting, lying, standing). Sleeping, scratching, vocalising, sniffing and escape attempts were excluded from statistical analyses as the combined total of these behaviours was less than 3% of the total time devoted to physical activity.

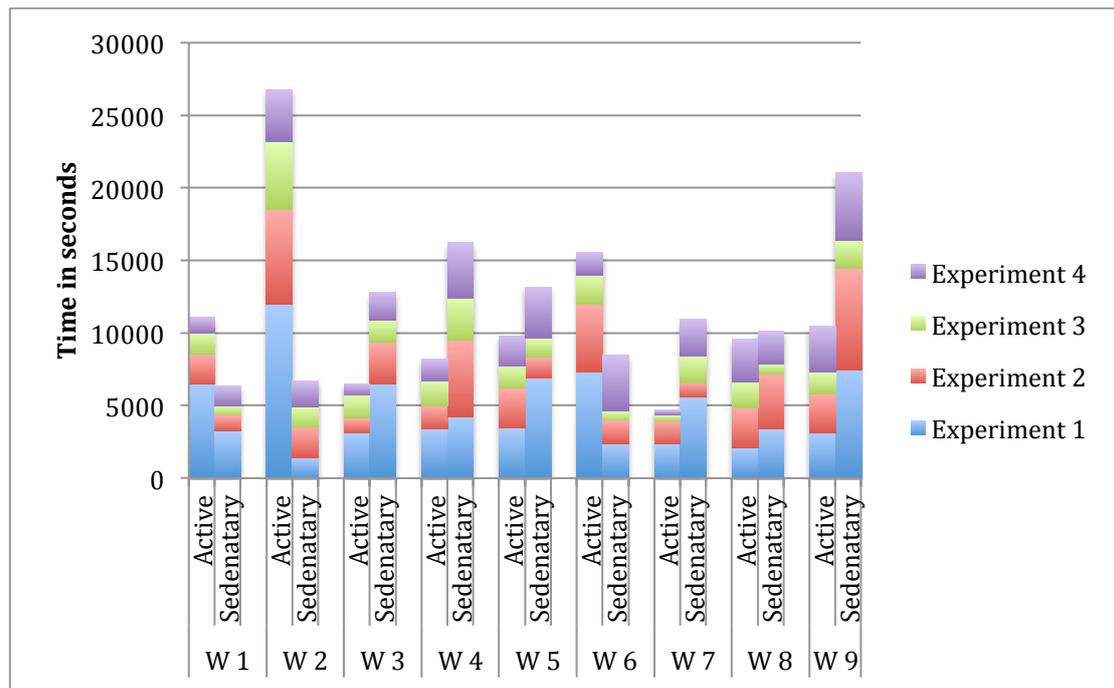
### Results

The results indicated there were marked differences in amounts of activity among all wombats. The average time devoted to performing all behaviours was recorded for all nine wombats as a percentage of all available time (Figure 4.2). Time spent eating (pellets and test food) equated to 55.9% of the total time available. The other observed behaviours recorded the following results: walking 18.6%, Sitting 5.9%, lying 6.5%, sleeping 0.05%, Digging 2%, Scratching 1.3%, Vocalising 0%, sniffing 0.6%, standing 8.1% and escape attempt 1.1% (Figure 4.2).



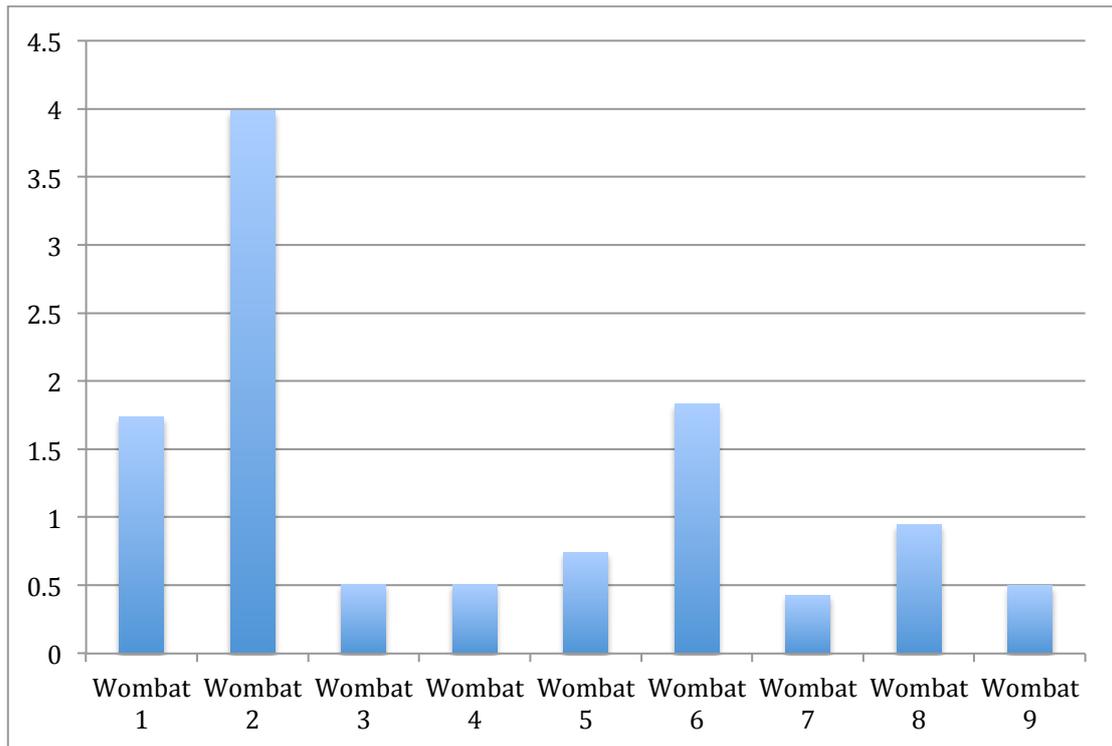
**Figure 4.2: Percentage of time spent performing each observed behaviour**

The time devoted to performing active behaviours compared with sedentary behaviours for each wombat during the four food choice experiments is depicted in Figure 4.3. Wombats 1, 2 and 6 were consistently more active than those that were more sedentary during all four experiments while the remaining wombats were consistently more sedentary than active. Wombats 3, 4 and 7 were the most sedentary during all four experiments (Figure 4.3).



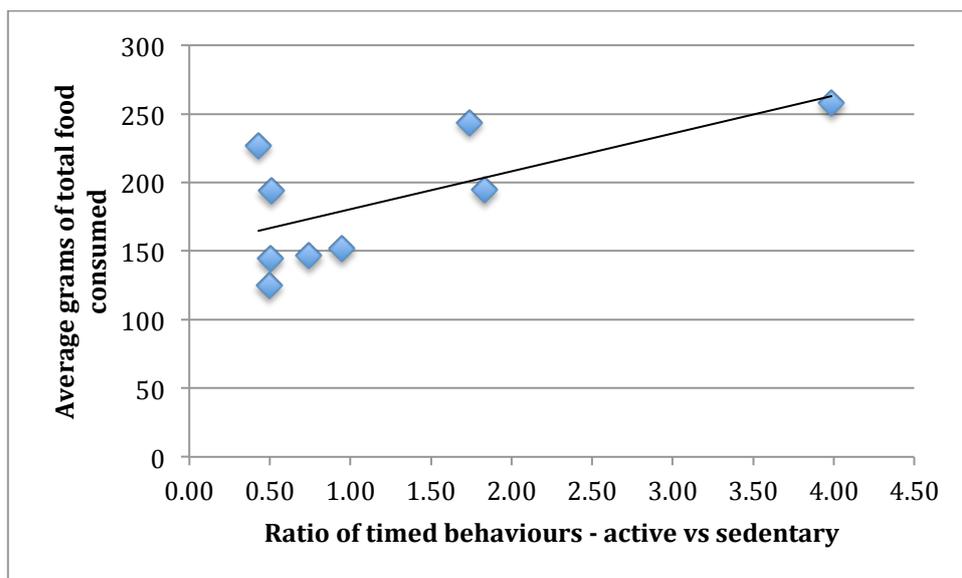
**Figure 4.3: Time spent performing active vs sedentary behaviours for all wombats during all experiments.**

The ratio of active behaviours compared to sedentary behaviours was calculated by dividing the time spent performing active behaviours by the total time spent performing sedentary behaviours (Table 4.4). A figure less than one indicates the wombat was more sedentary than active. A figure greater than one indicated the wombat was more active than sedentary. Wombats 1, 2 and 6 were consistently more active than sedentary. Wombat 2 was four times more active than sedentary (Figure 4.4).



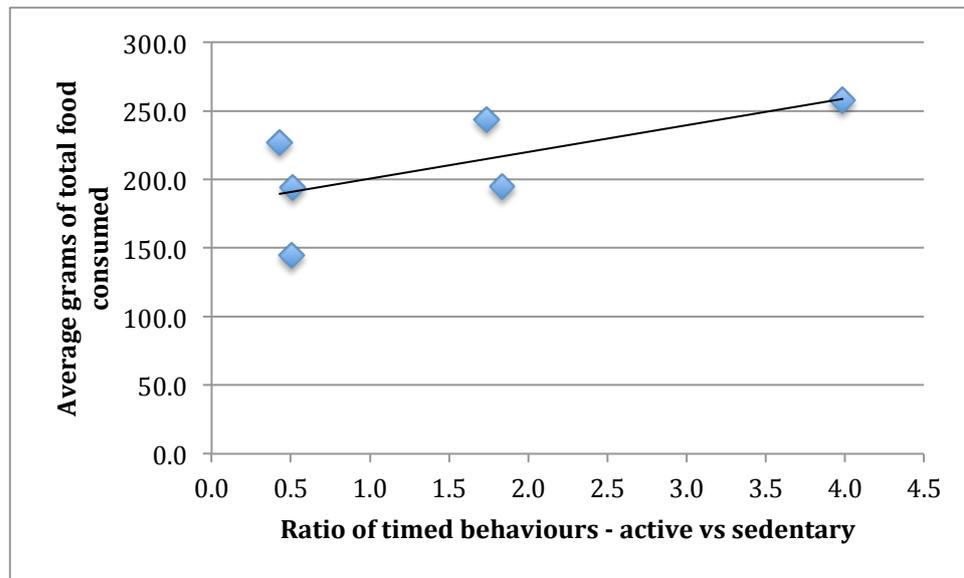
**Figure 4.4: Ratio of active compared to sedentary behaviours per wombat (values greater than one indicate the wombat was more active than sedentary)**

When the ratio of active vs. sedentary behaviours was calculated, the results were then plotted against the average of combined total foods consumed (i.e. the average of pellets and test food combined) to create a correlation between the amount of food consumed and the activity ratio (Figure 4.5). Increased activity positively correlated with increased food consumption (Figure 4.5).



