THE FEEDING ECOLOGY OF THE NORTHERN HAIRY-NOSED WOMBAT, *Lasiorhinus krefftii* (MARSUPIALIA: VOMBATIDAE)

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ABSTRACT

The northern hairy-nosed (NHN) wombat, *Lasiorhinus krefftii*, is critically endangered. It is restricted to a single population at Epping Forest National Park (EFNP) in semi-arid central Queensland. Apart from critically endangered status, the NHN wombat is unique among the larger grazing mammals because of its semi-fossorial lifestyle. This study investigated the feeding ecology of the NHN wombat, a key component in understanding the ecology of this species.

Epping Forest National Park is a tropical savanna. The burrows of the NHN wombat are associated with alluvial sands of a ancient streambed and an open eucalypt woodland. The rainfall at EFNP is seasonal but irregular, and the study was conducted in four years of below average rainfall. The seasonal trends in rainfall result in variation in the quality and quantity of forage available to the NHN wombat.

Fundamental to this thesis was measurement of temporal variability in the forage available to the NHN wombat. Forage quantity was monitored by the BOTANAL technique, allowing quantification of forage attributes including species composition, biomass and ground cover. In addition, primary productivity was monitored during the same period. Forage quality was measured by Near Infra-red Spectrometry (NIRS), calibrated to standard laboratory procedures. This technique allowed precise and reliable measurements of attributes of forage quality. These attributes included total nitrogen concentration, concentration of fibre (neutral detergent fibre, acid detergent fibre and acid-lignin), concentration of carbohydrates (water soluble carbohydrates), *in vitro* dry-matter digestibility (IVDMD) and organic matter content.

The biomass and species composition of forage available to the NHN wombat was dominated by the introduced grass *Cenchrus ciliaris* ($83.1 \pm 6.4\%$ of the total biomass). Of the native grasses, *Enneapogon* spp., *Aristida* spp. and *Chrysopogon fallax* dominated the biomass and species composition. In addition, the sedge *Fimbristylis dichotoma* occurred frequently, despite contributing little to

the overall biomass. The diversity of forbs at EFNP was high, dominated by *Waltheria indica*, *Salsola kalii* and species of the Malvaceae. However, forbs represented only a minor component of the biomass and total species composition. Overall the biomass of the forage available to the NHN wombat was high, above 1000 $kg.ha^{-1}$ (dry matter) year round. Primary productivity was positively correlated with rainfall.

The quality of forage species showed considerable temporal variation, with total nitrogen concentration in particular having a positive correlation with rainfall. Generally, the nutritional quality of the forage was poor, with seasonal variability in the concentration of nitrogen (range 0.3 to 2.5% of dry matter), concentration of fibre (range for neutral detergent fibre 54.9 to 86.8% of dry matter) and IVDMD (range 22.6 to 57.7% of dry matter). How the NHN wombat responded to such poor quality forage, both behaviourally and physiologically, was investigated.

Estimating the species composition of the diet of the NHN wombat is a key issue in an understanding of its feeding ecology. However, techniques used to describe the diets of herbivores have often been questioned for their precision. Consequently three methods were used to estimate the composition of the diet of the NHN wombat: stable carbon isotope analysis, conventional histological analysis and long-chain alkane analysis. Stable carbon isotope analysis partitioned the diet into plants consumed that had a C_3 photosynthetic pathway (essentially forbs) or a C_4 photosynthetic pathway (tropical grasses). Using faecal samples and hair samples (representative of assimilated carbon incorporated into body tissue), it was determined that the diet of the NHN wombat consisted primarily of tropical grasses, but this approach could not describe the botanical composition of the diet.

Analysis of epidermal residues and the concentration of long-chain alkanes in faeces were used to estimate the species composition of the diet. Conventional histological techniques compare the cell patterns of epidermal fragments in the faeces with a reference collection of epidermal patterns of plants available to the herbivore. Long-chain alkane analysis, however, takes advantage of the relatively

indigestible wax component of the epidermis of plants consumed by the herbivore. One of the chemical components of the epidermal wax is long-chain alkanes (C_{25} to C_{36}), each plant species with its own unique signature of alkanes. By comparing the patterns of long-chain alkanes in the faeces with a reference collection of plants available to the herbivore, through a least-squares optimisation procedure, an estimate of the composition of the diet could be generated.

The diet of the NHN wombat was dominated by *Aristida* spp. (20.6 ± 6.0% of epidermal residues), *Enneapogon* spp. (17.2 ± 6.3% of epidermal residues) and *C. ciliaris* (14.5 ± 5.5% of epidermal residues), and to a lesser extent by *W. indica* (2.0 ± 2.0% of epidermal residues) and *F. dichotoma* (1.9 ± 1.6% of epidermal residues). Unidentifiable epidermal fragments were the largest component of the diet determined by the histological technique (40.4 ± 13.2% of epidermal residues). The NHN wombat exhibited little temporal variation in the plants it consumes, suggesting it is a generalist in its forage choice. However, both the histological technique and the long-chain alkane analysis have limitations. Despite these limitations, the estimation of the diet of the NHN wombat in this study is the best possible with current methods.

Body composition of the NHN wombat was investigated by estimating levels of total body fat by Bioelectrical Impedance Analysis (BIA). The method of BIA was successfully calibrated on the southern hairy-nosed (SHN) wombat, *Lasiorhinus latifrons*, using stable isotopes of water and chemical analysis of the carcasses of SHN wombats. Multiple linear regression models using BIA plethysmograph measurements (resistance and impedance) and total body mass, were successful in predicting body fat ($r^2 = 0.90$, S.E. = 1.99) and total body water ($r^2 = 0.90$, S.E. = 1.64). Once calibrated, the BIA was applied to the NHN wombat.

The NHN wombat maintained a relatively constant level of total body fat (7.6 ± 3.5% of total body mass) despite seasonal variability in the quality of available forage. However, there was some evidence to suggest that altered states

of body composition (e.g. the storage of body fat) may be of considerable importance to females during lactation.

The behaviour and activity of the NHN wombat were investigated by radio telemetry with data loggers incorporated into the collar packages. The three-channel data logger measured activity, intensity of light and temperature, with a recording schedule of one record from the three channels every 5 minutes and a data storage capacity of 1 Mb. The ranging behaviour of the NHN wombat was conservative, with core home-ranges averaging only 6.8 ± 3.8 ha (70% harmonic mean activity contour). Likewise, the activity of the NHN wombat was also conservative. The mean number of bouts of activity in a twenty-four hour period was just 25.2 ± 11.1 with each bout lasting on average 7.6 ± 2.2 minutes. Bouts of activity occur both above-ground and within the burrow, although the significance of activities within the burrow is unknown. When above-ground, the NHN wombat maintained low levels of activity, rarely undertaking behaviour that resulted in high measurements of activity. Above-ground activity was positively correlated with rainfall, and hence the quality and quantity of the forage. When rainfall was high, activity was low. However, the ambient temperature may also impose important limitations on the behaviour and activity of the NHN wombat.

The habitat of the NHN wombat was shared by the eastern grey kangaroo, *Macropus giganteus*. The female adult body size of the eastern grey kangaroo is equivalent to the female adult body size of the NHN wombat allowing a comparison of aspects of their feeding ecology. Both herbivores have a preference for grasses with the species composition of their diets overlapping by more than 90%. Unlike the NHN wombat, the eastern grey kangaroo is not reliant on burrows. Following rainfall, the eastern grey kangaroo population declines in response to more favourable forage conditions beyond the boundaries of EFNP. They then return to EFNP as the quality and quantity of forage on surrounding pastoral properties decline. Although the density of eastern grey kangaroos is generally low on EFNP (seasonal range of 0.4 ± 0.6 to 16.9 ± 12.3 animals.km^{-2}), the similarities in forage and habitat preference indicate that there is potential for competition of food resources.

In summary, the lifestyle of the NHN wombat is one of extreme energy conservation, through its relationship with burrows, conservative behaviour and high digestive efficiency. The NHN is able to cope with the seasonally fluctuating savanna of northern Australia. The drought conditions encountered during this study resulted in a forage resource that was generally poor in quality. Despite possible nutrition restrictions, the NHN wombat exhibited little variation in its body composition or body condition throughout the study period. Therefore, the NHN wombat should be regarded as an extraordinary herbivore, well-suited to its semi-arid habitat.

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CHAPTER 1

INTRODUCTION: HERBIVORE FEEDING STRATEGIES

1.1 PREAMBLE

This thesis examines the feeding ecology of one of the three species of wombats, the northern hairy-nosed (NHN) wombat, *Lasiorhinus krefftii*. This wombat is of special significance because it is critically endangered (ANZECC 1995), consisting of a single population of only 65 individuals (Hoyle *et al.* 1995). Previous studies have described the diet of the NHN wombat but in limited detail (e.g. Flößer 1986 and Crossman 1988). These studies have compiled lists of plant species eaten, but have provided limited information on feeding ecology. In this thesis, "feeding ecology" is taken to include the relationships among food availability, diet selection, and feeding activity, and the effects of variation in the quality and quantity of food on feeding behaviour. Information of this kind is essential to predict the responses of the population to natural variation in its food supply, and to plan management of habitat with the objective of increasing the size of the population.

Wombats have a suite of ecological and physiological characteristics that set then apart from all other herbivores. Adult wombats can weigh as much as 40 kilograms yet they are capable of leading a semi-fossorial existence. This is truly unusual since wombats are more than twice the size of the next largest burrowing herbivore. Furthermore, compared to other mammals, wombats have very low maintenance energy and nitrogen requirements (Barboza *et al.* 1993, Wells 1978a). For a large herbivore with a capacious colon, wombats have long retention times of particulate matter and are capable of maintenance on relatively low rates of intake (Barboza 1989). The feeding ecology of the wombat should therefore reflect these characteristics.

Hairy-nosed wombats (*Lasiorhinus* spp.) occupy habitats that are characterised by low rainfall and high drought frequency, with low plant quality and productivity and generally very low nutrient status of soils and plant tissues. In any given year *Lasiorhinus* spp. can be exposed to a wide range of forage conditions. The feeding strategy of *Lasiorhinus* should therefore be capable of compensating for changes in food quality and quantity, as well as confering an ability to survive on a very low nutrient base. How the NHN wombat responds to changes in food quality and quantity is the central theme of this thesis.

The semi-arid tropical savanna habitat of the NHN wombat can be variable in climate and forage conditions. Several predictions of how the NHN wombat may respond to these variable conditions, both physiologically and behaviourally, can be made. First, the NHN wombat may vary food selectivity. This is a primary response by many herbivores to changes in food quality and quantity. Second, the NHN wombat may vary rates of intake, again as forage quality and quantity vary. Third, the NHN wombat may store and mobilise fat in response to changes in food quality and quantity, laying down fat when the quality and quantity of food are greatest and mobilise fat reserves when forage becomes limiting. Fourth, the NHN wombat may be able to maintain a continually low energy expenditure so that it is effectively buffered against fluctuations in food quality and quantity. This response may also include minimising the energy costs of reproduction. Although not examined in this thesis, minimising the energy costs associated with reproduction could occur if the rate of reproduction is low or reproduction is deferred until times when energy availability is relatively high. If energy expenditure is continually low, this may reduce the need for the wombat to employ any of the previously described strategies.

The scope of this thesis extends beyond the NHN wombat to include some aspects of the feeding ecology of the eastern grey (EG) kangaroo, *Macropus giganteus*. This grazing herbivore is similar in size to the NHN wombat (adult females are both approximately 30kg) yet has a different feeding strategy and digestive morphology. From a conservation and management perspective, the EG kangaroo has the potential to become a competitor for food resources with the

NHN wombat. This needs to be quantified. From an ecological perspective, study of the eastern grey kangaroo offers the opportunity to compare the feeding strategies of a sedentary herbivore (wombat) with a mobile herbivore (kangaroo).

In order to address many of the issues examined in this thesis, standard techniques have required critical validation and new techniques have been developed specifically for wombats and their habitat. For example, the extremely shy and nocturnal habits of the NHN wombat do not allow conventional approaches, such as direct observations, to describe above-ground activity or feeding. Instead, the uncertainties of these conventional techniques need to be overcome with new approaches. Likewise, the available methods to examine body condition have low sensitivity, therefore there is a need to investigate and calibrate alternatives. This is especially important given that a herbivore that feeds on a particularly low quality diet is unlikely to be able to rapidly lay down fat in a way that can be easily detected by morphological measurements or body weight changes. Indeed, the focus on the methodological aspects of this thesis is equally important to the interpretation of the feeding ecology of the NHN wombat. Furthermore, the methods established in this thesis will become central to the future research and management of the NHN wombat as well as other ecological studies of free-ranging mammals.

To establish the framework for this thesis the following introduction reviews some of the key determinants of herbivore foraging activities, specifically those relevant to the ecology of wombats. These include:
(1) digestive anatomy and physiology of herbivores and their consequences for forage selection and foraging behaviour;
(2) how temporal and spatial variation in the herbivore's food resource can place constraints on foraging and ranging behaviour of the herbivore; and
(3) wombat biology and the mechanisms available to wombats to compensate for variation in diet quality and quantity.

To begin the review it is necessary to first consider the foraging environment of the NHN wombat - tropical grasslands - and how this forage resource may affect diet selection.

1.2 DYNAMICS OF SAVANNA GRASSLANDS

Grazing ecosystems involve a complex, integrated flow of energy and nutrients between soil, plants and the animal (Georgiadis and McNaughton 1990, McNaughton and Banyikwa 1995). Among grazing ecosystems, tropical savannas represent a high degree of variability in energy and nutrient flow, affecting plant nutrition, biomass and species composition.

The foraging environment of the NHN wombat can broadly be classified as a tropical savanna grassland. Huntley (1982) describes a 'savanna' as an "ecosystem in which C_4 grasses dominate the herbaceous stratum". The Australian tropical savanna is characterised by a limited period of summer rain which stimulates rapid plant growth that then declines during a winter dry season. It is also characterised by both seasonal drought and long-term drought. Other characteristics of the Australian tropical savanna typically include high temperatures and low nutrient concentrations in the soil (Walker and Gillson 1982).

A broad generalisation is that the driving force of the tropical savanna is rainfall. Spatial and temporal variability of rainfall results in stochastic episodes of primary productivity leading to spatial heterogeneity in biomass, nutritional quality and species composition (McNaughton 1985). These pulses of primary productivity, associated with rainfall, are often unpredictable but result in tropical savannas having consistently greater quantity of plant biomass than temperate grasslands. During this rapid growing season, the nutritive value of the forage can be high but it is not sustained throughout the year. Therefore, the mean nutritive value of temperate grasses is greater than tropical grasses in the long-term.

Apart from rainfall, light, temperature, soil nutrients and grazing influence the physiology of grasses, and hence their nutritional quality for herbivores (Olubajo *et al.* 1974, Georgiadis and McNaughton 1990, Senanayake 1995). The combination of high light levels and high temperatures in tropical ecosystems results in rapid plant growth following rain. Rapid growth results in rapid

lignification of the cell wall with tropical grasses having a relatively higher fibre content than temperate grasses.

The herbivore's ability to absorb and metabolise plant material is reduced by the content of fibre, and in turn lignin (Van Soest 1965), with acid lignin resistant to the enzymatic and acid hydrolysis processes of digestion (Robbins 1983). Consequently the herbivore grazing in a tropical savanna environment needs to be adapted to a diet high in fibre and lignin but low in digestibility.

Other nutritional factors affecting herbivore foraging in a tropical savanna include low available protein and carbohydrates. Crude protein levels deteriorate from a peak in the growing season (up to 14% of dry matter in semi-arid Australian savannas) to levels that can affect the nitrogen balance of a herbivore at or well before the end of the dry season. Reserves of non-structural carbohydrate reserves in herbage follow similar temporal declines (Adjei *et al.* 1988).

The dynamics of a grazed savanna ecosystem are of major significance for diet choice of the grazing herbivore. Gross temporal changes in forage quality and quantity require the herbivore to be flexible in its feeding strategy. Generally, peaks in forage quality and quantity present the herbivore with abundant high-quality food. Some herbivores (e.g. wildebeest, impala and nyala) use these peaks to compensate for increased energy demands associated with breeding and lactation, and/or to store energy in the form of body fat (e.g. Van Rooyen 1993, Murray 1995).

Troughs in quality and quantity of forage limit forage choice by herbivores, forcing behavioural compensation and/or efficient mechanisms of digestive function. For any study of the feeding ecology of a grazing herbivore foraging in a tropical savanna habitat it is important to measure temporal changes in the quality and quantity of forage. Potential compensation mechanisms by the herbivore to temporal variation in the quality and quantity of the forage may occur such as variation in diet selection, physiological responses and behavioural responses.

1.3 HERBIVORE DIGESTIVE STRUCTURE & FUNCTION

The following section introduces some of the general concepts of herbivore digestion. In particular, it briefly describes the principal differences between foregut and hindgut fermenting herbivores. It then addresses the issues of body size, metabolic rate and digestive function, particularly in relation to the Vombatidae.

Diet selection by an individual herbivore is influenced by its morphological, physiological and behavioural characteristics; quality and quantity of food plants available in the immediate foraging vicinity; and community factors such as other herbivores or predators (Pratt and Gwynne 1977, Owen-Smith 1982, Jarman 1993). Large herbivores must be able to exploit their foraging environment in response to the spatial and temporal variability of the habitat (Georgiadis and McNaughton 1990). As a consequence, herbivores have a suite of adaptations and compensation mechanisms for a grazing lifestyle.

1.3.1 Food Choice and Mastication

Grasses generally have high silica and lignin concentrations (Georgiadis and McNaughton 1990). Consequently the consumption of grasses by herbivores has led to the evolution of efficient teeth in grazers (Hilderbrand 1988, Hume 1989, Sanson 1989). The primary function of the grazer's teeth is to structurally alter the food and reduce the particle size to maximise digestion (Hume 1989).

Two dental configurations may be used among the grazers: hypsodonty and continuously growing teeth (Hilderbrand 1988). Parallel evolution of hypsodonty has occurred in both the ungulates and the Macropodidae as they coevolved with the grasslands (McNaughton *et al.* 1985). Similarly, parallel evolution of continuously growing teeth has occurred amongst the rodents and the wombats (Hilderbrand 1988). The muscles and structure of the mandible are also important features of the grazer's teeth. The adductor mass of many grazers is dominated by the masseter muscles to enable sustained chewing and to overcome the heavily

lignified cell wall of grasses (Hilderbrand 1988, Hume 1989). The structure and position of teeth within the mandible constrain food choice by influencing bite dimensions, rates of chewing and consequently intake rates (Janis 1976, Trudell and White 1981, Gordon *et al.* 1996).

1.3.2 Digestive Morphology and Physiology

The chemical and physical processes of digestion facilitate the assimilation of nutrients for energy, maintenance, growth and reproduction (Stevens and Hume 1996). Specific details of digestion and digestive processes are described in detail by Stevens and Hume (1996), and specifically for wombats by Barboza (1989). However, for wombats the process of digestion (including the ingestive processes of searching and selection) and digestive morphology are important characteristics that make wombats unique (Barboza 1989).

Herbivores can be separated into two groups depending on the dominant site of fermentation: foregut or hindgut fermenters. The following describes principal elements of foregut and hindgut fermenters in relation to foraging activities and process of digestion.

1.3.2.1 Foregut Fermenters

Foregut fermenters are characterised by an enlarged multi-compartmental stomach that serves as the dominant site of microbial activity. This results in fermentative digestion occurring prior to gastric intestinal digestion (Parra 1978). Foregut fermenters can be differentiated further into ruminants and non-ruminants.

Ruminants

Ruminants are characterised by their ability to ruminate (regurgitation, remastication, and reswallowing) (Stevens and Hume 1996). The process of rumination allows rapid food consumption with little or no chewing at a grazing location. Then, at the animal's convenience, it regurgitates and comprehensively masticates the food before reswallowing, usually in a place of relative safety (Eckert *et al.* 1988). Ruminants digest more fibre than other groups of foregut fermenters because non-ruminant foregut fermenters have comparatively higher rates of passage through the forestomach (Dellow 1982, Dellow and Hume 1982,

Dellow *et al.* 1983). In addition, ruminants reduce food particle size further than non-ruminants through the remastication of food particles (Poppi *et al.* 1985).

Non-ruminant foregut fermenters

Non-ruminant foregut fermenters also use the forestomach for microbial fermentation but do not ruminate. Hume (1989) differentiated this group further on the basis of the 'chemical reactor theory' of Penry and Jumars (1986 & 1987) into dominant continuous flow, stirred tank reactors (CSTR) and plug-flow reactors (PFR).

The CSTR system operates on a continuous flow of material that is continuously mixed within the system (Penry and Jumars 1986 & 1987). All of the non-ruminant foregut fermenters (with the exception of some Macropodidae) and ruminants use a dominant CSTR type system (Penry and Jumars 1986, 1987). For maximum efficiency of the CSTR type system these non-ruminants have a sacciform forestomach to maximise retention of digesta for fermentation (Hume 1989).

The alternate PFR system (following a CSTR in some Macropodidae) is characterised by a continual flow of material through the system, with limited longitudinal mixing of digesta (Penry and Jumars 1986 & 1987). This process is employed by some of the larger members of the Macropodidae and is characterised by a grossly tubiform forestomach (Hume 1989). Food passes more rapidly through the tubiform forestomach compared to a sacciform stomach (e.g. McIntosh 1966, Delow and Hume 1982). However Macropodids masticate their food more thoroughly than ruminants, thereby physically aiding microbial fermentation. In addition, the series of PFR's used by the large macropods is similar in digestive process to that of large hindgut fermenters (Hume 1989). By maintaining relatively high rates of food intake (Hume 1989), non-ruminant foregut fermenters can compensate for a decreasing rate of fermentation along the tubiform stomach (Dellow *et al.* 1983).

1.3.2.2 Hindgut Fermenters

Hindgut fermenters are differentiated from foregut fermenters where by microbial fermentation occurs along the gastrointestinal tract. In hindgut fermenters, gastric and enzymatic digestion occurs before microbial digestion (Parra 1978).

Hindgut fermenters can be broadly divided into two functional groups (caecum or colonic fermenters) based on the site of microbial fermentation and digestive tract morphology. There is a clear body mass difference between these functional groups with caecum fermenters being small (range 0.014 to 79 kg), and colonic fermenters being large (range 175 to 7500 kg) with the exception of the Vombatidae (19 to 39 kg) (Table 1.1).

Caecum Fermenters

The caecum is a "pouch-like structure" (Hume 1989) found at the junction between the small and large intestines, and may function as a CSTR (Hume 1989). A characteristic of some hindgut fermenters is a relatively rapid passage of ingesta compared to ruminants. This group of herbivores adapts to the digestive constraints of plant fibre and rapid passage by feeding selectively on low fibre foods (Demment and Van Soest 1985).

When the protein content of forage plants becomes limiting some caecum fermenters may perform coprophagy (Demment and Van Soest 1985). The re-ingestion of faeces allows further microbial digestion and digestion of microbial protein in the small intestine. The few herbivores that regularly practice coprophagy have the ability to produce and distinguish two different types of faeces through selective separation of caecal contents at the ileo-caecal junction (Santomá *et al.* 1989). The re-ingestion of nitrogen-enriched faeces allows efficient utilisation of protein and other dietary components (vitamins and minerals) resulting in relatively high digestive efficiency (Santomá *et al.* 1989, Borges *et al.* 1996).

Table 1.1. Principal site of microbial fermentation in the gastrointestinal tract and range of adult body sizes for mammalian grazing herbivores. Sources: Nowak (1991) and Strahan (1995). Only non-arboreal grazing mammals have been included.

Order	Family	Ranges in Body Mass of Family* (kg)	Site of Microbial Fermentation
Marsupialia	Macropodidae	3.2 to 66	Foregut - Non Ruminant
Artiodactyla	Suidae	50 to 150	
Artiodactyla	Hippopotamidae	1000 to 4500	
Artiodactyla	Camelidae	55 to 690	Foregut - Ruminant
Artiodactyla	Cervidae	7 to 825	
Artiodactyla	Bovidae	14 to 1200	
Rodentia	Muridae	0.014 to 4	Hindgut - Caecum
Rodentia	Sciuridae	0.7 to 1.4	
Lagomorpha	Leporidae	0.125 to 7	
Rodentia	Caviidae	0.3 to 16	
Rodentia	Hydrochaeridae	27 to 79	
Marsupialia	Vombatidae	19 to 39	Hindgut - Colonic
Perissodactyla	Equidae	175 to 930	
Perissodactyla	Rhinocerotidae	1400 to 3600	
Proboscidea	Elephantidae	2700 to 7500	

* Grazing members only

Colonic Fermenters

Colonic fermentation uses the PFR process of digestion. By devoting a large proportion of the day to foraging, colonic fermenters are able to maintain high food intakes. For example, equids feed for up to 15 hours per day compared to only 10 for a bovid of comparable body mass (Duncan *et al.* 1990). Further, equids maintain dry matter intakes of 170 $g.kg^{-0.75}.d^{-1}$ compared to only 115 $g.kg^{-0.75}.d^{-1}$ for bovids of comparable body mass (Duncan *et al.* (1990). To cope with the bulk of the diet, the colon is extremely voluminous to facilitate microbial fermentation (Stevens and Hume 1996). Haustration of the colon in some hindgut fermenters aids in increasing retention time of digesta (Stevens and Hume 1996).

The relatively small body mass of wombats compared to other grazing colonic fermenters (Table 1.1) would suggest that they would be better served by caecum fermentation since they are within the range of body sizes of caecum fermenters and not colonic fermenters. Further, wombats should be inefficient as colonic fermenters because their semi-fossorial lifestyle, and thermal intolerance (Wells 1978a), is not conducive to long foraging bouts often necessary to maintain high rates of food intake. However, colonic fermentation in the Vombatidae is extremely efficient (Barboza 1989, Barboza and Hume 1992). Specific features of the wombat digestive system are a large ratio of gastrointestinal (GI) tract to total body length (TBL) and a particularly voluminous colon (Barboza 1989, Barboza and Hume 1992a&b). These features result in a large digestive capacity, a high potential for absorption and resorption of nutrients, and long retention time of digesta (Barboza 1989, Barboza and Hume 1992a, Barboza 1993a, Wells 1973). Further, efficient chewing of forage reduces the particle size of ingested forage, increasing the digestion of fermentable cell walls (Wells 1989).

1.3.2.3 Body Size and Digestion

The relationship between gut capacity and body mass in herbivores is essentially linear, with the ratio of metabolic rate (MR) to gut capacity (GC) declining with body mass (Demment and Van Soest 1985). Body mass therefore has direct implications for minimum required food quality (Illius and Gordon 1993)

with smaller herbivores having "less digestive capacity per unit of metabolic need" (Gross *et al.* 1996).

To test the notion that smaller herbivores have a lower digestive capacity, Gross *et al.* (1996) examined intra-specific digestion and passage rates two types of hay in the sexes of dimorphic Nubian ibex, *Capra ibex nubiana*. Demment and Van Soest's (1985) ideas suggests that male Nubian ibex (approximately three times larger in body size than females) should retain food longer in the digestive tract and digest it more completely (Gross *et al.* 1996). In general, Gross *et al.* (1996) found that males did have longer retention times than females, but the extent of digestion was comparable between males and females. While the rate of passage of both large particulate matter (determined by Cr-mordanted fibre) and small particulate matter (determined by Co-EDTA in a liquid phase) was consistent with the predictions of Demment and Van Soest (1985), Gross *et al.* (1996) attributed the digestion rate of forage in females to higher rates of mastication by females compared to males.

However, individual gut size is variable and needs to be considered when comparing rates of digestion and rates of passage, particularly in ecological studies where environmental conditions are not static. For example, Hammond and Wunder (1991) found that the prairie vole, *Microtus ochrogaster*, had a larger volume of the hindgut when the diet was high in fibre. Further, Hammond and Wunder (1991) found that prairie voles increased their rates of food intake in response to cold acclimation, that led to an increase in the size of the small intestine and caecum.

Demment and Van Soest (1985) suggest that the ratio of MR to GC (MR:GC ratio) is a key component in the evolution of body size and feeding behaviour of herbivores. These authors further suggest that the MR:GC dictates whether the herbivore should be a foregut or a hindgut fermenter (Demment and Van Soest 1985). Wombats, however, do not comply to this generalisation.

Generally, small mammalian herbivores have high metabolic requirements per unit body mass and consequently a high MR:GC. Limitations are therefore

imposed by digestive tract volume for small mammals, even though this volume can be variable (Hammond and Wunder 1991). Consequently, hindgut fermenting herbivores of small body size should select high quality food and/or practice coprophagy to maximise nutrient intake (Hume 1989, Yamauchi and Iwasa 1995). As body size increases, the retention time of forage also increases resulting in a higher digestive efficiency and allowing a greater return of energy per unit of food (Illius and Gordon 1993).

The complex structure of the forestomach in ruminants selectively delays the passage of ingesta depending on particle size (Demment and Van Soest 1985, Poppi *et al.* 1985). Demment and Van Soest (1985) suggest that medium sized ruminants have a comparatively higher rate of fibre digestion than a hindgut fermenter of similar body size. Consequently, the hindgut fermenter requires a higher dry matter intake to extract the same amount of nutrients as a foregut fermenting herbivore foraging on the same diet (Hume 1978). To compensate for the hind-gut fermenter's apparent lesser ability to extract nutrients than a foregut fermenter of equal body size, the large hindgut fermenter adopts a 'bulk feed' approach to foraging. To maximise the rates of intake, hind-gut fermenters usually forage for comparatively longer than a foregut fermenter and/or have an efficient bite size and rate (Bunnell and Gillingham 1985). If food is abundant and of poor quality, colonic fermentation becomes more efficient than rumination (Duncan *et al.* 1990), since the higher relative rate of passage allows greater relative food consumption, outweighing the consequent reduction in digestive efficiency (Illius and Gordon 1993).