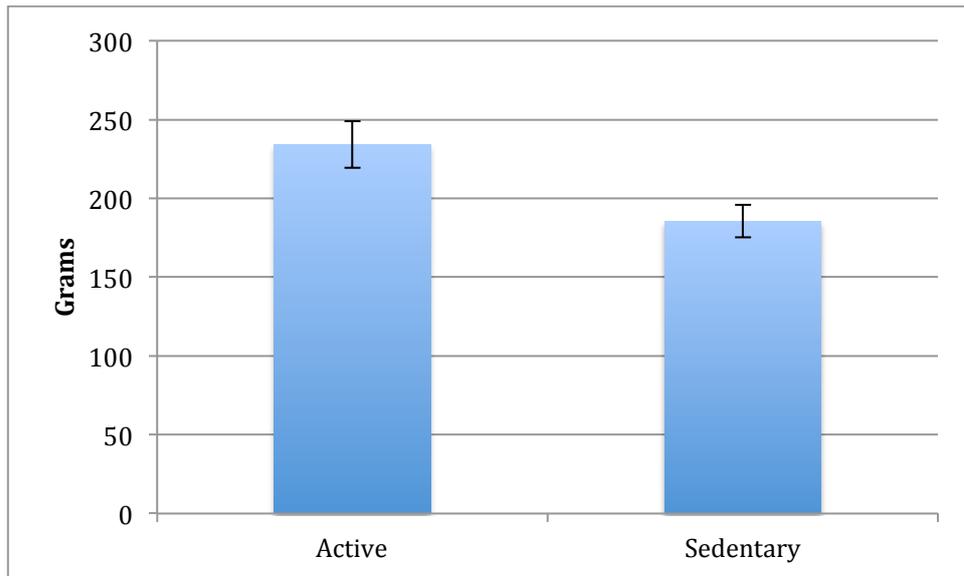
**Figure 4.5: Correlation between average combined total foods consumed and ratio of activity ( $r=0.67$ ).**

This result held true when calculated using the data for all nine wombats, and when using the data for just the 30% most active and 30% most sedentary wombats (Figure 4.6).



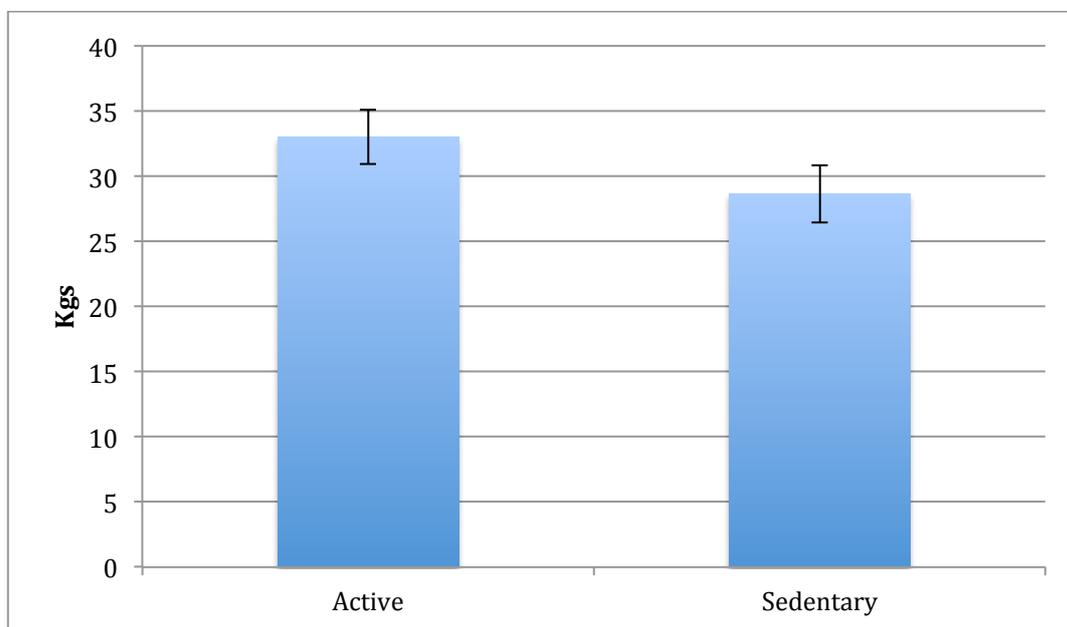
**Figure 4.6: Correlation between average combined total foods consumed and ratio of activity for 30% most active wombats and 30% most sedentary wombats ( $r=0.65$ ).**

The 30% most active wombats consumed significantly more food ( $234 \pm 14.8$  g) than the 30% most sedentary wombats ( $185.6 \pm 10.4$  g) (Figure 4.7). An Independent samples t-test between subjects revealed  $t=2.67$ ,  $df=189$  and  $p = 0.0082$ .



**Figure 4.7: Average amount of food consumed by the 30% most active and 30% most sedentary wombats**

While the active wombats were heavier in body weight ( $33 \pm 1.9$  kg) than the sedentary wombats ( $28.7 \pm 2.1$  kg), this difference was not significant ( $t=1.5$ ,  $df=4$ ,  $p = 0.2$ ) (Figure 4.8).



**Figure 4.8: Mean body weights of active and sedentary wombats**

## Discussion

The results demonstrated that physically active wombats consumed more food and were larger in body weight than sedentary wombats. The results indicate that wombats can regulate food intake based on energy expenditure. Physically active wombats would require a greater amount of food intake than sedentary wombats to satisfy their biological energy needs. Given sedentary behaviour is recognised as a behavioural strategy that wombats use to conserve energy it is likely that there could be links between energy requirements and levels of activity. Within the group of wombats used in this experiment, three were consistently more active than sedentary, while the remaining six wombats were consistently more sedentary than active. Of the three active wombats, two were male and one was a female. The sedentary wombats consisted of one male and five females. As these wombats have been fed the same diet and maintained in the same group housing arrangements for the past 5 years, it is assumed that they were all in a similar nutritional state prior to the commencement of the study, thus the differences in food choices and physical activity are likely to be a result of factors other than nutrition. These differences could relate to morphological, psychological or other differences such as genetic and epigenetic variations among wombats as well as social hierarchy. It is these individual differences that are of most interest for the captive management of this species. Zoos are becoming increasingly focused on maintaining the captive populations rather than supplementing with animals captured in their natural habitat and transferred to zoos. As zoo management gain a greater understanding of variations of dietary needs and preferences among animals, welfare outcomes will improve for all animal species housed in zoos and the longevity of animals such as wombats in captivity may be improved.

The SHNWs are different from other grazing herbivores in that they are social animals when sleeping and sharing dens, but tend to be solitary grazers (Descovich, Lisle et al. 2012). The zoo management must consider these behavioural differences from other species when designing enclosures and selecting animals to be housed together.

This present study provided evidence that there are differences among wombats in amounts of physical activity during the periods when they are feeding. Some animals were active throughout the 30 minute feeding period, inspecting the food bowls

multiple times, while others simply sat at the food bowl for extended periods. If two animals with different amounts of physical activity were to be housed together and fed together, there is a possibility that the more active wombat may not get sufficient access to the food bowl to allow adequate nutritional intake. Zoo staff may need to provide multiple food stations to accommodate a more desirable group housing configuration for wombats as compared with what exists at present.

The present study also provided evidence that more physically active wombats consumed more food on average than sedentary wombats. It is interesting that the more physically active wombats consumed more food and had greater body weights than the sedentary wombats. Perhaps in the group of wombats used in the present study, the larger animals happened to be more active while the smaller animals happened to be more sedentary. Another theory could be that the sedentary wombats were able to restrict their food intake so as not to gain weight. This demonstrates a form of nutritional “wisdom” – adjusting food intake to meet activity budgets. Likewise the more physically active wombats may have had biological sensing systems that resulted in greater food consumption to compensate for the greater energy expenditure that occurred due to greater amounts of physical activity of these animals.

The statistically significant differences in physical activity within this group of wombats could also be explained by innate genetic and epigenetic differences of the wombats. It was beyond the scope of this study to explore personality traits in more detail. This is a field of study, however, that is gaining emphasis within the animal behaviour sciences (Wemelsfelder, Hunter et al. 2001).

As the four food preference experiments were conducted over the course of one full calendar year, it can be assumed that any differences in amount of food consumption caused by seasonal climate variations were equalised over time.

In summary, this research found significant individual differences between wombats, which must be taken into account when designing feeding regimes for captive SHNW. There is more work to be done to explore the reasons for such individuality and it is hoped that this research has contributed to the knowledge of this species such that it may be applied to improve the welfare in captive environments.

### **Acknowledgements**

The authors acknowledge the support of Rockhampton Regional Council and the Rockhampton Zoo for providing the facility and the animals for this study. Thanks also to Judy Couper and her assistant, Mackenzie Hansler, for their assistance with the dry matter data collection. The following people assisted with data collection at various times and for this the authors offer thanks and gratitude; Lauren Thompson, Heather Campitell, Alexis DuPont, Tiffany Palmer and Andrew Fenning.