If colonic fermenters are more efficient in extracting more nutrients per day than ruminants when food is available *ad libitum*, "why is the grazing herbivore biomass dominated by medium sized ruminants?" (Duncan *et al.* 1990). Duncan *et al.* (1990) suggest three answers to this question. First, digestible protein rather than dry matter digestibility may be a more appropriate measure of nutrient extraction, particularly in tropical foragers. Second, the ability to convert digestible energy to metabolic energy may differ between colonic fermenters and ruminants.

Third, energy requirements for maintenance may be slightly higher in colonic fermenters than ruminants.

The comparisons made by Duncan *et al.* (1990) were between equids and bovids. In a comparison between wombats (colonic fermenters) and large macropods (non-ruminant foregut fermenters) that are matched almost exactly for female body mass, wombats have lower dietary nitrogen requirements (Barboza *et al.* 1993) and lower field-measured basal metabolic rate (Wells 1973, Evans unpublished data) than large macropods. This suggests that the first and third predictions made by Duncan *et al.* (1990) do not apply in this comparison and the second prediction is unlikely.

The previous discussion shows that the body mass of the wombat is small in relation to other colonic fermenters (Table 1.1) and suggests it should have a high MR:GC ratio. Consequently the wombat should be forced to maintain high rates of intake and inhabit areas of high food abundance to compensate. So how can the wombat survive using such an apparently inappropriate digestive system? By virtue of its burrowing behaviour the wombat maintains one of the lowest mammalian field basal metabolic rates (Wells 1973, Murray Evans unpublished data). This in turn reduces the MR:GC ratio, allows efficient use of comparably modest rates of intake and has resulted in wombats occupying a variety of habitats where the quality and quantity of forage can be variable.

1.4 Herbivore Foraging Decisions

1.4.1 Mechanistic Approach to Foraging

The interaction between a herbivore and the plants it consumes is complex. The foraging decisions of a herbivore can be directly attributed to the consequences of consuming a biologically complex food resource (McNaughton and Georgiadis 1990, Hirakawa 1997). A herbivore should aim to optimise the intake of energy, protein, water, minerals and vitamins, all of which are necessary for maintenance and reproduction (Robbins 1983). However, the plants consumed by the herbivore may vary temporally and/or spatially in nutritional quality, or present a herbivore with a suite of anti-feedants. Therefore, selection of food by a

herbivore is complex. The following section details one of several theoretical explanations of food selection by a herbivore: the mechanistic approach to foraging.

The mechanistic explanation of foraging suggests that the herbivore will forage in response to one or more nutritional requirements (Belovsky 1984). That is, to interpret an action (forage selection) in relation to one or more reactions (environmental and/or morphological or physiological adaptations). Parameters often investigated include nutrient requirements, diet quality, plant secondary compounds, thermal balance, species interaction, body size and morphophysiology, or resource density (e.g. Owen-Smith 1982, 1992, 1993; Spalinger and Hobbs 1992, Kohlmann and Risenhoover 1994, Gordon *et al.* 1996, Gross *et al.* 1996, Tixier *et al.* 1997). Predictions about foraging choice can then be generated. This approach can also generate mathematical models, like the optimal foraging approach. The mechanistic approach to foraging can also be used to test or generate new optimal foraging models and thereby complements optimal foraging theory. The following discussion highlights several of the parameters of specific relevance in this thesis.

Diet Quality & Quantity

Two approaches have been used to evaluate the types of foods eaten by herbivores. The first approach examines diet choice in relation to specific chemical components of the plant ingested by the herbivore such as protein (e.g. Coppock *et al.* 1983), various carbohydrates (e.g. Adjei *et al.* 1988, Radojevic *et al.* 1994), trace elements such as sodium (e.g. Georgiadis and McNaughton 1990, Wallis de Vries and Schippers 1994), and secondary compounds. For example, Tixier *et al.* (1997) found that roe deer, *Capreolus capreolus*, selected plants with a high concentration of soluble sugars, high concentrations of protein-binding phenolics and elevated *in vitro* dry matter digestibilities (IVDMD), but avoided plants with high fibre concentrations and were indifferent to varying concentrations of total protein. These results suggest that *C. capreolus* selects plants on the basis of their chemical composition.

The second approach is essentially a measure of digestibility. It assumes the digestibility of a forage is a major determinant of intake if there is little post-digestion loss of energy (Van Soest 1965, Hirakawa 1997). However, this approach is an extension of the first with the quantification of IVDMD effectively describing the digestive process of the herbivore in relation to the quality of the ingested forage (Hirakawa 1997).

When the mechanistic approach to forage choice is applied to herbivores grazing in tropical savannas, key factors influencing forage choice may include crude protein levels, digestibility and forage quantity. In tropical grasslands, crude protein content of forages can often be limiting during the dry season (Owen-Smith 1982). Protein quality (e.g. limited amino acids) may result in the herbivore struggling to consume enough protein for maintenance, thereby affecting its body composition and condition. Conversely, fibre content of tropical forages is higher than temperate grasses so much so that it may deter forage consumption. Further, aspects of forage quantity can and do influence forage consumption by herbivores.

Growth stage, available biomass, forage density and composition, and the influence of previous feeding bouts may influence herbivore diet selection (Robbins 1983). As forage biomass and density decline, large herbivores modify their behaviour by increasing bite size (Demment and Greenwood 1988), and reduce specialisation on plant species and plant parts (Weckerly 1994). The increase in bite size allows the herbivore to maintain a constant rate of intake but the quality of plant parts ingested is reduced, as is the time spent cropping and chewing the plants (Trudell and White 1981, Demment and Greenwood 1988). In conjunction with changes in bite size and bite rates, the time allocated to foraging and home range size increases as the availability of biomass declines (Trudell and White 1981).

Plant structure is also a key component of herbivore grazing systems. Generally, grazing time increases and intake decrease as pastures become shorter (Arnold and Dudzinski 1969). At a finer resolution, the leaf length of a pasture can affect rates of intake by a herbivore (e.g. Arnold and Dudzinski 1969). While it is likely that plant structure may influence foraging decisions by the NHN wombat

this has not been examined in this thesis. This is primarily because the pastures in the habitat of the NHN wombat tend to be consistently sparse and open with little variation in growth form among the plants they consume with the exception of an introduced grass, *Cenchrus ciliaris*.

Herbivore-Herbivore Interaction

Forage selection by one herbivore may be influenced by the presence of another herbivore within the same ecosystem. The presence of two herbivores in the same ecosystem may result in interspecific competition if the food resource becomes limiting (Dawson *et al.* 1992). Interspecific competition can eventually lead to competitive exclusion of one herbivore from a habitat (e.g. Krausman 1978). Alternatively the resources maybe partitioned. Resource partitioning allows species to co-occur through preference for different food types (plant species or plant parts) and/or feeding styles (Jarman 1974, Jarman and Sinclair 1979).

Competition and resource partitioning between herbivores can often interchange. For example, the European rabbit *Oryctolagus cuniculus*, the western grey kangaroo *Macropus fuliginosus*, the red kangaroo *M. rufus* and the southern hairy-nosed (SHN) wombat *Lasiorhinus latifrons* are four grazers of the Australian semi-arid zone that occur in sympatry in the Murraylands District of South Australia (Dierenfeld 1984). During non-drought conditions there is a high overlap in dietary preference among the four herbivores (Wells 1973, Dierenfeld 1984, St John and Saunders 1989, Kean 1990) with no measurable resource competition. When resources become limiting the potential for competition between the herbivore species exists. However, Dierenfeld (1984) found that when forage resources became limiting through drought, the herbivores coexisted through resource partitioning and did not compete for limited forage resources. This would maximise each herbivore's ability to survive periods of drought.

1.4.2 Variation in Food Resources - Herbivore Compensation Mechanisms

One problem that governs food choice by herbivores is temporal variation in quantity and quality of their food resource. This is most apparent in the distinctly seasonal environments of grasslands, savanna grasslands, tundra and alpine

regions. To cope with seasonal variability, herbivores employ both behavioural and physiological compensation mechanisms.

1.4.2.1 Behavioural Compensation Mechanisms

Herbivores living in fluctuating tropical savanna environments are confronted with two hypothetical choices: (1) move to an area of higher nutritional potential or (2) remain in residence and cope with fluctuations in nutritional quality and quantity. The benefits of migration are clear: it allows the herbivore to access resources that are higher in quality. However, there are substantial costs, the primary one being the energetic cost of long-range movements. Also, the benefits derived from migration can only be gained if a herbivore can find a resource of higher quality. The alternate strategy is where a herbivore remains sedentary. A sedentary herbivore, therefore, needs to be able to survive periods when the nutritional quality of the food resource is low.

The classic example of the two approaches is the tropical savanna ecosystem of the Serengeti-Mara ecosystem of Tanzania and Kenya. The animal biomass of the Serengeti-Mara ecosystem is dominated by herbivores that have very large home ranges and are considered migratory herbivores, but herbivore biodiversity is dominated by resident herbivores faithful to small home ranges (Maddock 1979, Fryxell *et al.* 1988, McNaughton and Banyikwa 1995). The migration of the herbivores is driven by the wet-dry seasonality of the ecosystem, which also dictates foraging behaviour. Migration occurs in succession, with first the Zebra, second the Wildebeest, followed finally by Thomson's gazelle (Gwynne and Bell 1968). During the wet, each of the herbivores follow the rains, and graze in a limited area for an extended period of time (McNaughton 1985, McNaughton and Banyikwa 1995). The dry season is characterised by 'rotational-passage' grazing, where the herbivores spend little time in one area, but cover a large area and often re-cover areas already grazed (McNaughton 1985, McNaughton and Banyikwa 1995). Within the migratory herbivore community, diversity of food preference, particularly preferred plant parts, results in resource partitioning and allows co-occurrence (Gwynne and Bell 1968, Jarman and Sinclair 1979, McNaughton 1985).

The resident herbivore communities in the Serengeti-Mara ecosystem are not uniformly distributed and appear to concentrate on areas with higher rainfall and high availability of some nutrients such as sodium and potassium (McNaughton and Banyikwa 1995). It has also been suggested that aggregations of one herbivore may facilitate the presence of another by reducing the threat of predation or enhancing forage quality through nutrient enhancement (Giraldeau 1984, Sinclair 1985, McNaughton and Banyikwa 1995, Fryxell 1995).

Within the resident herbivore communities different feeding styles are used to cope with variations in food resources (Jarman 1974). Some groups of herbivores have high selectivity of plant species and/or plant parts. Others adopt a non-selective foraging style and flexible home ranges. These alternate feeding styles allow coexistence through resource partitioning.

The current study presents the opportunity to compare the response of two grazing herbivores of similar body size to seasonal changes in quality and quantity of the forage resource. The eastern grey kangaroo, *Macropus giganteus*, is considerably more mobile than the NHN wombat (Johnson 1991a). The eastern grey kangaroo therefore has the opportunity to move in response to changes in forage conditions, but the movements of NHN wombat are somewhat restricted because of their dependence on burrows and could therefore be regarded as sedentary. Diet selection of the NHN wombat and the eastern kangaroo are described in Chapters 3 & 6 respectively, and movements of the NHN wombat and eastern grey kangaroo are discussed in Chapters 5 & 6 respectively.

1.4.2.2 Physiological Compensation Mechanisms

Changes in quality and quantity of the forage resource represent a change in energy potentially available to a herbivore. In large herbivores foraging on a naturally variable food resource such as in savannas, the changes in nutritional quality, and hence energy intake, can be substantial. For example, a model by Gordon and Illius (1996) describes the allometric relationship between body mass and forage quality. They suggest that the rate of energy assimilation "is the product of rate of intake and rate of digestion of forage" (Gordon and Illius 1996) which is affected by the quality of the forage (Illius and Gordon 1999).

As discussed above, herbivores can respond to these changes in forage quality by a change foraging behaviour. Many animals, however, adapt to seasonally variable habitats physiologically by altering the dimensions of the gastrointestinal tract (Hammond and Wunder 1991) or storing energy in the form of body fat (Pond 1978, Prince and White 1985). The deposition and mobilisation of body fat, and to a lesser extent body protein, serve as compensation mechanisms in a seasonally variable environment (Parker *et al.* 1993), with fat deposition occurring during times of 'plenty' and mobilisation of fat occurring when food becomes limiting. However energy storage is not the only function of body fat; it also serves to insulate the body, provide mechanical support and protection for organs, and can provide sexual and social signals (Pond 1978).

Investigations of body composition of SHN wombats suggest the storage of fat is an important ecological adaptation to seasonal variability of available forage (Gaughwin 1981). Body fat in wombats is likely to primarily serve as an energy store rather than a mechanism for thermoregulation (insulation) or as a secondary sexual character because thermoregulation in wombats is regulated by behaviour and social interactions are largely through olfaction (Wells 1973, Gaughwin 1981). Gross changes in body fat of the NHN wombat are likely to represent an energy storage mechanism to compensate for extreme changes in quality and quantity of the food resource. This is investigated in Chapter 4.

Although not investigated in this thesis, changes in gut morphology may be an important adaptation to changes in the forage environment of the NHN wombat. Barboza (1989) found that the gut contents of captive common wombats, *Vombatus ursinus,* and SHN wombats were less (28-33% and 13% respectively) than predicted values for gut contents derived from Demment and Van Soest (1985). However, Barboza (1989) found that the predicted value for wild caught SHN wombats was only 2% less than the predicted value. Although not investigated in detail by Barboza (1989) it appears that gut volume of both common wombats and SHN wombats is plastic in response to changing forage quality and quantity.

1.4.2.3 Burrowing as a Compensation Mechanism

Burrowing is quite common among mammalian herbivores, particularly those of small body size (under 5 kg). The burrow offers shelter from the environment, provides a means of predator avoidance, offers a site for food storage, or in the truly fossorial mammals such as the naked mole rat (*Heterocephalus glaber*), a foraging environment (Reichman and Smith 1990). There are however costs associated with a burrowing lifestyle. The burrow is central to the ranging behaviour of wombats, influencing both behaviour and physiology. Therefore the following section reviews the cost and benefits of living in a burrow, with particular emphasis on their implications for wombat biology.

Energetic Cost of Construction and Maintenance

The construction of mammalian burrows has several constraints: the physical dimensions of the burrow; geological substrate in which the burrow is constructed; and the energetic cost of construction. Wombats construct burrows in a variety of soil types. The NHN wombat however restricts burrow construction to alluvial sands. Burrow construction by the pocket gopher (*Thomomys bottae*) in alluvial sand is approximately 10 times more energy efficient than burrowing in clay soils (Vleck 1979), but offers less structural support. Consequently, 86% of NHN wombat burrow entrances use tree roots for structural support (Crossman 1988).

Construction of a burrow has substantial energetic costs to the burrower, with ongoing energetic costs through the maintenance of the burrow. This initial outlay of energy can be substantial. For pocket gophers, the energy required for burrowing is between 360 to 3400 times that of walking the equivalent distance that has been burrowed, depending on the substrate in which the burrow has been constructed (Vleck 1979). Consequently, the long term gains of living in a burrow need to exceed the initial energetic cost of construction. As this initial energetic outlay is substantial, wombats minimise the initial energetic costs by extending existing burrows and inheriting maternal burrows (e.g. Johnson and Crossman 1991).

Thermoregulation

An animal's ability to balance heat production and heat loss is a function of its body mass and thermal conductance. Burrowing mammals exhibit lower mass specific metabolic rates than other terrestrial mammals (McNab 1979), with hairy-nosed wombats having the lowest field metabolic rate of all marsupials so far examined (Wells 1973, M. Evans unpublished data). If heat is difficult to dissipate in a burrow, then a low metabolic rate may be advantageous for a fossorial or semi-fossorial lifestyle (McNab 1979, Conteraras and McNab 1990). Large burrowing mammals have a relatively high thermal conductance to reduce heat storage in the burrow and compensate for their smaller surface to volume ratio (McNab 1979). Also, the burrow environment contributes to the burrowing mammal's ability to thermoregulate by providing a microclimate stable in temperature and humidity (Withers 1978, Maclean 1981). This in turn reduces heat loss through conduction, convection and evaporation to minor pathways of heat loss in a burrowing animal (Maclean 1981).

Gas Exchange and Water Balance

Burrow architecture influences gas composition and relative humidity within the burrow, thereby affecting respiration and water balances of the burrower (Withers 1978). High relative humidity within the burrow can minimise evaporative water loss when burrow temperature is sufficiently high (Reichman and Smith 1990). Coupled with a reduction in water use associated with reduced metabolic rate, the burrow environment offers a means of balancing the water budget (McNab 1979). For *Lasiorhinus* spp., the ability to maintain a water balance, aided by their burrow, is of critical importance in semi-arid savannas without access to free-standing water.

The burrow and wombat foraging

Among burrowing herbivores, only the Vombatidae (30 kg) and Old-world Porcupines (16 kg) achieve an adult body mass above 10 kilograms (Johnson 1997). The old-world porcupines, *Hystrix* spp., have a relatively high energy diet for a herbivore, feeding upon fruits and tubers, as do other large mammalian burrowers that are insectivores such as the giant pangolin, *Manis gigantea*, and the

aardvark, *Orycteropus afer*. The wombat can then be considered as an anomaly, being large relative to other burrowers but feeding upon grasses of poor nutritional quality.

Wombats use burrows for environmental shelter, specifically as a means to maintain thermoregulatory and water balance (Wells 1973, Wells 1978b). They require little shelter against predation as they have few natural predators, apart from humans (Flannery 1994) and dingoes (Corbett 1995). They do not require a burrow as a site for food storage and they only consume subterranean vegetative matter (e.g. roots) as an indirect consequence of burrow construction. Consequently, the physiological costs and benefits of a fossorial lifestyle are central to the ecology of wombats.

1.5 THE VOMBATIDAE - AN OVERVIEW

The Vombatidae, consist of three extant species from two genera (*Vombatus* and *Lasiorhinus*) and four extinct genera (Figure 1.1). *Vombatus ursinus*, the common wombat, is generally a forest dweller grazing in clearings and is found in eastern Australia (Figure 1.2). The hairy-nosed wombats of the genus *Lasiorhinus* are both grazers inhabiting semi-arid grasslands and savanna. The SHN wombat inhabits southern Australia, and the NHN wombat is confined to a single population in Central Queensland (Figure 1.3). All three species share the following morphological adaptations: open rooted continuously growing teeth; stocky powerful limbs for digging; short necks; broad flattened skulls; a large broad sacrum; and a dermal shield on the rump (Wells 1989). The morphological adaptations are well suited to their nocturnal, burrowing and grazing habits.

1.5.1 The ecology of wombats

1.5.1.1 Herbivorous habits

Common wombats predominantly eat grass with some dicots and roots (McIlroy 1973, Rishworth *et al.* 1995a&b). McIlroy (1973) observed the feeding behaviour of the common wombat in forests near the Australian Capital Territory. He found that the common wombat showed a preference for the leaves of grasses.

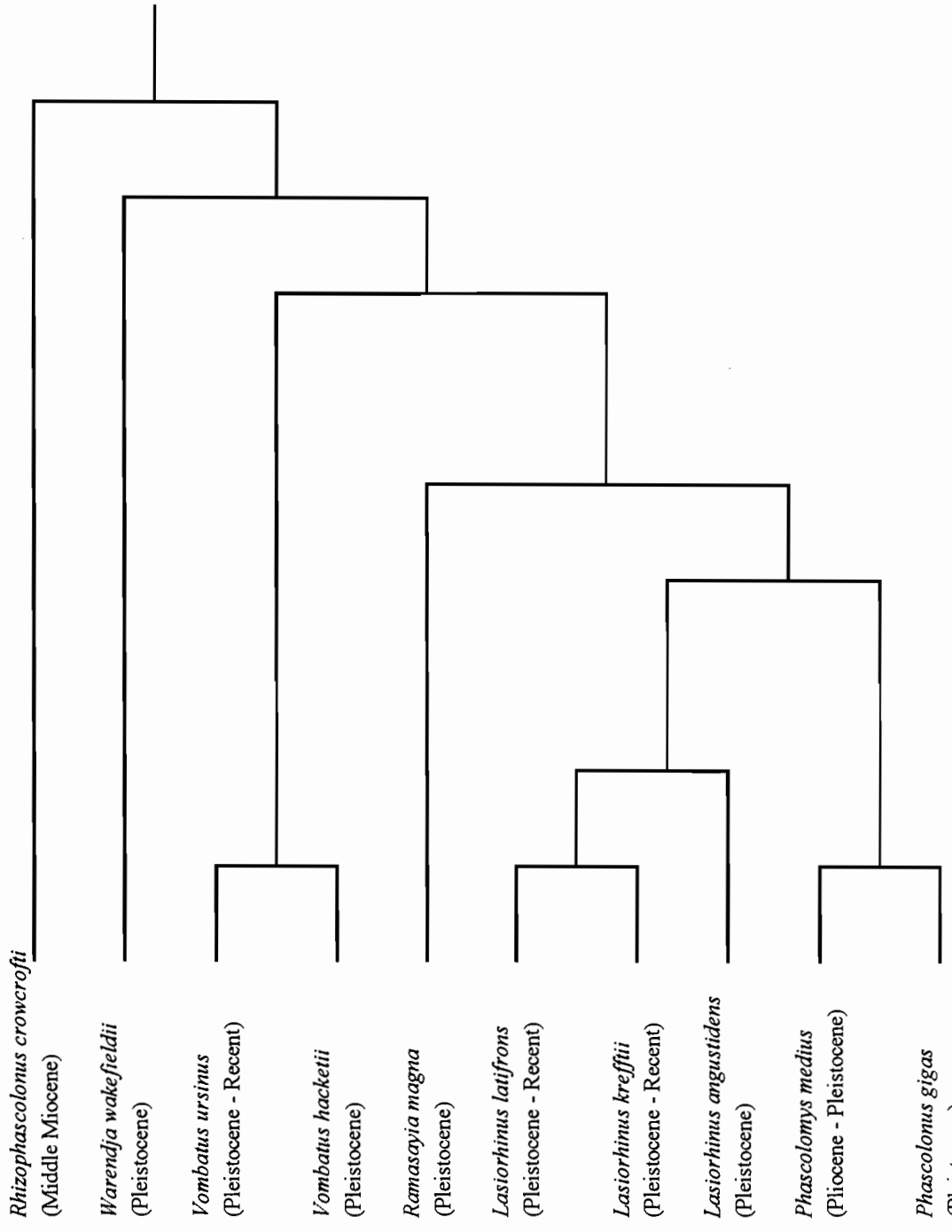


Figure 1.1. A cladogram of the Vombatidae reproduced from Archer (1984).

Figure 1.2. Current distribution of the common wombat. Figure is from Triggs (1988).

Figure 1.3. Current distribution of both the southern hairy-nosed wombat and the northern hairy-nosed wombat. Figure is from Triggs (1988). More detailed distributions of extant and extinct populations of the NHN wombat are shown in Figures 2.1 and 2.3.

However, McIllroy (1973) observed the common wombat to consume such a diverse array of food items that he considered the Common wombat to be a non-selective feeder.

The SHN wombat also has a preference for grasses, particularly *Stipa* spp. (Wells 1973, Wells 1978a, Dierenfeld 1984, Kean 1990). Direct observations of the SHN wombat suggest that the SHN wombat shows a strong preference for new shoots of *Stipa* spp. (St John and Saunders 1989). However, the habitat and forage resource of the SHN wombat is highly variable and characterised by a high frequency of drought. In response to variation in forage resources, particularly drought, the SHN wombat is flexible in its foraging decisions. Dierenfeld (1984) demonstrated that SHN wombats foraging in conditions of drought consumed chenopod shrubs when grasses were limiting. Further, Gaughwin *et al.* (1984) found that SHN wombats were able to suffer body mass losses of up to 50% before death during drought.

The diet of the NHN wombat has been investigated by Flößer (1986) and Crossman (1988). Both accounts indicate that the diet of the NHN wombat consists almost solely of grasses with a small contribution from forbs. However, both studies were brief with temporal variability and variability between individual NHN wombats not considered in their examination of the diet of the NHN wombat. Consequently, this study aims to address these issues. It also aims to describe the response of the NHN wombat to variation in quantity and quality of its food resource which was not described in the previous studies but is so important in tropical savanna ecosystems.

1.5.1.2 Adaptations to Grazing

Digestive Processes and Nutritional Adaptations of Wombats

Feeding on grass, the teeth of wombats are subjected to considerable wear from silica inclusions present in plant cells (Wells 1989). The open rooted, continuously growing teeth of wombats allow the wombat to maximise the physical breakdown of plant particles despite continual wear of the teeth (Wells 1989). This is an important adaptation, especially because hairy-nosed wombats are long-lived (*L. krefftii* can live for more than 27 years in captivity, J.Dennis pers. comm.).

Wombats forage in response to the nutritional quality of plants and may actively seek plants high in nitrogen, low in fibre and with a high digestibility (Wells 1973). To compensate for the generally low concentrations of nitrogen of forage species consumed by wombats, wombats have one of the lowest dietary nitrogen requirements known for any mammal (Barboza *et al.* 1993). Barboza *et al.* (1993) found that the daily dietary nitrogen requirements were 158 $mg.kg^{-0.75}.d^{-1}$ for the common wombat and 201 $mg.kg^{-0.75}.d^{-1}$ for the SHN wombat. Further, the capacious colon of the wombat allows the retention time of the consumed forage to be maximised. By having a long retention time, the fermentation of grasses and the potential energy gain per unit time of digestion of the forage consumed can be maximised (Dierenfeld 1984).

Water Balance and Energetic Adaptations

The low metabolic rates of wombats compared to other herbivores (Wells 1973, Wells 1978b, Barboza 1989, Barboza 1993b) results in a high water-use efficiency which, for the hairy-nosed wombats, is an important physiological characteristic for living in a semi-arid habitat. The physiological benefits of living in a burrow, as described above, combined with nocturnal foraging and short foraging bouts help keep water turnover to a minimum.

Besides the benefits gained by living in burrows, wombats are subjected to restrictions imposed by their semi-fossorial lifestyle. Wombats are dependent on their burrows for thermoregulation and are intolerant to extremes in ambient temperature (Wells 1978b). This greatly compromises time devoted to foraging and other activities which must be confined to periods when above-ground conditions are optimum (Wells 1973, Wells 1978b, Johnson 1991a&b)

1.5.2 The Northern Hairy-nosed Wombat

Until recently, little was known of the biology of the NHN wombat. The first non-fossil specimens were reported by De Vis (1900) from the Moonie River near St George in south-central Queensland, Kershaw (1909) from near Deniliquin in south-central New South Wales, and Longman (1939) from Epping Forest Station near Clermont in central Queensland. Confusion surrounded the taxonomic

status of specimens from each of the three localities which was further confounded by the demise of the former two populations. In 1983, Dawson revised the taxonomy suggesting that specimens from all three localities were the one species, *Lasiorhinus krefftii*, distinct from the SHN wombat *L. latifrons*. Taylor *et al.*(1994) genetically confirmed Dawson's (1983) taxonomy, and went on further to suggest that speciation occurred about 2 million years ago.

Conservation Status

The NHN wombat is restricted to a single population at Epping Forest National Park, in Central Queensland. At present, the population is estimated to consist of only 65 individuals (Hoyle *et al.* 1995) and has very low allelic diversity compared to *L. latifrons* (Taylor 1995). Among large mammals only the Javan Rhinoceros, *Rhinoceros sondaicus*, has the dubious honour of being more endangered with a population estimated to be only 50 to 54 individuals (Khan 1985). Historical accounts suggest that the NHN wombat was rare at the time of European settlement, but its decline was accentuated by a combination of grazing practices and drought (Crossman 1988, Johnson 1991a, Crossman *et al.* 1994).

1.6 AIMS AND APPROACH

The NHN wombat is an unusual herbivore because of its behaviour and morphology, and in particular the benefits and limitations imposed by a semi-fossorial lifestyle. It is also subjected to extremes in quality and quantity of its food resource associated with a dynamic tropical savanna habitat. This thesis therefore aims to:

1. Describe and quantify temporal variation of the savanna habitat of the NHN wombat.

2. Describe and quantify the botanical composition of the NHN wombat diet, and investigate temporal variation in diet composition.

3. Investigate potential physiological compensation mechanisms (energy storage in the form of body fat) associated with temporal variation in quality and quantity of the food resource.

4. Examine the activity and ranging behaviour of the NHN wombat.

5. Compare the diet, population dynamics and distribution of the sympatric eastern grey kangaroo to that of the NHN wombat.

Results are discussed in relation to diet choice and the consequences for the digestive strategy and foraging behaviour of wombats. The findings of each chapter are then used to describe the feeding strategy of the NHN wombat and the implications for the conservation of the NHN wombat.

CHAPTER 2

FORAGE QUANTITY AND QUALITY

2.1 INTRODUCTION

Epping Forest National Park (EFNP) in central Queensland (146°41'E and 22°21'S) is a tropical savanna, with a semi-arid climate (Steinbeck 1994). As a tropical savanna ecosystem, with irregular rainfall, the food resource of the grazing wombat is constantly changing.

This chapter aims to quantify the variation in quantity and quality of the forage available to the NHN wombat. Before quantifying changes in the quantity and quality of the forage at EFNP, it is necessary to begin with a brief description of EFNP. The chapter culminates in a synthesis of the forage environment of the NHN wombat followed by a critical review of the technique used to quantify forage quality and its application to EFNP and beyond.