## References

- Barboza, P. S. (1989). The Nutritional Physiology of the Vombatidae. Doctor of Philosophy PhD Thesis, The University of New England.
- Barboza, P. S. and I. D. Hume (1992). "Hindgut fermentation in the wombats: two marsupial grazers." Journal of Comparative Physiology B **162**: 561-566.
- Descovich, K., A. Lisle, et al. (2012). "Differential responses of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*) to the presence of faeces from different species and male and female conspecifics." Applied Animal Behaviour Science **138**: 110-117.
- Descovich, K., A. Lisle, et al. (2012). The effect of group size on vigilance and activity budgets in a captive solitary marsupial (*Lasiorhinus latifrons*). Brisbane, The University of Queensland: 73-90.
- Descovich, K., A. Lisle, et al. (2012). Space allowance and the behaviour of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*), The University of Queensland.
- Fenning, Y. and D. Swain (2017). Food preferences of captive southern hairy-nosed wombats and the implications for captive management of this species. Unpublished thesis. Rockhampton, CQUniversity.
- Finlayson, G. R., G. A. Shimmin, et al. (2003). "Monitoring the activity of a southern hairy-nosed wombat, *Lasiorhinus latifrons*, using temperature dataloggers." Australian Mammalogy **25**: 205-208.
- Finlayson, G. R., G. A. Shimmin, et al. (2005). "Burrow use and ranging behaviour of the southern hairy-nosed wombat (*Lasiorhinus latifrons*) in the Murraylands, South Australia." Journal of Zoology **265**: 189-200.
- Hogan, L., C. Phillips, et al. (2009). "Remote monitoring of the behaviour and activity of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*)."  
Australian Mammalogy **31**: 123-135.
- Hume, I. D. (1999). Marsupial Nutrition. London, Cambridge University Press.
- ISIS (2016). ZIMS: Zoological Information Management System.
- Nicolson, V. (2012). Southern hairy-nosed wombat population management plan. Annual report and recommendations. Gold Coast, Dreamworld: 23.
- Triggs, B. (2009). Wombats: second edition. Collingwood, CSIRO Publishing.

Wemelsfelder, F., T. E. A. Hunter, et al. (2001). "Assessing the "whole" animal: a free choice profiling approach." Animal Behaviour **62**: 209-220.

## Chapter 5

### Food intake relative to time of day in captive southern hairy-nosed wombats

#### Abstract

This research examined the effects of time of day during individual feeding trials of nine captive southern hairy-nosed wombats housed at Rockhampton Zoo.

The experimental design, described in chapters 3 and 4, allowed for each of the nine wombats to be observed for 32 individual 30 minute feeding periods; a total of 288 observation periods. The feeding periods were conducted between 1500 and 2100 hours over the total time period of 15 months (September 2013-December 2014). To account for changes in feeding patterns/choices among wombats feeding at the 1500 compared to wombats feeding later in the evening, the individual wombats were randomly selected each day for order that they would be fed during the daily feeding period. This allowed for each wombat to be observed over multiple time periods. The results showed that as the hour became later, the wombats consumed more of the pelletised diet and less of the test food items. That is, they increased consumption of pellets as the evening became later, while they consumed the same or less of the test food items (carrot, kangaroo grass, guinea grass and sweet potato) than earlier in the evening.

#### Keywords

Feeding time, time of day, food intake

### Introduction

This research examined the effect of time of day on the feeding behaviours of a group of nine captive SHNWs. A survey of feeding regimes in zoos that house this species showed inconsistency regarding the feeding schedules of these animals. No study has examined the effects of feeding time on captive SHNWs. While it was beyond the scope of this project to examine the effects of feeding wombats in the morning as apposed to the evening, this series of feeding experiments explored the effects of delayed feeding time compared to the usual feeding time of this particular group of wombats – 1500 (3pm).

The aims for these experimental observations were as follows –

12. To determine if total food intake was affected by time of day
13. To determine if time of day affected food choice

The observations for this study were carried out in combination with observations conducted for the previous two research projects discussed in Chapters 3 and 4.

### Methods

The experimental methods used for this project have been described in detail in Chapters 3 and 4. The data for this set of experimental aims was collected during the previously described feeding trials of chapters 3 and 4, and as such will not be described again here.

Activity routines of this group of wombats, reported average den emergence times and time devoted to foraging/feeding within a 24 hr period have been previously described (Hogan et al., 2009). Data from Hogan's (2009) research helped inform the design of this feeding experiment with den emergence times being used to establish a time of day to initiate the feeding periods. The wombats in the previous study (Hogan et al., 2009) were active for 2 to 4 hour periods, which gave the approximate start and finish times for the series of experiments reported in this thesis. The seasonal data (i.e., earlier den emergence in the winter months compared with summer) from the previous study (Hogan et al., 2009) were also used to determine the approximate start

and finish times for the feeding periods during the experiments reported in this thesis. During the winter months, the experimental feeding periods commenced earlier in the afternoon (around the 1500 hour) while in the warmer months of the year, the feeding periods for the experiments commenced later (around the 1700 hour). Feeding periods were concluded within the 2000 hour.

### *Data collection*

Throughout each 30 minute feeding period the wombats were observed from an elevated position and behaviours recorded on a Microsoft Excel spread sheet in a continuous fashion. At the end of the 30 minute feeding period, the data collected provided numeric documentation of the amount of time that had elapsed while the wombat was eating. The food that was not eaten was weighed, disposed of and the data for weights of this food were then entered into the spread sheet along with numeric values for temperature and humidity at the time of the period. The majority of the data collection experiments were conducted by the primary study investigator, with several other trained individuals used during different experiments to observe the wombats. These individuals were continually supervised by the primary study investigator at all times and there were no differences found in the recording/observation of the data by the various observers. This was further controlled for by randomisation across groups and wombats.

### *Statistical analysis*

Graphical representations of the data collected were created using Microsoft Excel. All data was presented as mean  $\pm$  SEM with analysis of variance and tests evaluating preferences for naturalness compared with familiarity conducted using the Graphpad Prism statistical program. T-tests for significance were used at  $P \leq 0.05$ .

### *Results*

The data demonstrates that as the day became later, the wombats chose to eat more of the pellets (Figure 5.1) but consumption of the test food items was not affected by time of day (Figure 5.2).

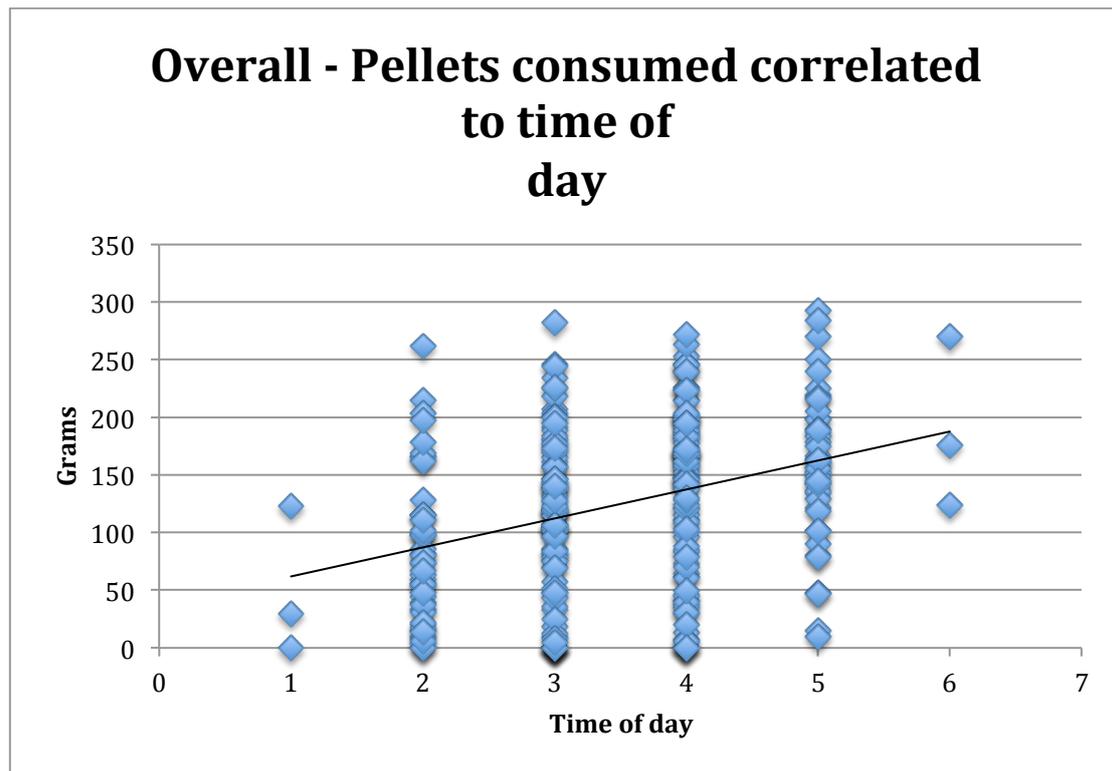


Figure 5.1: Average pellets consumed correlated to time of day ( $n=288$ ,  $r=0.35$ )