2.2 CHARACTERISTICS OF HABITAT OF THE NORTHERN HAIRY-NOSED WOMBAT

2.2.1 Climate

Epping Forest National Park is within the catchment of the Belyando River (Figure 2.1). The climate of this region has been formally described by Fitzpatrick (1967) with Clermont, a regional centre (110 km south east of EFNP) providing long term climate data. Mean monthly temperatures at Clermont Post Office during this study ranged from a maximum of 37.5°C in January 1994 to a minimum of 6.4°C in July 1995. Mean bimonthly temperatures during this study followed a sinusoidal pattern (Figure 2.2), with daily temperatures being consistent around the mean maximum and minimum temperatures.

Figure 2.1. The distribution of the northern hairy-nosed wombat in Queensland from Horsup (1998), adapted from Crossman (1988). Closed circles (●) are confirmed populations of the NHN wombat, including the extant Epping Forest National Park Population and the extinct Moonie River population. Open circles (○) are unconfirmed records of the NHN wombat.

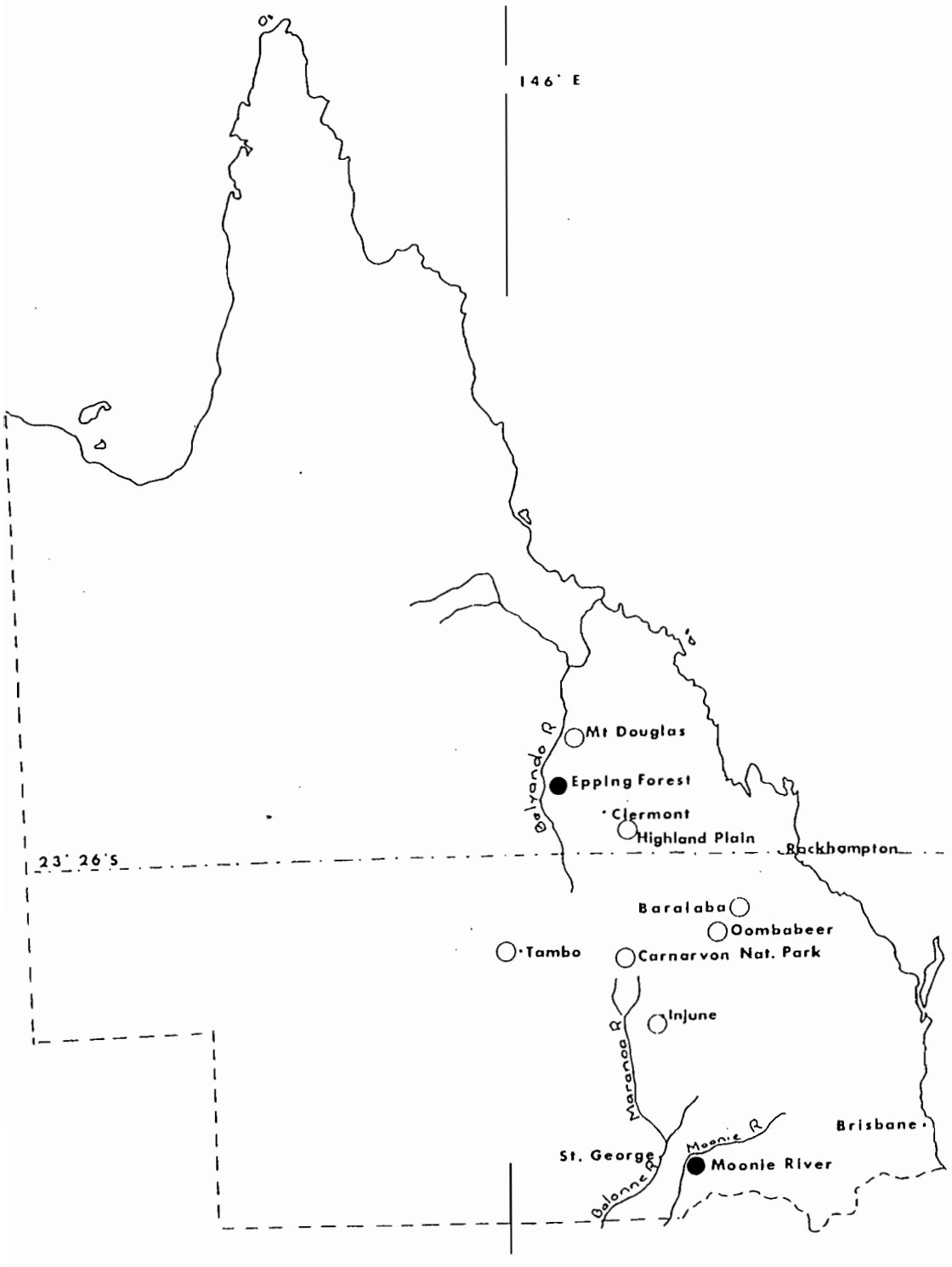


Figure 2.2. Mean bimonthly maximum and minimum temperatures during the study period. Temperatures were recorded at the Clermont Post Office located 110 kilometres south east of Epping Forest National Park.

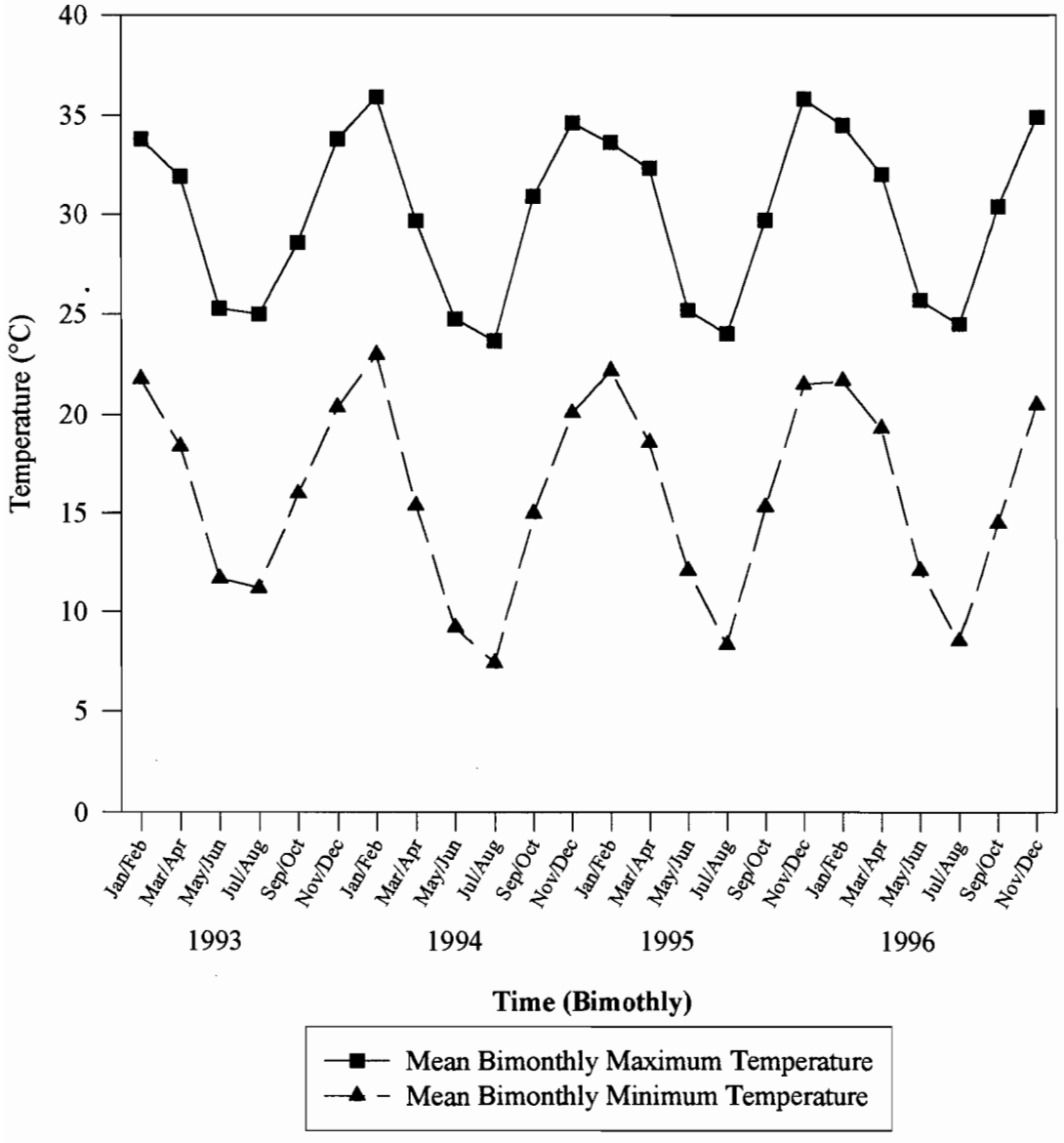


Figure 2.3. The Epping Forest region highlighting Epping Forest National Park (EFNP) and the surrounding pastoral properties including Epping Forest Holding to the north of EFNP and Waltham Holding to the south of EFNP. Areas once occupied by the NHN wombat are indicated by the approximate date they were last seen. Figure is from Horsup (1998) after Crossman (1988).

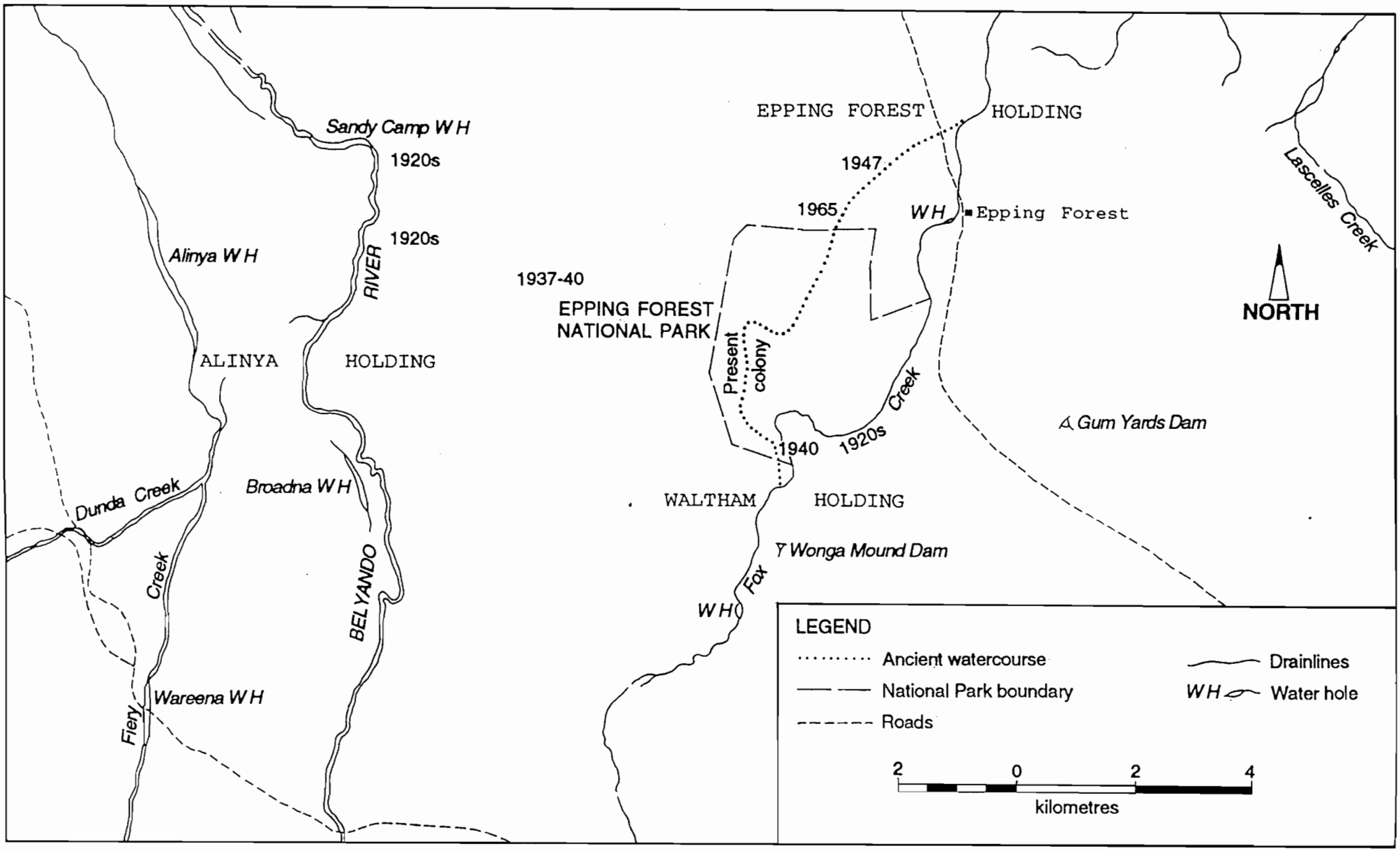
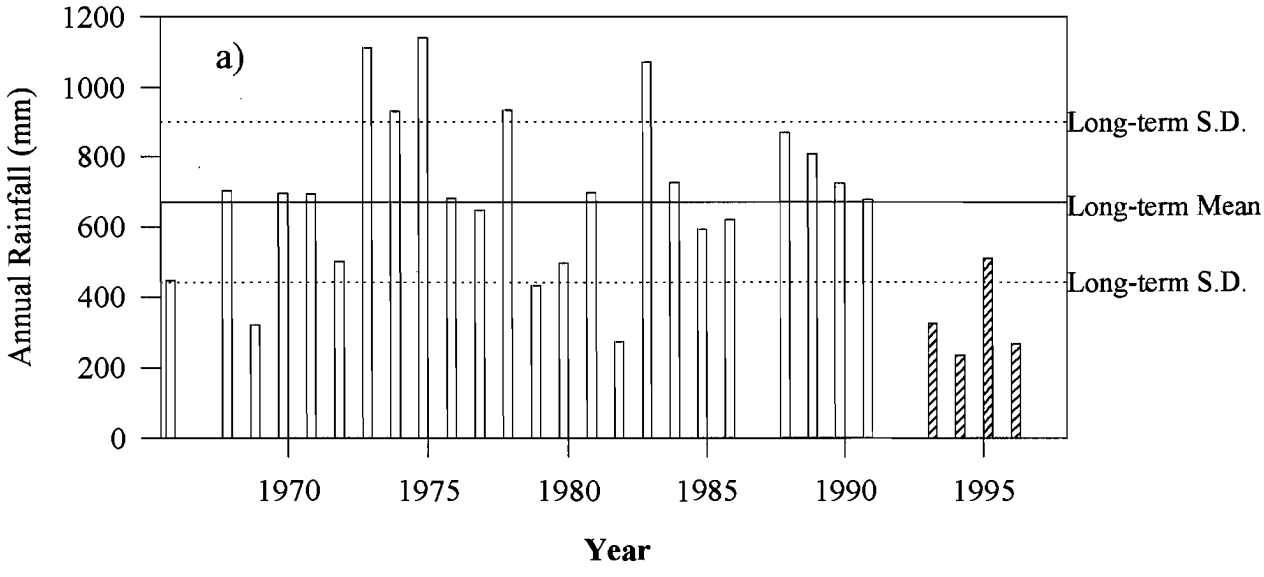
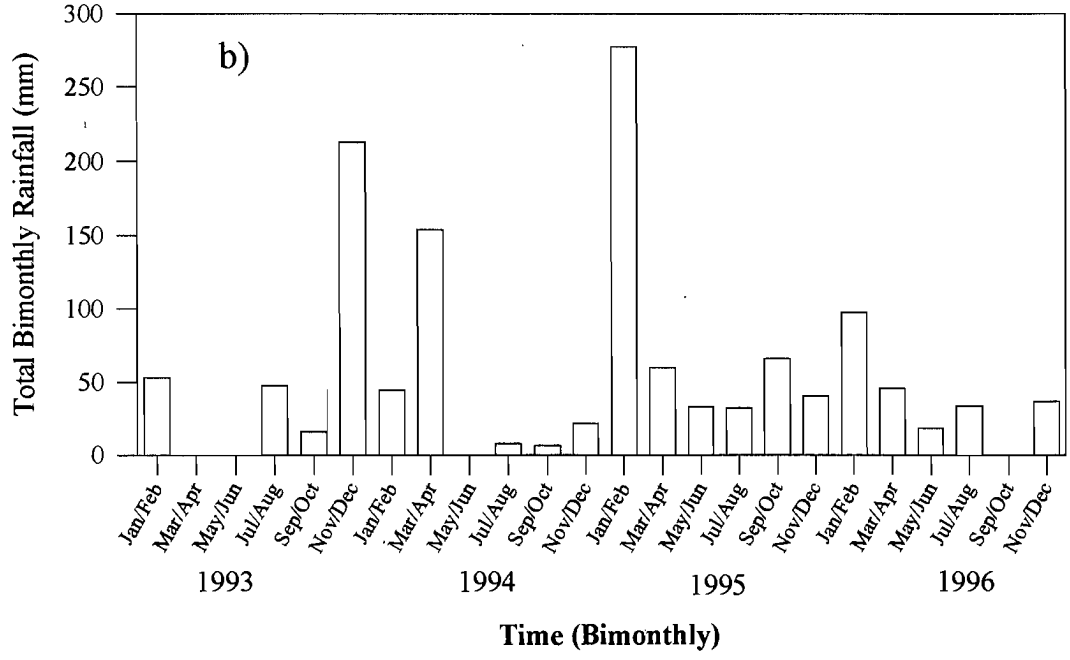