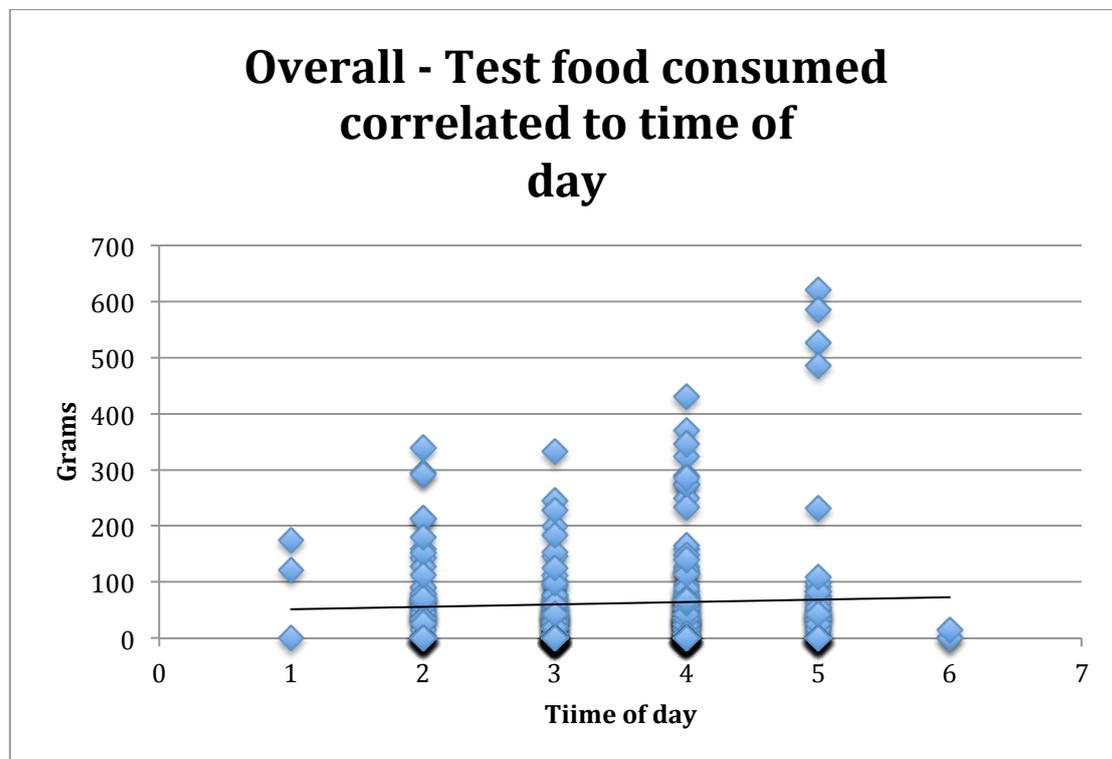
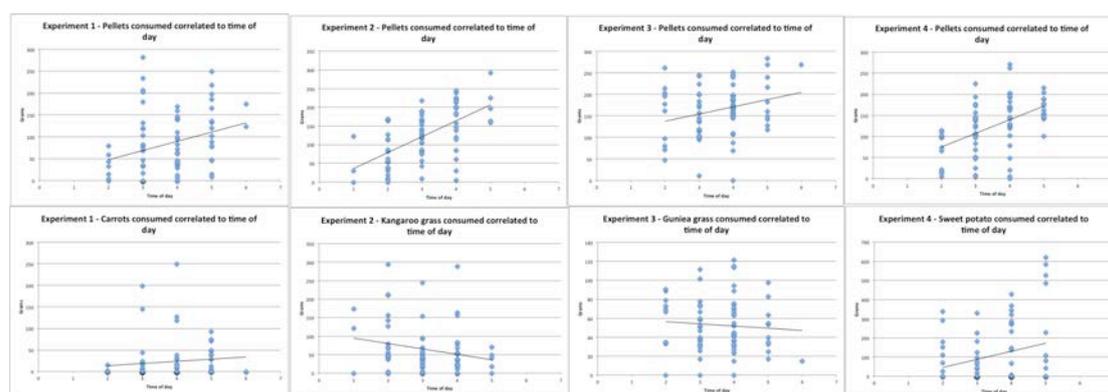


Figure 5.2: Average test foods consumed correlated to time of day ( $n= 288$ ,  $r=0.04$ )

Examination of each experiment in isolation reveals this overall trend to be true for all four experiments regarding pellet consumption, but is less accurate for the test foods (Figure 5.3). In all four experiments, pellet consumption increased with time of day. Experiments 1 and 4 showed a slight increase in test food consumption correlated to time of day, while Experiments 2 and 3 showed a slight drop in test food consumption correlated to time of day (Figure 5.3).



**Figure 5.3: Pellets and test food consumption correlated to time of day per experiment**

**Table 5.1: Correlation coefficient values for graphs in figure 5.3**

		Experiment 1	Experiment 2	Experiment 3	Experiment 4
<b>r value</b>	Pellets	0.30	0.60	0.27	0.48
	Test food	0.11	-0.22	-0.08	0.25
<i>n</i> =72					

A more detailed assessment of these results revealed differences among some of the wombats and between food choices (Figures 5.4 and 5.5). Only four wombats had a negative correlation of pellets consumed to time of day (Wombat 1 Experiment 1, Wombat 2 Experiment 1, Wombat 6 Experiment 1 and Wombat 7 Experiment 4). All the remaining wombats showed a positive correlation to amount of pellets consumed to time of day. In contrast, there were 16 negative correlations and seven neutral correlations of the test foods (Figure 5.5). The data trend lines are the important aspect of the graphs presented in figures 5.4 and 5.5. The wombats that had a difference in trends to the overall graphs

depicted in Figures 5.1 were Wombats 2, 5, 6 and 9 (Figures 5.4 and 5.5). The individual graphs for wombats 2, 5, 6 and 9 are presented larger for easier reading (Figures 5.6-5.13). Correlation coefficient values are given for the discussed wombats (Wombats 2, 5, 6 and 9) but have not been presented for every wombat in Figures 5.4 and 5.5 for ease of reading.

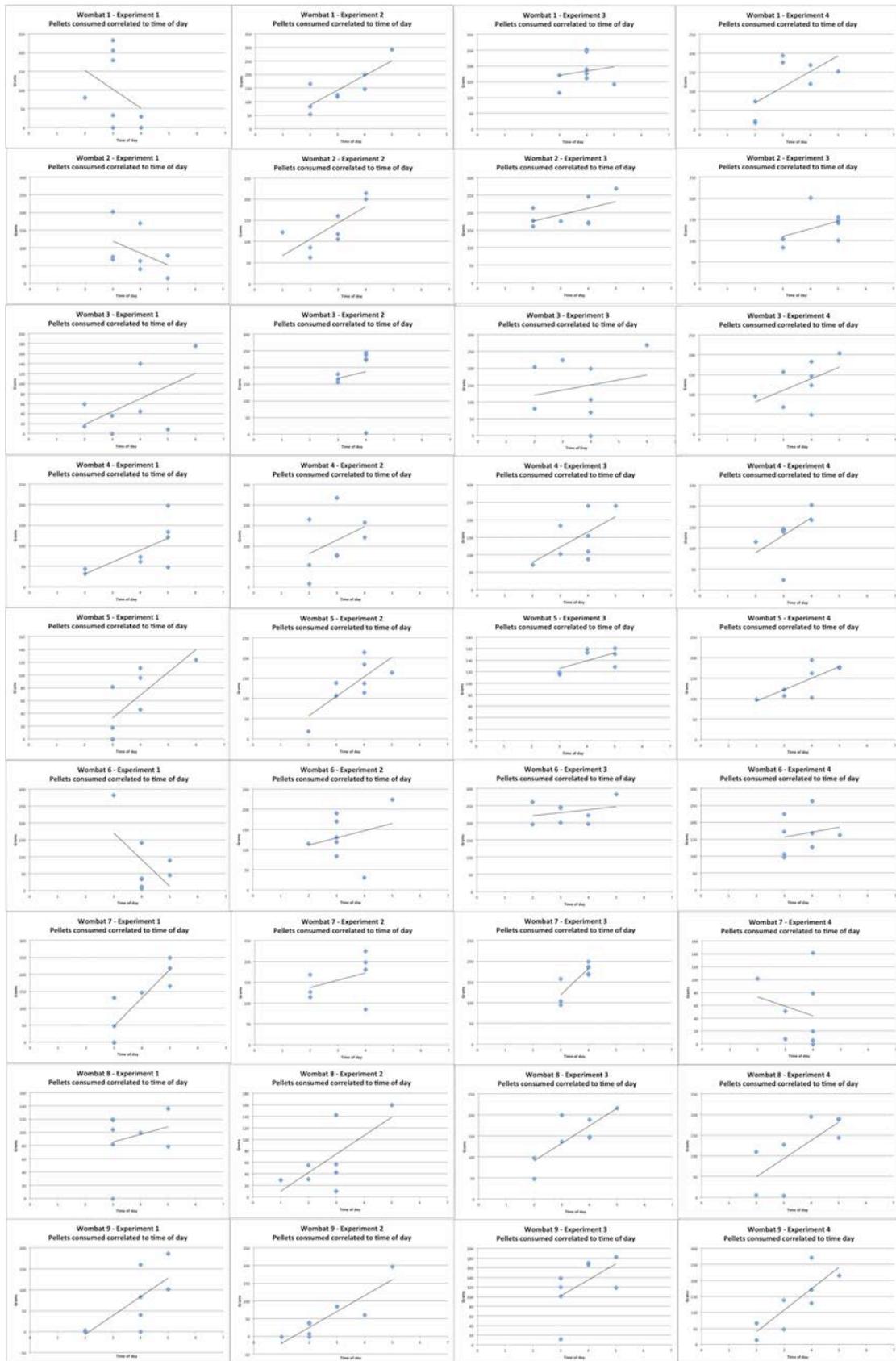


Figure 5.4: Average pellet intake correlated to time of day for each wombat for each experiment (n=8)



**Figure 5.5: Average test food intake correlated to time of day for each wombat for each experiment ( $n=8$ )**

During all four experiments, Wombat 2 showed a consistent consumption of pellets. The amount of pellets consumed did not vary greatly depending on the time of day (Figure 5.6). Consumption of the test food items by Wombat 2 did

not increase over time. The later the feeding period in the day, the more test food was consumed (Figure 5.7). This finding was influenced greatly by this particular wombat's strong preference for sweet potato in Experiment 4, as described previously in Chapter 4 of this thesis.

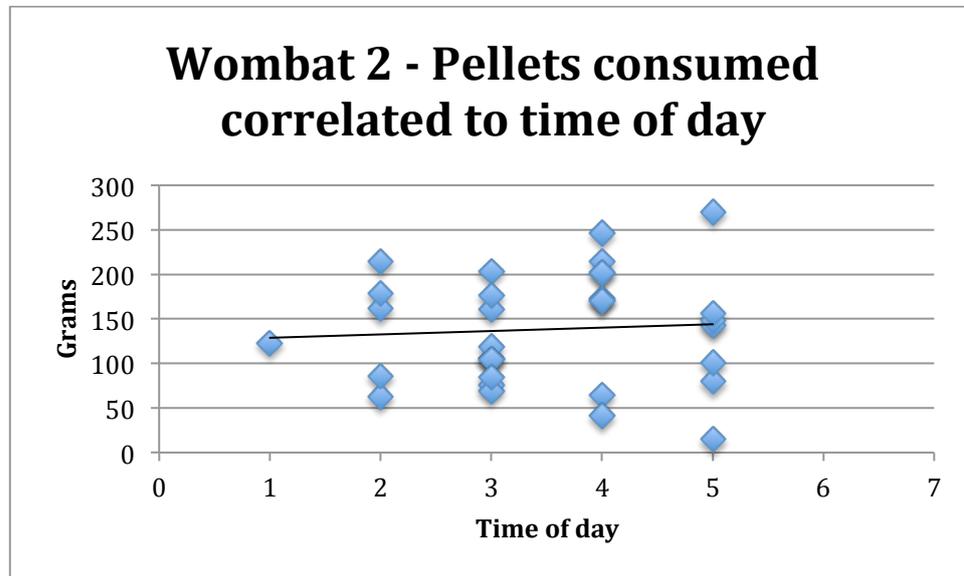
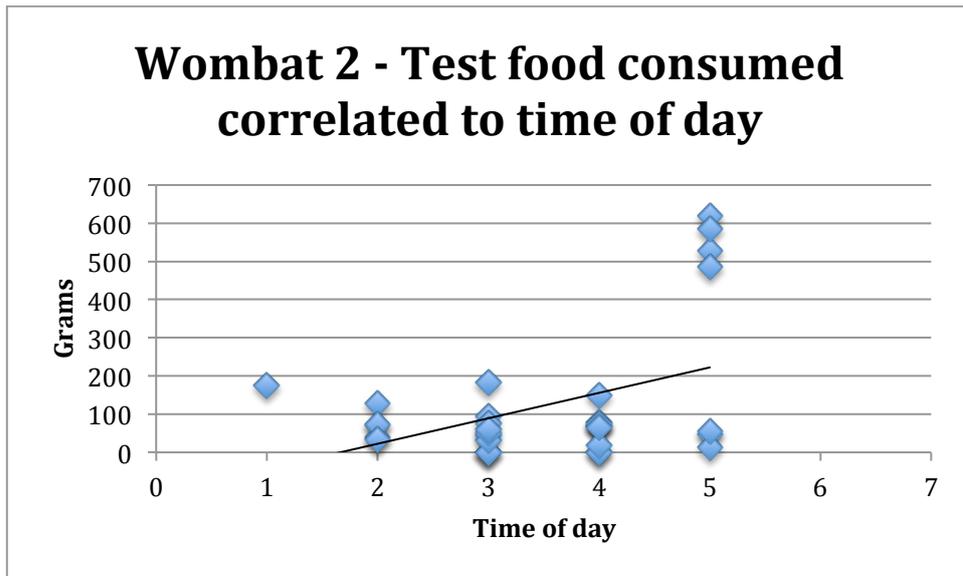


Figure 5.6: Wombat 2, total amount of pellets consumed correlated with time of day ( $n=32$ ,  $r=0.07$ )



**Figure 5.7: Wombat 2, Total amount of test foods consumed correlated with time of day ( $n=32$ ,  $r=0.43$ )**

Wombat 5 differed from the overall trend of wombat food consumption by increasing consumption of both pellets and test foods as the duration of the experiments advanced (Figures 5.8 and 5.9). Wombat 5 did not consume any carrots during Experiment 1 or any sweet potato in Experiment 4. The results for this individual, therefore, only provide data for the consumption of pellets and the two grasses.

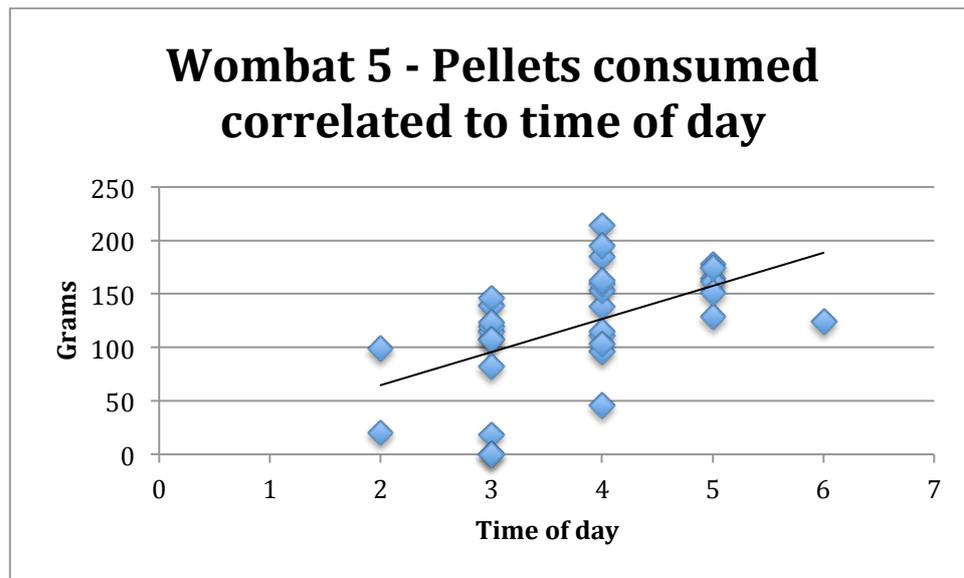


Figure 5.8: Wombat 5, Total amount of pellets consumed correlated with time of day ( $n=32$ ,  $r=0.55$ )

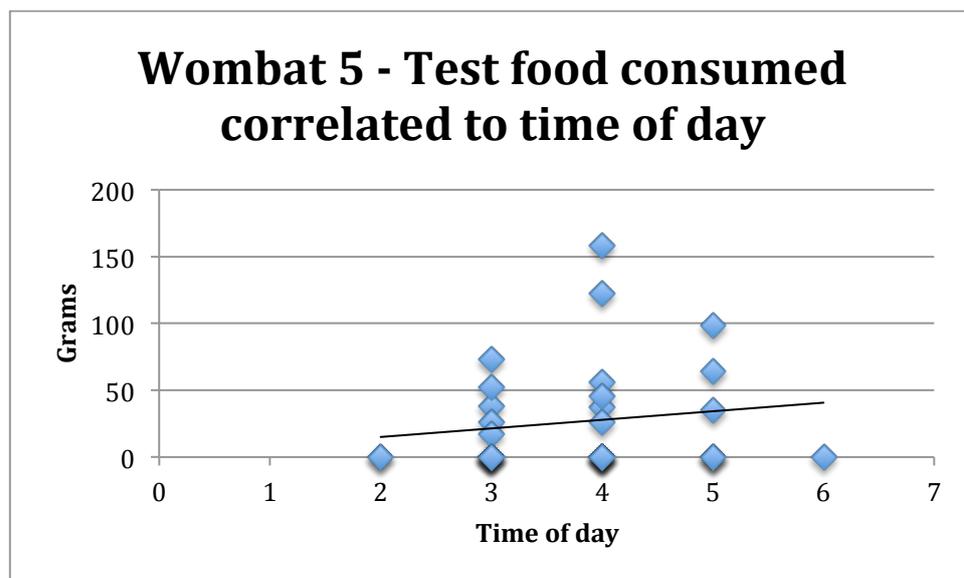


Figure 5.9: Wombat 5, Total amount of test foods consumed correlated with time of day ( $n=32$ ,  $r=0.14$ )

Wombat 6 showed an opposite trend of food consumption compared with the trend when all animals were included, by decreasing food consumption over time of both pellets and test foods (Figures 5.10 and 5.11). These results may have been influenced by this particular wombat's aversion to two of the test foods, carrot and sweet potato. Wombat 6 consumed only very small amounts of

these test items, preferring pellets and kangaroo grass compared with all other foods offered.

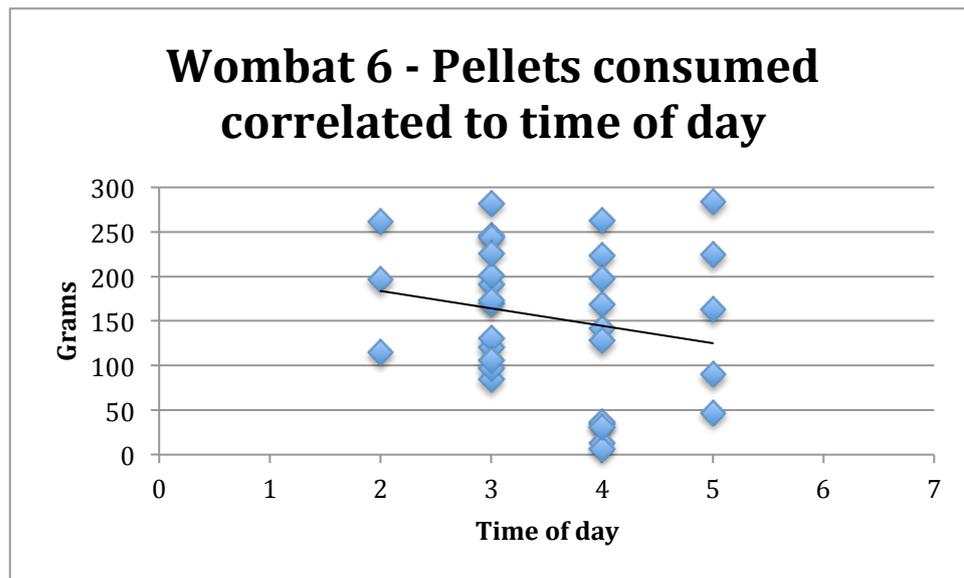


Figure 5.10: Wombat 6, Total amount of pellets consumed correlated with time of day ( $n=32$ ,  $r=-0.21$ )

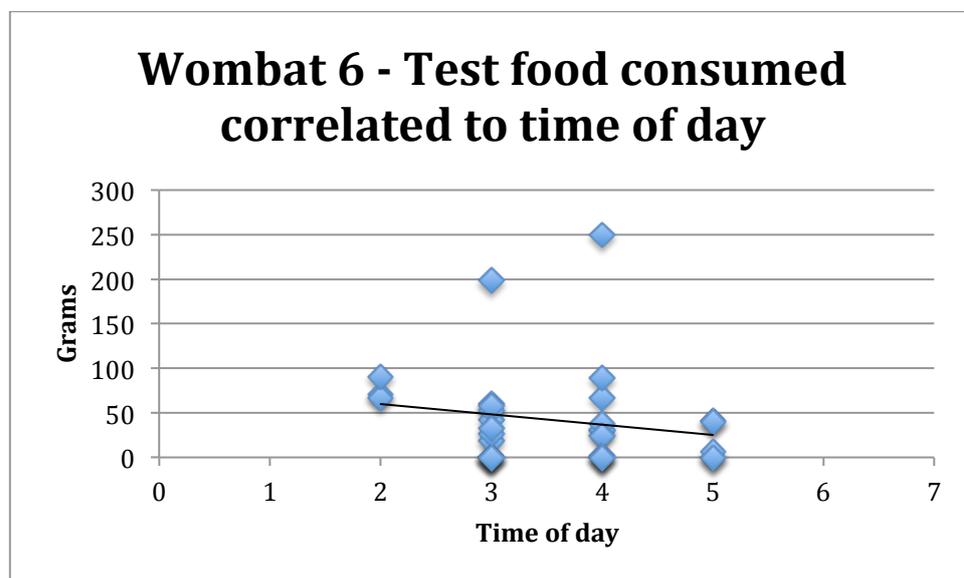


Figure 5.11: Wombat 6, Total amount of test foods consumed correlated with time of day ( $n=32$ ,  $r=-0.18$ )

Wombat 9 increased consumption of both pellets and test foods as the duration of the experiments advanced (Figures 5.12 and 5.13). This result is interesting because Wombat 9 did not consume any sweet potato in the final experiment and

had a decreased consumption of kangaroo grass as the duration of the experiments progressed. The data obtained from Wombat 9, therefore, indicate the great preference of this individual for pellets, carrots and kangaroo grass; that is, all the familiar food items.