Figure 2.4. Rainfall trends at Epping Forest National Park.

a) Long-term trends in total annual rainfal collected at Clermont Post Office, 110 km south-east of Epping Forest National Park. Hatched bars are the total annual rainfall during the current study. Data was incomplete for 1967, 1987 and 1992.



b) Total bimonthy rainfall throughout study period. Local rainfall data collected at Walthum Station homestead, approximately 15 km south-east of Epping Forest National Park. Absence of data in some bimonthly periods represents no rainfall.



Annual rainfall in the Epping Forest region (defined in Figure 2.3) is highly variable (Figure 2.4a) and is characterised by a high frequency of drought. This study was conducted during four years of below average rainfall (Figure 2.4a). Bimonthly rainfall during the study was distinctly seasonal but very irregular, with the majority falling in the summer months of December, January and February (Figure 2.4b). Another characteristic of rainfall in the Epping Forest region is its spatial variability. Daily rainfall records can differ significantly between Walthum Holding and Epping Forest Holding, the two properties surrounding EFNP (shown in Figure 2.3). Likewise, differences in total rainfall were recorded between three rain gauges located throughout the range of the wombat habitat. Over a direct distance of approximately 4.5 km, the three rain gauges within wombat habitat recorded 14.25 mm, 4.8 mm and 1.5 mm of rainfall between October and December 1994. In the same period, Walthum Station homestead recorded 22.7 mm. Localised variation in rainfall was not examined during this study because rainfall is irregular and often low, and the rain gauges on EFNP are checked infrequently and often not immediately following rain. However, localised variation of rainfall may temporarily influence foraging behaviour and population distribution. This is examined in Chapter 5.

2.2.2 Soils

The distribution of wombat burrows at EFNP (Figure 2.5) is strongly associated with the deep uniform sandy soils that follow an ancient stream bed (Figure 2.6) (Steinbeck 1994). These sandy soils, often exceeding 5.5 metres in profile, provide a suitable burrowing substrate with extra structural support provided by Bauhinia trees, *Lysiphyllum hookeri,* at the entrance of many burrows (Crossman 1988, Steinbeck 1994). Other soils types on the Park are typically clays that support brigalow (*Acacia harpophylla*) and gidgee (*Acacia cambagei*) communities, but are unsuitable for wombat burrowing (Steinbeck 1994).

2.2.3 Vegetation Community

Epping Forest National Park is a small 3300 ha parcel of the brigalow, *Acacia harpophylla*, belt with six vegetation communities defined by Steinbeck (1994): grassland with trees; open patchy woodland; dense patchy woodland; open

Figure 2.5. Epping Forest National Park showing the distribution of NHN wombat burrows and seven BOTANAL transects. Each BOTANAL transect is 400 metres in length, bisecting the firebreak passing through the habitat of the NHN wombat. Map was produced from data collected by Steinbeck (1994).

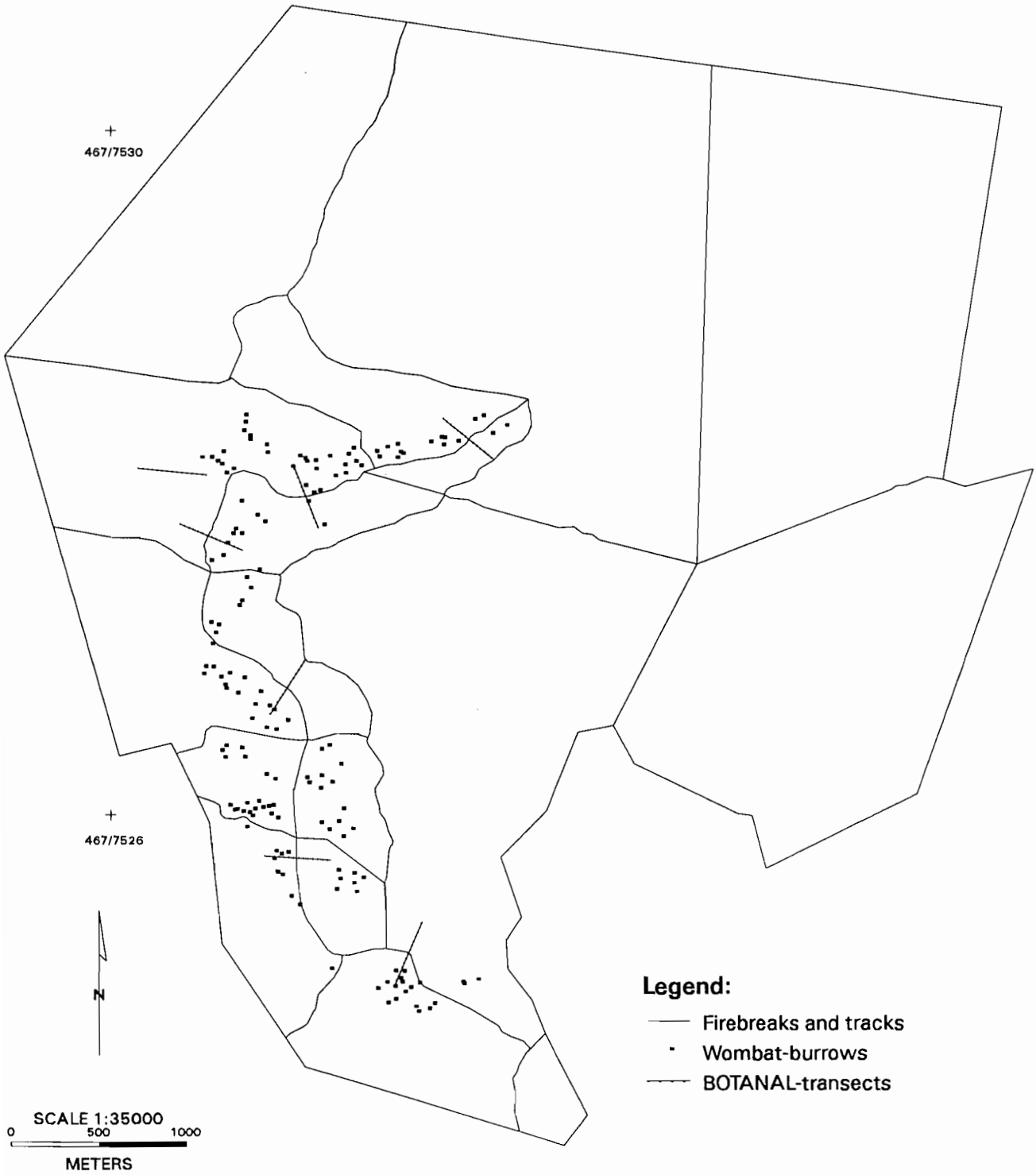


Figure 2.6. Soil map of Epping Forest National Park, highlighting the deep uniform sandy soils of the ancient water course. Map was produced from data collected by Steinbeck (1994).

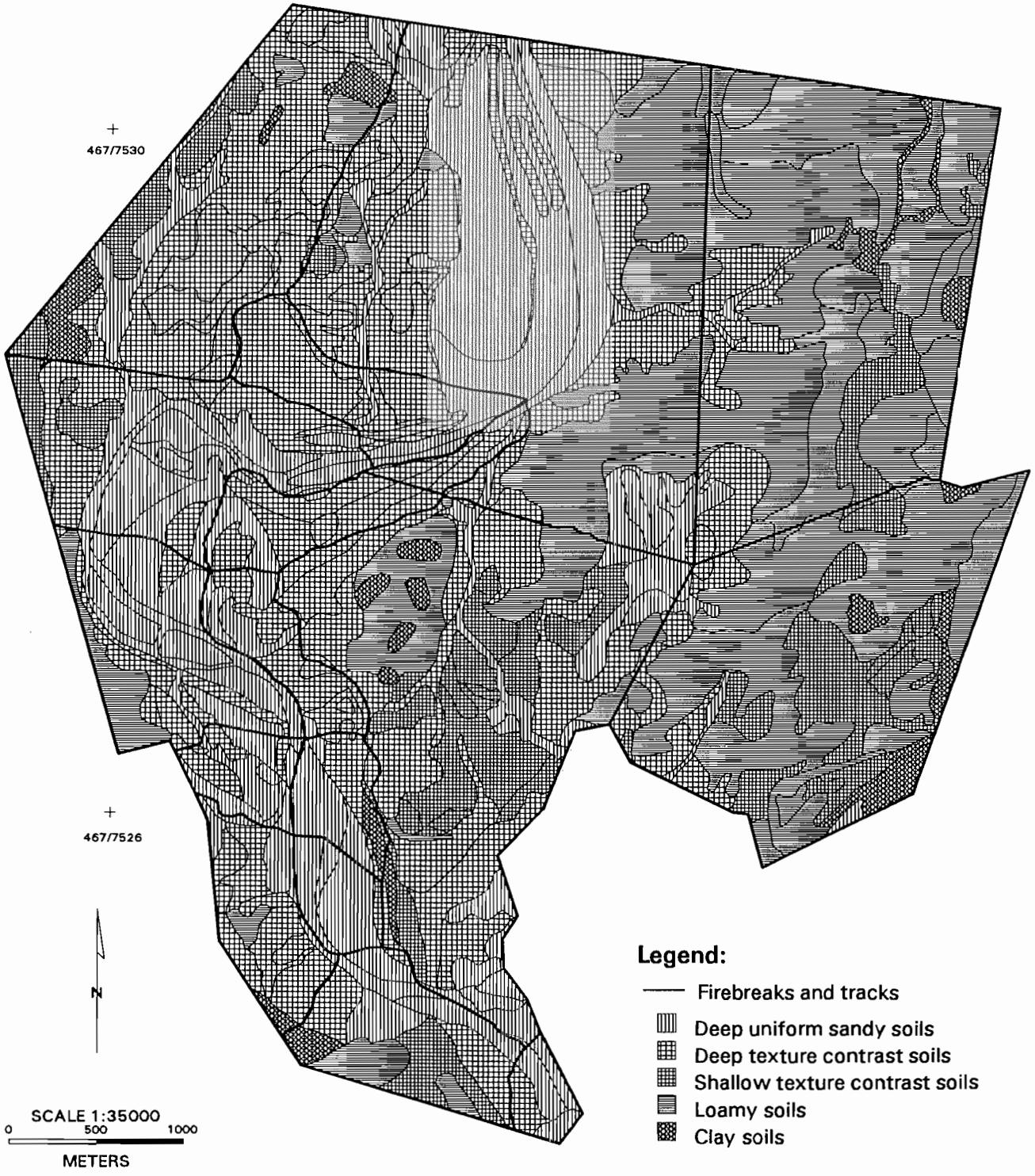
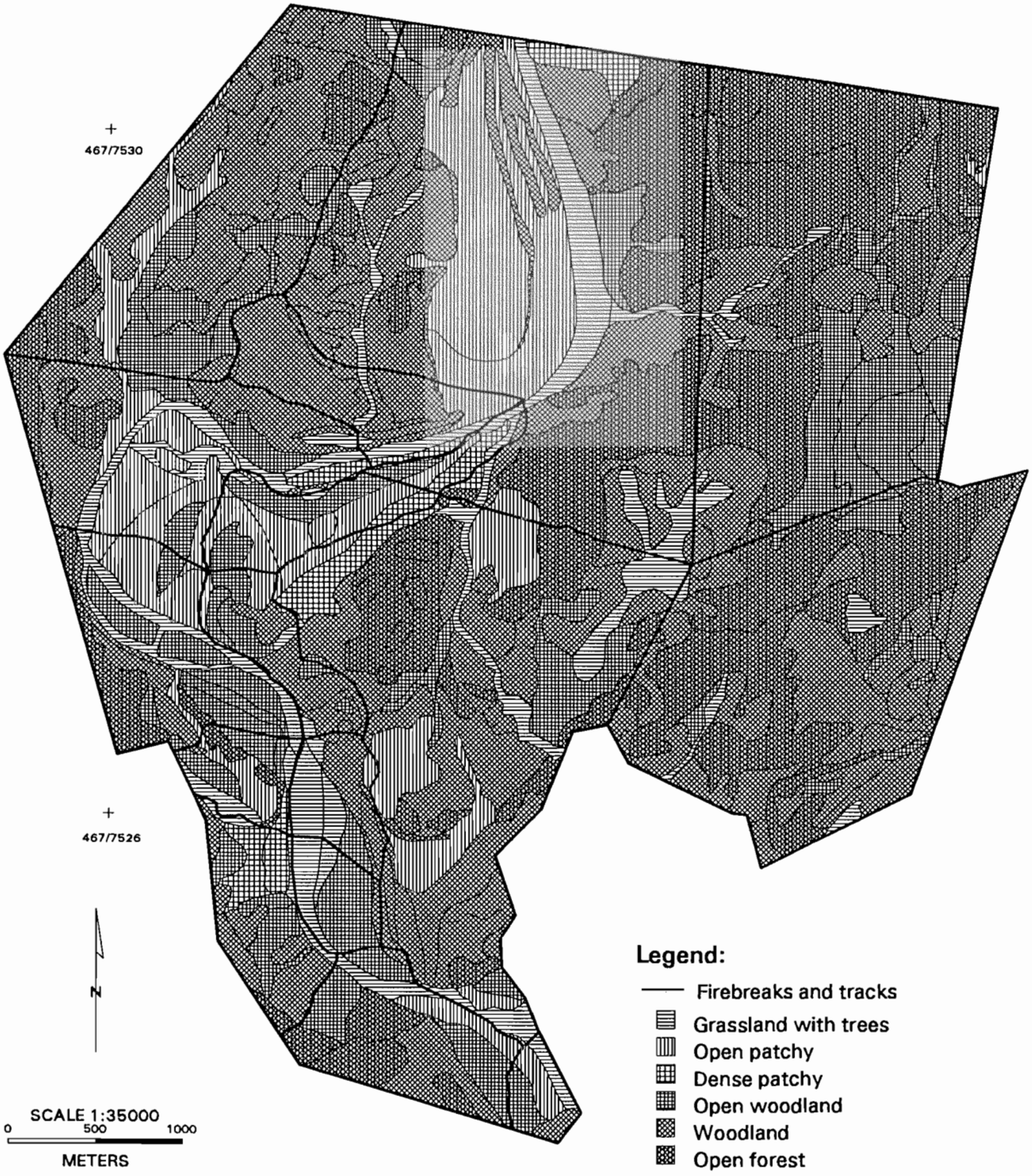