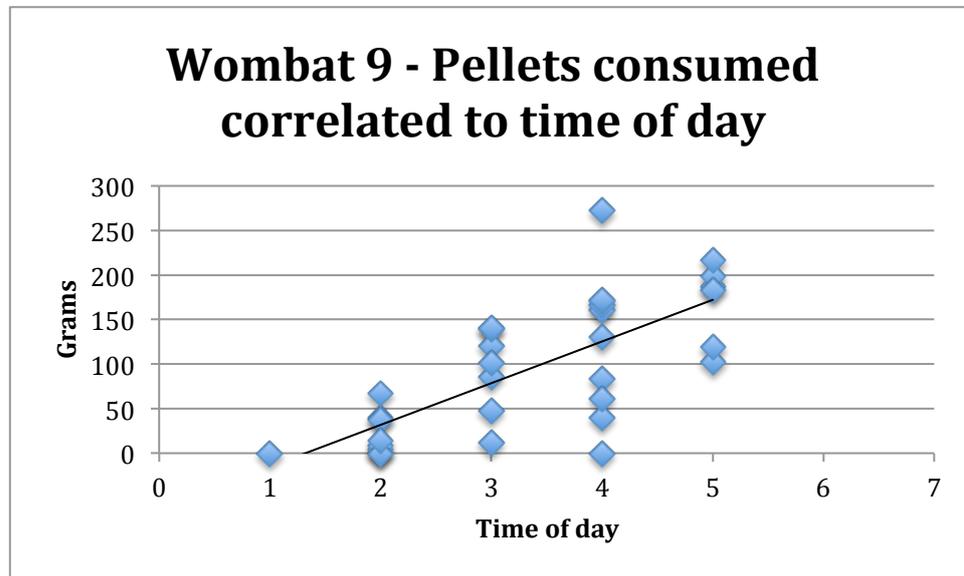


Figure 5.12: Wombat 9, Total amount of pellets consumed correlated with time of day ( $n=32$ ,  $r=0.71$ )

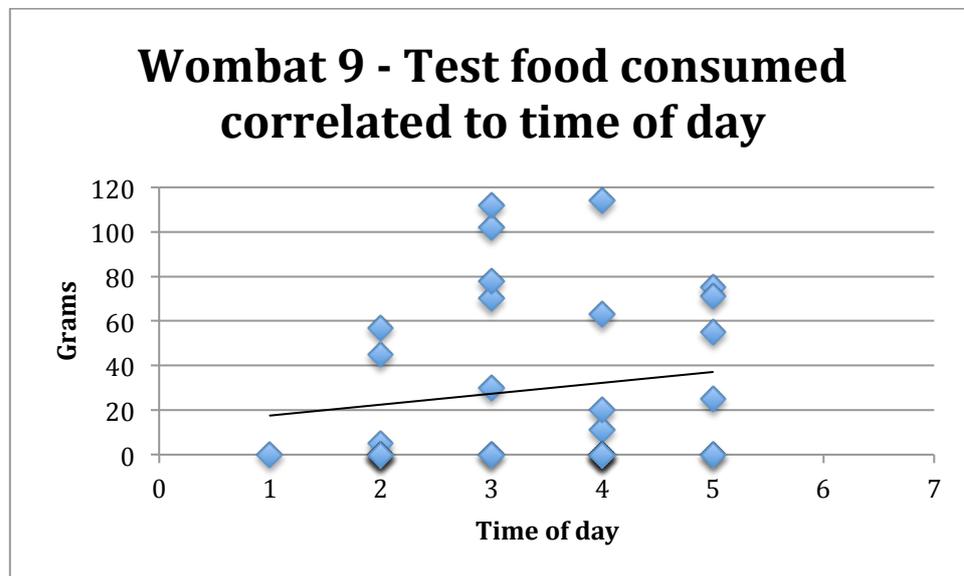


Figure 5.13: Wombat 9, Total amount of test foods consumed correlated with time of day ( $n=32$ ,  $r=0.15$ )

#### Discussion

These results indicate that as the evening progressed, the wombats became hungrier, but less curious. They consumed more of the control food, pellets, the later the feeding period occurred during the day, while their rate of consumption for the test food items remained unchanged. As the wombats became hungrier they were more likely to choose a familiar food source with known nutritional and digestive consequences (pellets), than an unfamiliar food that is comparatively less nutritionally balanced or of unknown digestive consequence.

Over the course of four experiments, wombats did adjust their food choices. While there were some differences between individual wombats, the most prevalent choice of food was the nutritionally balanced and familiar food source, pellets. The deliberate choice to leave pellets as the control food item in each experiment was done to eliminate the chance of the wombats becoming nutritionally compromised and to clearly demonstrate that the wombat were making a clear choice to sample the other food item even in the presence of a known and reliable food source.

These findings could have implications for captive wombats. As discussed in Chapter 3, wombats offered a choice of food items are better able to express their individuality, which allows for improved welfare status of the individual. This experiment showed that hungry wombats were less likely to select difference food options. Therefore, if wombats are to be offered a varied diet, this would be best offered *ad libitum*, to prevent the wombats from experiencing a level of hunger that alters their food preferences. Wombats that have the freedom to feed on a choice of foods at a time of day natural for their species will consumed a variety of food types according to their own individual preferences. This would be a positive welfare outcome for these wombats.

## Chapter 6

### Summary conclusions

The overall aim of this thesis was to investigate the food preferences of captive SHNWs. These aims were defined at the beginning of each results chapter (Chapter 3, 4 and 5) and were as follows-

1. To determine how much of the test food item was consumed
2. To determine how many times the test food item was selected
3. To determine how many times the consumption of the test food item exceeded consumption of pellets.
4. To establish if the test food items influenced the consumption of pellets
5. To determine if food preference is affected by variables such as rate of consumption and environmental conditions
6. To assess differences in physical activity levels between wombats.
7. To establish if activity level alters feeding behaviour.
8. Does food choice make a wombat behave differently.
9. To determine if total food intake was affected by time of day
10. To determine if time of day affected food choice

Previous studies have reported on the food choices of wombats in their natural habitat (Evans, Macgregor et al. 2006; Triggs 2009), however, this was the first study of food preferences in this species in captivity. Captive wombats were provided an opportunity to explore food choices using a more structured experimental methodology. An effective experimental design protocol was established to provide a reliable method of recording the wombats' food choices. Factors that may have influenced the wombats' choices were investigated, including seasonal weather conditions at the time the experiments were

conducted and time of day, as well as individual differences in physical activity during the feeding experiments. It was anticipated that the wombats would have a preference for natural/familiar as compared with unnatural/unfamiliar foods. The findings reported in each of the preceding chapters will be discussed subsequently in this final section of this thesis.

At the time the experiments commenced for this project a survey of diets for captive wombats used in the zoos of Australia provided information indicating that the captive diet resembled little of the diet of wombats in their natural habitat. The experiment in Chapter 3 was designed to assess whether institutionalised captive wombats would prefer a natural diet (grass) as compared with the diets that are commonly provided by zoo management (pasture replacement pellets, carrots and sweet potato). These wombats consumed more pellets during the feeding periods when the experiments were conducted than any other food item. This was not a surprising result. Zoo records had indicated that this group of wombats reliably consumed the pellet diet daily for many years prior to the commencement of this research. For this reason the wombats were offered pellets as one of two choices of feeds across four different test food trials. If the wombats chose to eat one of the test foods over the pellets, then it was clear they were making a choice to try that test food despite the presence of their familiar and well accepted staple diet.

The wombats were influenced by the four different food choices. Grass, both familiar and unfamiliar, positively correlated with the total amount of food consumed and was also more readily chosen by all nine wombats. Carrot was negatively correlated with total food consumed and sweet potato caused stark differences in personal preference among the wombats. The naturalness of the items influenced consumption more than familiarity. The wombats did not avoid the unfamiliar foods. These results are inconsistent with reports from previous studies on other captive herbivores, such as sheep that avoided novel food items in several studies ((Villalba, Provenza et al. 2007; Villalba, Manteca et al. 2009; Villalba, Provenza et al. 2011)) This would be an interesting topic to study further: Are domesticated animals more neophobic than non-domesticated animals when offered novel food items? Neophobia was not evident in this

group of SHNWs as consumption of the unfamiliar food items remained consistent over the course of eight feeding trials. That is, the wombats did not appear to increase or decrease consumption of the unfamiliar foods as they became more familiar over the eight feeding trials.

The inclusion of pellets during each feeding period ensured the wombats were able to maintain a nutritionally balanced diet to maintain their physiological status. Had the pellets been omitted from the feeding, the wombats would have been nutritionally compromised and may have made different food choices. Further experiments would have to be conducted to explore food choices for wombats that were nutritionally compromised.