Figure 2.7. Vegetation structure of Epping Forest National Park. Map was produced from data collected by Steinbeck (1994). Vegetation classifications are described in full by Steinbeck (1994).



woodland; woodland; and open forest (Figure 2.7). Each of the vegetation communities, with their associate floristic types, are strongly associated with the different soil types (Table 2.1). Wombat burrows occur close to trees in the grassland vegetation community (Crossman 1988). Wombat feeding ranges are typically associated with grasslands with trees, open woodland, woodland, and open patchy vegetation communities where grasses dominate the understorey (Johnson 1991a).

The grassland community of the wombat habitat is dominated by the introduced *Cenchrus ciliaris* (buffel grass) and native species including *Aristida* spp. (three-awned grasses), *Enneapogon* spp. (bottle washers), *Eragostris lacunaria* (purple lovegrass), *Chrysopogon fallax* (golden beard grass) and the sedge *Fimbristylis dichotoma*. The frequency of occurrence of the plants fluctuates temporally (Figure 2.8). For example, *Heteropogon contortus* (black spear grass) had a frequency of occurrence of approximately 20% prior to the 1990's (Back 1997) but the effect of long-term drought has markedly reduced its frequency of occurrence (Figure 2.8). Recently, the introduced grass *Cenchrus ciliaris* (buffel grass) has become increasingly prominent in the grassland community, increasing from 8% to 48% in frequency from 1987 to 1997, and 13% to 54% of total biomass from 1987 to 1994 (Back 1997, Horsup 1998).

2.3 FORAGE QUANTITY

2.3.1 Introduction

2.3.1.1 Forage quantity and herbivory

Consider the foraging environment from the perspective of a wombat. Once the wombat has decided where to forage, its forage selection is dictated by the plant species it encounters, or the frequency with which it encounters different species and the composition of those species. The species composition is intrinsically related to the species frequency of the pasture and species biomass. Foraging decisions by the wombat must therefore initially consider the three primary attributes of quantity: species composition, species frequency and species biomass.

Table 2.1. Vegetation community structure and soil types at Epping Forest National Park adapted from Steinbeck (1994). Soils types are described in detail by Steinbeck (1994) using standard Northcote (1971) soil classifications.

Vegetation Structure	Floristic Types	Major Soil Types	Minor Soil Types
Grassland with Trees	Grasslands with isolated trees	Deep Uniform Sandy Soils	-
Open Patchy	Inland Bloodwood[1]/ Moreton Bay Ash[2]	Deep Uniform Sandy Soils	Deep Texture Contrast Soils
Open Patchy/Open Woodland/Woodland	Bauhinia[3]	Deep Uniform Sandy Soils	Deep Texture Contrast Soils
Open Patchy/Open Woodland/Dense Patchy	Boonaree[4]	Deep Uniform Sandy Soils/Deep Texture Contrast Soils	-
Open Patchy/Open Woodland/Woodland	Brown's Box[5]	Deep Texture Contrast Soils	-
Open Woodland	Brigalow[6]/Browns Box[7]	Shallow Texture Contrast Soils	-
Open Woodland/Open Forest	Brigalow[6]	Loamy Soils	-
Open Forest	Brigalow[6]/Gidgee[7]	Loamy Soils	-
Open Forest	Brigalow[6]/Gidgee[7]/ Coolibah[8]	Loamy Soils	-
Open Woodland/Open Forest	Brigalow[6]/ Yellowwood[9]	Loamy Soils	-
Open Forest/Open Woodland	Gidgee[7]	Loamy Soils	Clay Soils
Woodland/Open Woodland	Coolibah[8]	Loamy Soils	Shallow Texture Contrast Soils/ Clay Soils

(1) Inland Bloodwood: *Corymbia terminalis*

(2) Moreton Bay Ash: *Eucalyptus tessellaris*

(3) Bauhinia: *Lysiphyllum hookeri*

(4) Boonaree: *Heterodendrum oleifolium*

(5) Brown's Box: *Eucalyptus brownii*

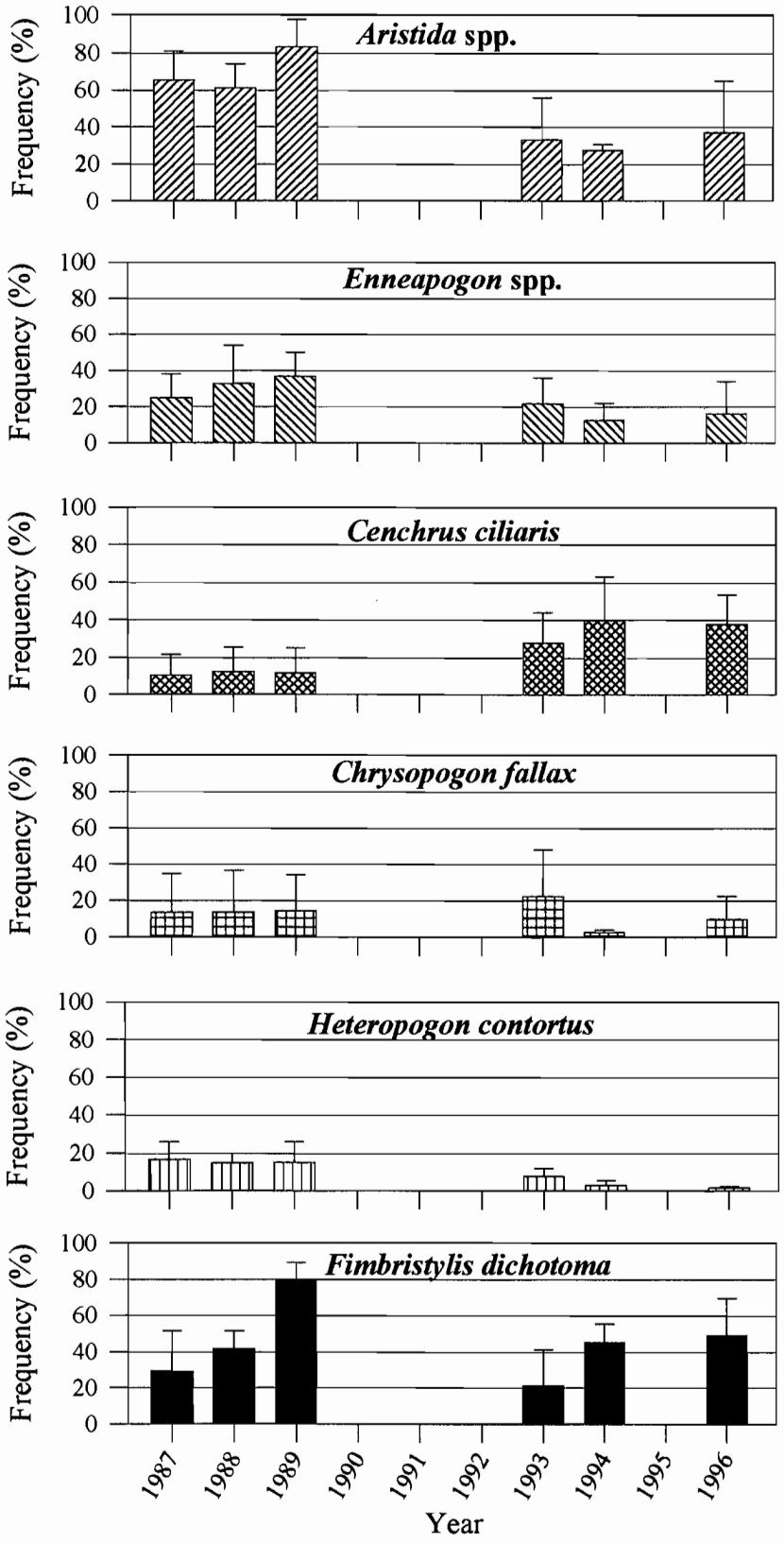
(6) Brigalow: *Acacia harpophylla*

(7) Gidgee: *Acacia cambagei*

(8) Coolibah: *Eucalyptus microthera*

(9) Yellowwood: *Terminalia oblongata*

Figure 2.8. Long-term frequency of occurrence (as a percentage ± S.D.) of dominant forage species at Epping Forest National Park using BOTANAL methodology. Data were not collected in the years 1990-1992 and 1995. Data for the figure is from a study measuring long-term trends of species composition and frequency by Back (1997).



The grazing environment of the NHN wombat is heterogeneous (Steinbeck 1994). Therefore, forage species consumed by the NHN wombat may be dependent upon other factors besides the aforementioned attributes of forage quantity. Specifically, forage selection by the NHN wombat may be influenced by the nutritional quality of the forage species (Kohlmann and Risenhoover 1994). However, in a savanna environment, species composition, frequency and biomass can greatly influence foraging decisions of herbivores (McNaughton and Banyikwa 1995). In addition, primary productivity, which is strongly associated with total rainfall in tropical savannas (McNaughton and Banyikwa 1995), can also influence foraging decisions by herbivores.

In order to quantify attributes of forage quantity it is necessary to begin with a brief review of the merits of the techniques used to describe forage quantity, specifically focussing upon the application of the BOTANAL method.

2.3.1.2 Forage Quantity: Applications and Techniques

There are three common measures of forage quantity: species frequency, species composition and species biomass. Each attribute is usually defined in terms of a given area (e.g. a quadrat). Species frequency is defined as the "percentage of quadrats where the species (or other attribute) is present" (Tothill *et al.* 1992). Species composition or botanical composition is defined as "the percentage a plant species or group contributes to the total biomass" (Tothill *et al.* 1992). Species biomass describes the dry matter yield of each plant species within a pasture.

There are two classes of methods of estimating species frequency, composition and biomass in grazing systems - destructive and non-destructive techniques. Application of the appropriate technique depends on the accuracy required, cost and scale of measurements (Catchpole and Wheeler 1992). The accuracy of non-destructive sampling is less than destructive sampling, but at reduced cost and greater scale of application (Catchpole and Wheeler 1992). In addition, a non-destructive technique reduces the potential detrimental impact on the grazing herbivore's food resource.

BOTANAL - Measuring the Botanical Composition of Grazed Pastures

BOTANAL is a non-destructive technique and data analysis package designed for measuring attributes of forage quantity important in grazing systems (Tothill *et al.* 1992). Users of the BOTANAL technique can measure biomass and botanical composition of pastures, with potential for measurements of species frequency, ground cover and other non-botanical attributes such as the presence or absence of faecal pellets deposited by herbivores. The major advantage of the BOTANAL technique is that it allows attributes of forage quantity to be estimated with high precision over a relatively large area.

BOTANAL operates on a comparative yield method, following the dry-weight-rank method of Mannetje and Haydock (1963). Briefly, the dry weight method ranks species on the basis of a visual estimate of dry weight. Within a quadrat of defined size, the species ranked one is the species with the greatest estimated dry weight or yield (Evans 1992). Comparisons are made, via an algorithm, with a range of reference standards of known dry weight that are destructively sampled. The BOTANAL analysis algorithm calculates yield and botanical composition using the assumption that the plant species occupying the first three ranks in any given quadrat are given the values of 8.04 for rank 1, 2.41 for rank 2, and 1.0 for rank 3. By summing the total rank values for each plant species in all quadrats of a given experimental area, the yield or botanical composition can be expressed as a percentage of the total scores of all plant species (Tothill *et al.* 1992). This calculation algorithm essentially follows 70, 21, and 9 percent distribution of the first three ranks. However, by allocating a value for each of the first three ranks (e.g. 8.04 for rank 1) rather than a percentage (e.g. 70% for rank 1), plant species that contribute greater than 70% of the yield will not be underestimated.

The method makes three basic assumptions (from Tothill *et al.* 1992):

1. The majority of sample quadrats contain at least three species.
2. The species order varies between quadrats.
3. Each plant species has an equal probability of contributing to yield with no single species dominating yield because of its morphology.

Two additional assumptions were also used in this study to interpret BOTANAL analyses in terms of forage quantity available to the NHN wombat:

4. That all species recorded by BOTANAL were equally available to the NHN wombat for consumption.
5. The NHN wombat eats only above-ground plant material.

Validity of Assumptions: Specifically for Epping Forest National Park

Using the set of multipliers and rank scores of Tothill *et al.* (1992), when sample sizes are small, the BOTANAL analysis algorithm tends to overestimate the rare plant species and underestimate the common plant species (Tothill *et al.* 1992). Therefore, to increase the precision of the BOTANAL technique, a minimum sample size per experimental unit is required (estimated to be 30 quadrats/experimental unit), with precision increasing as sample size increases (Tothill *et al.* 1992).