The experimental design used for all four feeding experiments provided data that could be used to test food choice. Although the wombats had a nutritionally balanced food item available, the wombats still chose to consume the other foods provided during the experiments. The food selected by the wombats provides evidence that the wombats did make choices and these choices were statistically significant and individually unique. This finding has implications on how wombats can be fed in captivity to aid in improving their welfare state. Allowing for individual expressions of food preferences will greater meet the needs of the individual animal within the group thus allowing greater agency.

By studying the wombats individually, the effects of feeding competition among wombats were negated. This allowed for the individuality of each wombat to be examined. To explore the effects of group dynamics on food choices, further experiments would need to be designed and conducted. Such experiments were beyond the scope of this project.

Wombats used in the present study had unique individual preferences for foods and great variance in amounts of physical activity during the experiments. If animals have the choice of expressing their uniqueness within the feeding environment it has been theorized that this will benefit the population as a whole (Provenza, Villalba et al. 2003).

Chapter 4 reported the data on the findings for amount of physical activity during the feeding experiments. Physically active wombats consumed more food

than more sedentary wombats. This result is not surprising, as the energy expended by a physically active wombat would require a greater intake of energy from its food source than that required by a more sedentary wombat if body energy reserves are to be sustained. This finding has implications for the captive management of SHNWs into the future. The sedentary wombats in this study tended to sit for long periods at the food bowl consuming their chosen item. The more active wombats visited the food bowl multiple times within each feeding trail. If a zoo housed two wombats together, one being more sedentary in nature and the other being more active in nature, then there may need to be multiple feeding stations established to ensure each wombat has equal access to the required amount of food. If only one feeding station is provided, a sedentary wombat may sit at the food trough for extended periods which may inhibit access to the feeding station by the more active wombat.

Chapter 5 presented the findings of other data recorded during the four experiments that may have influenced the feeding behaviour of the wombats. Temperature and humidity had little to no impact on the feeding choices of these wombats. The time of day of feeding period did contribute to some variability in feeding patterns among wombats. The 5-hour time duration during which the feeding periods occurred each evening meant that the first wombat was fed during the 1500 hour, while the last wombat to be fed during the feeding period was fed between the 1900 and 2100 hour of the day. This difference in time of feeding during the 5-hour period over which feedings occurred may have resulted in the wombats that were fed later in the period to be hungrier at the time of the feeding period was initiated. These effects were minimised by the random order of selection of each wombat every day as to the sequence that they would be fed during the 5-hour feeding period. The results indicated that as the wombats became hungrier as a result of receiving food later in the 5 hour feeding period, the more they chose to eat pellets and the less they chose to eat the experimental food items. There were individual differences among wombats that were described in Chapter 5.

The small sample size ( $n=9$ ) is acknowledged as a limiting factor in this research. This limitation was not considered prohibitive to the study overall, as the overall

population of wombats in captivity is small (58 animals in total world wide). Therefore, this group of animals represented 15.5% (9/58 captive wombats) of the captive population of wombats in zoos. It was beyond the scope of this research masters project to design and implement an experiment that would incorporate all captive SHNWs without eliminating all the variables associated with wombats housed in many different facilities in different climactic conditions and on differing base diets. Rockhampton Zoos population of SHNW is the largest captive population of SHNW held in one place. Discounting the research on the grounds of the small *n* number would limit the knowledge of this species in general. It was therefore considered important to conduct this research in order to contribute to the knowledge of this species and to understand individual differences of these animals when housed in captivity. The repetitive measures experimental design was used to strengthen the statistical analysis of the results.

There were other limitations on this research project that could not be controlled for. These were as follows

- Innate differences in genetic, epigenetic, physiology, morphology, personality, age and reproductive status of the wombats
- The influence of humidity could not be fully accounted for due to limited data available for analysis

Future research could build upon the findings of this project by-

- Randomising the test foods across all four experiments
- Removing pellets from the feeding period so that the wombats only have options to choose the test food items

In conclusion, this thesis examined the food preferences of a group of SHNWs when offered the choice of familiar and unfamiliar, and natural and unnatural foods. Many factors that may have influenced the individual wombats food choices were discussed. The main conclusions drawn from this research are as follows:

1. Naturalness rather than familiarity was established as the more potent influence on food preference.
2. For this group of captive wombats, carrot should be reconsidered as a more infrequent feeding option given the negative impact on overall food consumption during the test period and fresh grasses should form an essential element of captive diets
3. When wombats were offered a control food of nutritionally balanced pellets, all wombats chose to eat some of the experimental food items. This indicates that the wombats were making clear choices regarding their food intake.
4. There were marked individual differences among wombats regarding their food preferences. Some wombats consumed very large quantities of a particular food item, while other wombats avoided that item completely. These differences were due to factors other than nutritional requirements.
5. As wombats became hungrier, they became less likely to consume the experimental food items, choosing to consume more of the pellets.
6. Temperature did not affect the food intake of these wombats.
7. The effect of humidity on the food intake of these wombats could not be properly analysed due to missing data caused by equipment malfunction. There appeared to be some relationship between humidity and food intake in this group of wombats, which would require further investigation.
8. Amount of physical activity of individual wombats influenced the amount of food consumed. Active wombats consumed more food than sedentary wombats.
9. The experimental design used throughout this study was effective in allowing for collection of data that when analysed yielded significant results that are repeatable in multiple experiments.

10. Over the course of four experiments, wombats did adjust their food choices. While there were some differences between individual wombats, the most prevalent choice of food was the nutritionally balanced and familiar food source, pellets.

These findings may have impacts of the welfare states of captive SHNWs. Welfare states are becoming more scrutinised by governing bodies such as the Australasian Zoo and Aquarium Association (ZAA). For member institutions to receive accreditation from ZAA they must demonstrate commitment to the five domains of animal welfare. It is hoped that this work has contributed to the broader knowledge of SHNWs in captivity. The individual differences expressed by the wombats during this series of experiments demonstrates a need for the industry to acknowledge such individuality in their animals and to provide increasing opportunities for captive animals to express their unique choices. Zoos can do this by examining their own wombats and collecting data as to the individual differences expressed by their animals. Diet choices provided should be not only nutritionally sound, but also varied to allow for individuality between wombats. By increasing opportunities for choice captive wombats may experience improved welfare states.

Suggested improvements to current basic husbandry practices are as follows:

- Offer a variety of dietary choices to the wombats, noting individual differences between wombats. Record these differences and learn from them.
- Offer multiple feeding stations to account for different activity levels.
- Offer opportunities for the wombats to graze on live growing grasses.
- If grazing is not available, allow the wombats access to food throughout the day/night, rather than at one feeding time only.

- Record daily food intake and weather condition data to better inform future generations of the effects weather conditions can have on food consumption in this species.
- Grass, due to its positive correlation with total food consumption, should form an essential component of the diet for all captive wombats.

## References

- Arnold, G. W., de Boer, E. S., Boundy, C. A. P (1980). "The influence of odour and taste on the food preferences and food intake of sheep." Australian Journal of Agricultural Research **31(3)**: 571-587.
- Atwood, S. B., F. Provenza, et al. (2001). "Influence of free-choice vs. mixed ration diets on food intake and performance of fattening calves." Journal of Animal Science **79**: 3034-3040.
- Barboza, P. S. (1989). The Nutritional Physiology of the Vombatidae. Doctor of Philosophy PhD Thesis, The University of New England.
- Barboza, P. S. (1993). "Digestive strategies of the wombats: feed intake, fiber digestion, and digesta passage in two grazing marsupials with hindgut fermentation." Physiological Zoology **66(6)**: 983-999.
- Barboza, P. S. (1993). "Effects of restricted water intake on digestion, urea recycling and renal function in wombats (Marsupialia: Vombatidae) from contrasting habitats." Australian Journal of Zoology **41**: 527-556.
- Barboza, P. S. and I. D. Hume (1992). "Digestive tract morphology and digestion in the wombats (Marsupialia: Vombatidae)." Journal of Comparative Physiology B **162**: 552-560.
- Barboza, P. S. and I. D. Hume (1992). "Hindgut fermentation in the wombats: two marsupial grazers." Journal of Comparative Physiology B **162**: 561-566.
- Barboza, P. S., I. D. Hume, et al. (1993). "Nitrogen metabolism and requirements of nitrogen and energy in the wombats (Marsupialia: Vombatidae)." Physiological Zoology **66(5)**: 807-828.
- Bechara, A., D. Tranel, et al. (1995). "Double dissociation of conditioning and declarative knowledge relative to the amygdale and hippocampus in humans." Science **269**: 1115-1118.
- Berteaux, D., M. Crete, et al. (1998). "Food choice by white-tailed deer in relation to protein and energy content of the diet: a field experiment." Oecologia **115**: 84-92.

- Carlstead, K. and D. Shepherdson (2000). Alleviating stress in zoo animals with environmental enrichment. The biology of animal stress: basic principles and implications for animal welfare. G. Moberg and J. Mench. Wallingford, CAB International: 337-354.
- Clauss M, Kohlschein G-M, et al. (2013). "Short-term digestible energy intake in captive moose (*Alces alces*) on different diets." Zoo Biology **32**: 484-489.
- De R, Moio L, et al. (2002). "Influence of flavor on goat feeding preferences." Journal of Chemical Ecology **28(2)**: 269-81.
- Delbridge, A., J. Bernard, et al., Eds. (1992). The Macquarie Dictionary. Sydney, The Macquarie Library.
- Descovich, K., A. Lisle, et al. (2012). "Differential responses of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*) to the presence of faeces from different species and male and female conspecifics." Applied Animal Behaviour Science: 110-117.
- Descovich, K., A. Lisle, et al. (2012). The effect of group size on vigilance and activity budgets in a captive solitary marsupial (*Lasiorhinus latifrons*). Brisbane, The University of Queensland: 73-90.
- Descovich, K., A. Lisle, et al. (2012). Space allowance and the behaviour of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*), The University of Queensland.
- Druery, G. V. (2004). Development of assisted breeding techniques in the southern hairy-nosed wombat, *Lasiorhinus latifrons*. Faculty of Arts, Health and Sciences. Rockhampton, Central Queensland University. **Master of Applied Science**: 256.
- Evans, M. C. (1999). Ecological energetics of wombats. Armidale, University of New England. **Doctor of Philosophy**.
- Evans, M. C., C. Macgregor, et al. (2006). "Diet and feeding selectivity of common wombats." Wildlife Research **2006**: 321-330.

- Everitt, J. H., Gonzalez, C.L., Scott, G., Dahl, B. E (1981). "Seasonal food preferences of cattle on native range in the south Texas plains." Journal of Range Management **34(5)**: 384-388.
- Favreau, F., P. J. Jarman, et al. (2009). "Vigilance in a solitary marsupial, the common wombat (*Vombatus ursinus*)." Australian Journal of Zoology **57**: 363-371.
- Felton, A. M., Felton, A., Raubenheimer, D., Simpson, S. J., Krizsan S. J., Hedwall, P.-O., et al. (2016). "The Nutritional Balancing Act of a Large Herbivore: An Experiment with Captive Moose (*Alces alces*)." PLoS ONE **11(3)**: e0150870. doi:10.1371/journal.pone.0150870
- Fernandez E, Dorey N et al. (2004) "A two-choice preference assessment with five cotton-top tamarins (*Saguinus Oedipus*)." Journal of Applied Animal Welfare Sciences **7(3)**: 163-9.
- Finlayson, G. R., G. A. Shimmin, et al. (2003). "Monitoring the activity of a southern hairy-nosed wombat, *Lasiorhinus latifrons*, using temperature dataloggers." Australian Mammalogy **25**: 205-208.
- Finlayson, G. R., G. A. Shimmin, et al. (2005). "Burrow use and ranging behaviour of the southern hairy-nosed wombat (*Lasiorhinus latifrons*) in the Murraylands, South Australia." Journal of Zoology **265**: 189-200.
- Forbes, J. M., and Kyriazakis, I. (1995). "Food preferences in farm animals: why don't they always choose wisely?" Proceedings of the Nutrition Society **54(2)**: 429-440.
- Higgins A, Bercovitch F, et al. (2011). "Dietary specialization and Eucalyptus species preferences in Queensland Koalas (*Phascolarctos cinereus*)." Zoo Biology **30**: 52-58.
- Hogan, L., C. Phillips, et al. (2009). "Remote monitoring of the behaviour and activity of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*)." Australian Mammalogy **31**: 123-135.
- Hogan, L. (2010). The behaviour and reproductive biology of captive southern hairy-nosed wombats (*Lasiorhinus latifrons*). School of Animal Studies. Brisbane, The University of Queensland. **Doctor of Philosophy**: 271.

- Hogan, L., S. Johnston, et al. (2010). "Stereotypes and environmental enrichment in captive southern hairy-nosed wombats, *Lasiorhinus latifrons*." Applied Animal Behaviour Science **126**: 85-95.
- Hogan, L., C. Phillips, et al. (2010). "Monitoring male southern hairy-nosed wombat (*Lasiorhinus latifrons*) reproductive function and seasonality in a captive population." Animal Reproduction Science **118**: 377-387.
- Hogan, L., C. Phillips, et al. (2010). "Non-invasive methods of oestrus detection in captive southern hairy-nosed wombats (*Lasiorhinus latifrons*)." Animal Reproduction Science **119**: 293-304.
- Hogan, L., Phillips, C., Horsup, A., Janssen, T., Johnston, S. (2011). "Technique for faecal marking in group-housed southern hairy-nosed wombats *Lasiorhinus latifrons*." Australian Zoologist **35 (3)**: 649-654
- Horsup, A. (2004). Recovery plan for the northern hairy-nosed wombat *Lasiorhinus keffrii* 2004 - 2008. Brisbane, The State of Queensland, Environmental Protection Agency: 36.
- Horsup, A. (2012). "Fact Sheet: Wombats of Australia." Retrieved 18.05.2012, 2012.
- Hume, I. D. (1989). "Nutrition of marsupial herbivores." Proceedings of the Nutrition Society **48**: 69-79.
- Hume, I. D. (1999). Marsupial Nutrition. London, Cambridge University Press.
- Hutson, G. D. and S. C. van Mourik (1981). "Food preferences of sheep." Australian Journal of Experimental Agriculture and Animal Husbandry **21(113)**: 575-582.
- ISIS (2016). ZIMS: Zoological Information Management System.
- IUCN. (2017). "The IUCN Red List of Threatened Species." Retrieved 13/03/2017, 2017, from <http://www.iucnredlist.org/details/11343/0>.
- Konarzewski, M. and J. Diamond (1995). "Evolution of basal metabolic rate and organ masses in laboratory mice." Evolution **49(6)**: 1239-1248.

- Kyriazakis, I. and J. D. Oldham (1993). "Diet selection in sheep: the ability of growing lambs to select a diet that meets their crude protein (nitrogen x 6.25) requirements." British Journal of Nutrition **69**: 617-629.
- Kyriazakis, I. and J. D. Oldham (1997). "Food intake and diet selection in sheep: the effects of manipulating the rates of digestion of carbohydrates and protein of the foods offered as a choice." British Journal of Nutrition **77**: 243-254.
- Langvatn, R. and T. A. Hanley (1993). "Feeding-patch choice by red deer in relation to foraging efficiency: an experiment." Oecologia **95**: 164-170.
- Lian, X., T. Zhang, et al. (2010). "Group size effects on foraging and vigilance in migratory Tibetan antelope." Behavioural Processes **76**: 192-197.
- Manteca, X., J. J. Villalba, et al. (2008). "Is dietary choice important to animal welfare?" Journal of Veterinary Behaviour **3**: 229-239.
- Mellor, D., Beausoleil, N. (2015). "Extending the 'Five Domains' model for animal welfare assessment to incorporate positive welfare states." Animal Welfare **24**: 239-251
- Morgan, K., Tromborg, C. (2007). "Sources of stress in captivity." Applied Animal Behaviour Science **102**: 262-302
- Nicolson, V. (2012). Southern hairy-nosed wombat population management plan. Annual report and recommendations. Gold Coast, Dreamworld: 23.
- Papachristou, T. G., L. E. Dziba, et al. (2006). "Patterns of diet mixing by sheep offered foods varying in nutrients and plant secondary compounds." Applied Animal Behaviour Science **108**: 68-80.
- Prince, J. S., W. G. LeBlanc, et al. (2003). "Design and analysis of multiple choice feeding preference data." Oecologia **138**: 1-4.
- Provenza, F., J. J. Villalba, et al. (2003). "Linking herbivore experience, varied diets, and plant biochemical diversity." Small Ruminant Research **49**: 257-274.

- Rees, P. A. (2005). "Will the EC Zoos dietetic increase the conservation value of zoo research?" *Oryx* **39**: 128-131.
- Rogosic, J., J. A. Pfister, et al. (2005). "Sheep and goat preferences for and nutritional value of Mediterranean maquis shrubs." *Small Ruminant Research* **64**: 169-179.
- Simpson, S. and D. Raubenheimer (2012). *The Nature of Nutrition: A unifying framework from animal adaptation to human obesity*. Princeton, Princeton University Press.
- Taggart, D. A., G. R. Finlayson, et al. (2007). "Growth and development of the southern hairy-nosed wombat, *Lasiorhinus latifrons* (Vombatidae)." *Australian Journal of Zoology* **55**: 309-316.
- Taggart, D. A. and P. T. Robinson (2008). "*Lasiorhinus latifrons*." *IUCN Red List of Threatened Species: Version 2011.2* <iucnredlist.org>. Retrieved 26 April, 2012.
- Thom, B., Ed. (2002). *Geographica: The complete illustrated reference to Australia and the World*. Sydney, Random House.
- Treby, D. (2005). Southern hairy-nosed wombat (*Lasiorhinus latifrons*) husbandry manual, Australasian Zoo Keeping: 54.
- Triggs, B. (2009). *Wombats: second edition*. Collingwood, CSIRO Publishing.
- Unknown (2012). "Animal welfare in context." Retrieved 20/05/2012, 2012, from [http://www.ourfutureplanet.org/newsletters/resources/Animals/What is Animal Welfare.pdf](http://www.ourfutureplanet.org/newsletters/resources/Animals/What%20is%20Animal%20Welfare.pdf).
- Villalba, J. J. and F. Provenze (1999). "Effects of food structure and nutritional quality and animal nutritional state on intake behaviour and food preferences of sheep." *Applied Animal Behaviour Science*. **63(2)**: 145-163
- Villalba, J. J. and F. Provenza (2007). "Self-medication and homeostatic behaviour in herbivores: learning about the benefits of nature's pharmacy." *Animal* **1(9)**: 1360-1370.

- Villalba, J. J., X. Manteca, et al. (2009). "Relationship between reluctance to eat novel foods and open-field behaviour in sheep." Physiology & Behaviour **96**: 276-281.
- Villalba, J. J., F. Provenza, et al. (2011). "Preference for diverse pastures by sheep in response to intraruminal administrations of tannins, saponins and alkaloids." Grass and Forage Science **66**: 224-236.
- Villalba, J. J., F. Provenza, et al. (2007). "Learned appetites for calcium, phosphorus, and sodium in sheep." Journal of Animal Science **86**: 738-747.
- Wemelsfelder, F. (2007). "How animals communicate quality of life: the qualitative assessment of behaviour." Animal Welfare **16**(S): 25-31.
- Wemelsfelder, F., T. E. A. Hunter, et al. (2001). "Assessing the "whole" animal: a free choice profiling approach." Animal Behaviour **62**: 209-220.
- Woolnough, A. P. and W. J. Foley (2002). "Rapid evaluation of pasture quality for a critically endangered mammal, the northern hairy-nosed wombat (*Lasiorhinus krefftii*)." Wildlife Research **29**(1): 91-100.
- Woolnough, A. P. (1998). The feeding ecology of the northern hairy-nosed wombat, Lasiorhinus krefftii (Marsulialia: Vombatidae). PhD, James Cook University.
- Zinner, D. (1999). "Relationship between feeding time and food intake in Hamadryas baboons (*Papio hamadryas*) and the value of feeding time as predictor of food intake". Zoo Biology **18**: 495-505
- Zoo and Aquarium Association, A. (2016). "ZAA website." Retrieved 5/5/16, 2016, from <http://www.zooaquarium.org.au/>.
- Zoo and Aquarium Association, (2018). "Animal Welfare Position Statement." Retrieved 26/4/18, 2018, from <https://www.zooaquarium.org.au/index.php/welfare/accreditation/>.