Assumption 1 is dependent on the sampling intensity. Sampling intensity is dependent on the size of the quadrat used in sampling, and to a lesser extent observer training, the number of observers used, and observer intensity. The size of the quadrat used in BOTANAL sampling should be large enough so that at least three species will be present in the majority of quadrats, thereby allowing use of the BOTANAL algorithm (Tothill *et al.* 1992). The size of the quadrat is therefore influenced by the vegetation type, with sparse vegetation communities requiring larger quadrats (Tothill *et al.* 1992). However, other considerations influence selection of quadrat size. Williams (1971) suggested that quadrat size should be as large as possible, preferably 1 m^2, so that each quadrat would contain the maximum potential information. Haydock and Shaw (1975) suggested a smaller quadrat size of 0.25 m^2 was preferable as it represents the maximum quadrat size for the minimum time invested in scanning the quadrat for BOTANAL measurements. A compromise of 0.5 m^2, used in this study, would therefore be optimum: large enough to encounter an average of more than three species per quadrat, yet small enough to minimise time required to record every species within the quadrat.

Assumptions 2 and 3 are dependent on the heterogeneity of the habitat being examined. In general, the habitat of Epping Forest National Park is heterogeneous (Steinbeck 1994) making assumption 2 valid. Further, the morphologies of native grass and sedge species are similar, validating assumption 3. However, individual *Cenchrus ciliaris* plants are comparatively larger in yield than native grasses species (personal observation). Therefore, the validity of assumption 3 is dependent on the contribution of *C. ciliaris* to the total biomass. Assumptions 4 and 5 are both valid for the NHN wombat to a limited degree. Some plant species have a higher frequency of occurrence than others. Also, the frequency of occurrence of some plant species may vary with time (Figure 2.8) which in turn will alter the availability of forage species available to the NHN wombat.

BOTANAL and Epping Forest National Park

The BOTANAL technique is well suited for an application at Epping Forest National Park. A broad-scale sampling regime can easily cover the entire range of the NHN wombat with acceptable precision. Further, since BOTANAL is essentially a non-destructive technique, the impact on the forage resource of the NHN wombat is negligible.

2.3.2 Methods

2.3.2.1 BOTANAL

Preliminary BOTANAL measurements began in July 1993 with comprehensive bimonthly measurements conducted from October 1993 to June 1996. A random design of seven transects was laid across currently occupied wombat habitat. Each transect was 400 metres long and located along the central fire break bisecting the NHN wombat habitat with each transect separated by one kilometre (Figure 2.5). Each transect passed through a vegetation ecotone but was primarily in the vegetation community and soil type associated with NHN wombat habitat.

Eighty quadrats (0.5 m^2) were placed along each transect allowing statistically robust comparisons to be made between transects and eliminating the

potential for overestimating rarer species (Tothill *et al.* 1992). Within each transect the following variables were recorded:

1. Species. Species were ranked from the most to least abundant using the set of multipliers and rank scores of Tothill *et al.* (1992). Also, ranks could be tied if the contribution of one plant species in the quadrat was equal to another. Using the BOTANAL analysis program, the species data provided an estimate of species composition and species frequency. A total of 55 'species' categories were recorded by BOTANAL, although some 'species' records were actually genera (e.g. *Enneapogon* spp.), families (e.g. Malvaceae) or broad groups (e.g. Other Grasses, Other Forbs or Other Legumes).

2. Yield. A measure of yield from 0 to 20, with zero representing bare ground and 20 representing the maximum potential biomass encountered at the time of sampling. Yield was calibrated with an average of nine 'standards' (cut quadrat) per BOTANAL collection (nine standards = one set of standards). Each standard was scored across the range of the yield, cut and dried at 60°C for 48 hours and weighed. Dry weights of the set of standards were then analysed by least squares linear regression, with the regression equation predicting an estimate of dry matter biomass of each yield score from 0 to 20. Least-squares linear regression of the set of standards consistently explained more than 90% of the variation for each set of standards during the study. The slope and intercept values of the regression equation for each set of standards were then used in the BOTANAL algorithms for species biomass, composition and frequency calculations.

3. Ground Cover. An estimate of total percentage ground cover was allocated to each of the quadrats.

4. Additional Attributes. Six additional attributes were recorded in the collection of BOTANAL data primarily to describe spatial distribution of the three dominant herbivores - wombats, macropods and termites.

- *Faecal Pellet Distribution - within quadrats*. The number of wombat and macropod faecal pellets was counted within each quadrat.

- *Termite Activity - within quadrats.* A score of 0, 1, 2 or 3 was allocated to above ground activity by termites within a quadrat. Above ground termite activity was indicated by surface tunnel construction and clear signs of termite grazing. A score of zero represented no termite activity, a score of one or two represented moderate levels of termite activity, with a score of three represented extensive termite activity.

- *Faecal Pellet Distribution - between quadrats.* A score of one or zero was allocated to the presence or absence respectively of wombat and/or macropod faecal pellets in the five metre interval between quadrats.

- *Termite Activity - between quadrats.* Likewise, a score of zero or one was allocated to presence or absence of evidence of termite activity between quadrats.

The spatial relationship between wombat and macropod scats is discussed in Chapter 6. The level of termite activity above-ground was considered low. For example, the average occurrence of termite activity was $1.80 \pm 2.33\%$, that is for every 400 metres transect, evidence of termite activity occurred in an interval of only 7.2 metres. The majority of above-ground activity was low to moderate (scored 1 or 2). Consequently, the above-ground activity of termites was not examined further.

One-way ANOVA was used to examine any differences in species biomass, species frequency, species composition and ground cover with time or between the seven transects. To best illustrate bimonthly trends of BOTANAL attributes, data were pooled into five groups: Native Grasses, Sedges, the introduced *Cenchrus ciliaris*, Forbs and Malvaceae. The direct influence of rainfall and the long-term consequences of rainfall on the attributes of forage quantity, measured by BOTANAL, was determined by correlation analysis. The effect of time was examined by calculating the correlation coefficient for the attribute at time t and the rainfall at $t-1$ in a time-series.

2.3.2.2 Primary Productivity

An experiment was conducted to quantify primary productivity. It attempted to incorporate spatial and temporal variation, while controlling for potential biomass elimination through grazing pressure.

Primary production was monitored at five NHN wombat burrows. Burrows were selected on the criterion of a high rate of NHN wombat occupancy (determined by burrow monitoring, D. Crossman, C. Johnson and A. Horsup unpublished data). The area surrounding selected burrows also shared similar forage characteristics (species composition, ground cover and biomass). Three vegetation characteristics were targeted and were considered typical characteristics of the vegetation community in the vicinity of a NHN wombat burrow. These included:

- the grasses that surround a wombat burrow. These are typically characterised by a dominance of *C. ciliaris*;
- native grass and forb communities (approximately 15 metres from the burrow); and
- free growing *C. ciliaris* not associated with disturbed soil of a burrow (approximately 30 metres from the burrow).

For each of the three vegetation characters, a mesh fence exclosure measuring 1 m x 1 m excluded all vertebrate herbivores from grazing new growth. Each fenced 'experimental' plot was matched with an unfenced 'control' of the same dimensions, one metre from the fenced plot, that allowed grazing by all vertebrate herbivores.

Primary productivity was measured bimonthly at each of the control and experimental plots. Analysis of variance was used to examine the relationship between primary productivity and rainfall, spatial variation (within and between sites), and potential influence of vertebrate grazing on primary productivity. The relationship between rainfall and primary productivity was also examined by correlation analysis.

2.3.3 Results

2.3.3.1 BOTANAL Attributes

Temporal and Spatial Characteristics

The total number of plant groups recorded by the BOTANAL technique was positively correlated with rainfall (r = +0.17) but not significantly [$r_{(p \leq 0.05)}$ = 0.811; 4 df]. Generally, the number of plant groups recorded followed a seasonal pattern, with highest diversity associated with the months of high rainfall (Figure 2.9). This trend however may have been influenced by observer experience and sampling intensity. As experience increases, the time devoted to scoring each transect decreases and rare species may be over-looked or the converse may occur.

Percentage ground cover followed a seasonal trend, with highest levels occurring during the months with high rainfall (e.g. March/April 1994, January/February 1995, January/February 1996 Figure 2.10). However, ground cover was not significantly correlated with rainfall or with the rainfall time-series (Table 2.2). Ground cover did however vary with time and between BOTANAL transects (Table 2.3).

Seasonal trends in species biomass (Figure 2.11), species composition (Figure 2.12) and frequency (Figure 2.13) of selected forage groups were less discernible. The lack of clear seasonal trends of species biomass, frequency and composition (Figures 2.11 to 2.13) may be a result of the differing response of each plant species to rainfall but undetected in the correlation analysis or, more likely, an artefact of the BOTANAL technique. The dry-weight technique may be the prime source of error, particularly because of the heterogeneous nature of the habitat and the dominance of *C. ciliaris* in its contribution to the species composition and biomass. Some species such as *Fimbristylis* spp. do exhibit strong seasonal patterns and were influenced, but not statistically significant, by rainfall (Table 2.2).

The BOTANAL technique focuses upon species biomass and species composition. However, the frequency of occurrence is more likely to be a true reflection of what the NHN wombat encounters while foraging if it feeds at random. Some species, such as *Fimbristylis dichotoma*, have relatively high

Figure 2.9. Number of plant groups detected by the BOTANAL technique. Plant groups includes plant species, plant genera (see text for details). Data from March/April 1996 were not collected.

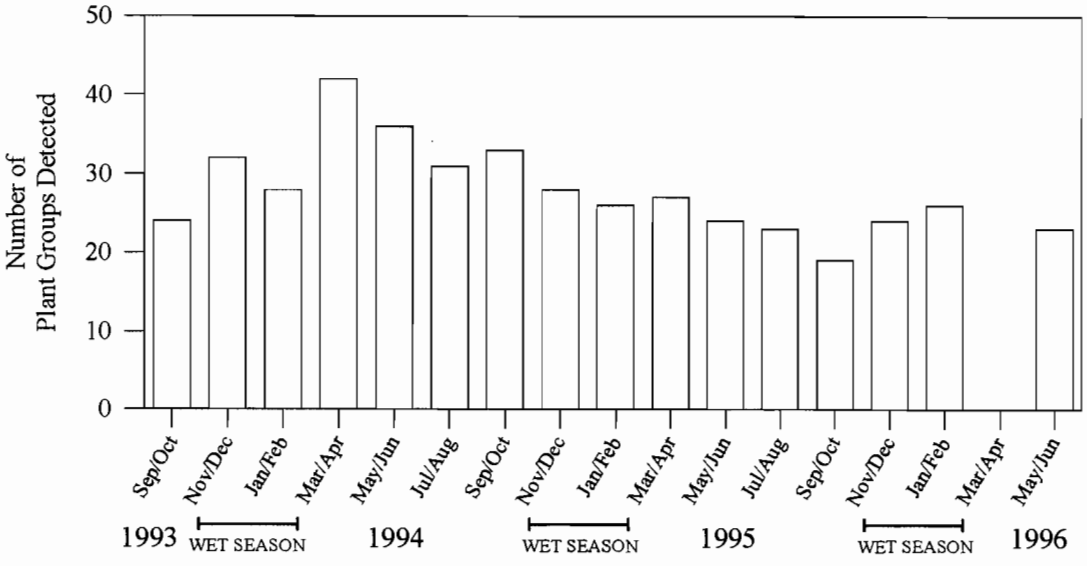


Figure 2.10. Bimonthly trends in total ground cover(percentage). Percentage ground cover expressed as a mean of the seven BOTANAL transects ± one standard deviation. Data from March/April 1996 were not collected..

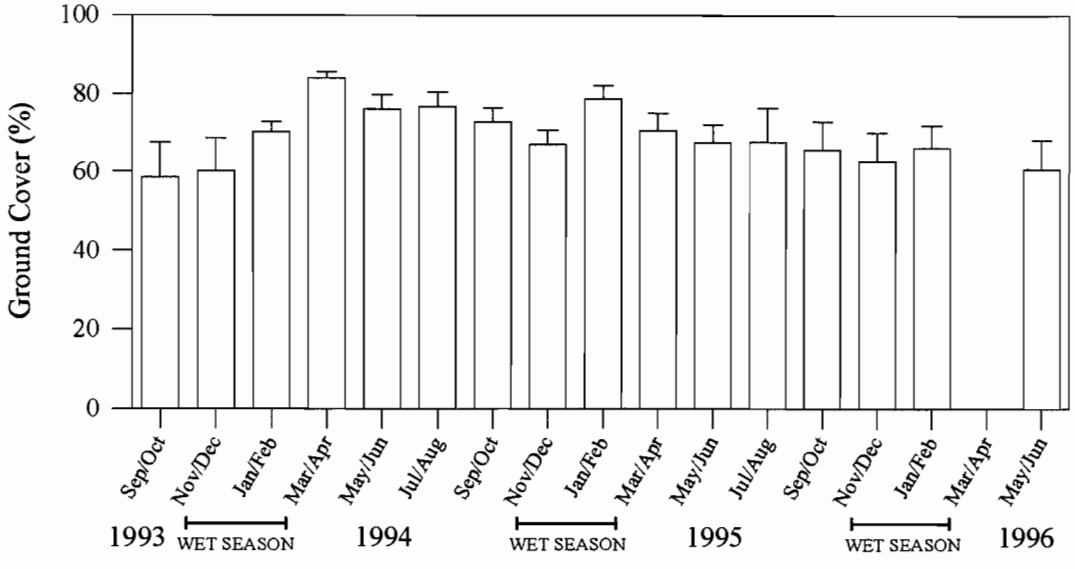


Table 2.2. Correlation coefficients (r) of BOTANAL attributes with rainfall, and the long-term effect of rainfall associated with bimonthly time lags. Significant correlations with rainfall are highlighted in bold underlined text ($r = 0.81$; $p \leq 0.05$; 4 df).

BOTANAL Attribute	Plant Species	TIME LAG (MONTHS)					
		0	2	4	6	8	10
Ground Cover		0.24	-0.49	-0.22	0.03	0.35	0.65
Species Composition	*Cenchrus ciliaris*	-0.01	0.14	0.25	-0.16	-0.25	-0.63
	Chrysopogon fallax	-0.32	0.01	0.12	0.33	0.27	-0.25
	Enneapogon spp.	-0.19	-0.05	-0.29	0.35	0.09	0.51
	Aristida spp.	-0.41	0.02	-0.12	0.39	0.14	0.36
	Heteropogon contortus	-0.50	0.47	0.07	0.16	-0.21	0.20
	Fimbristylis dichotoma	0.73	0.38	-0.18	-0.17	-0.26	-0.17
	Brachiaria spp.	0.04	-0.26	-0.35	0.07	0.46	0.59
	Salsola kalii	-0.12	-0.10	-0.04	0.23	0.03	0.42
	Malvaceae	0.09	0.32	-0.18	0.10	-0.10	0.21
	Waltheria indica	0.19	-0.38	-0.22	0.15	0.51	0.59
	Total Monocots	-0.30	0.17	0.24	0.08	-0.05	**-0.81**
	Total Dicots	0.29	-0.16	-0.24	-0.08	0.05	**0.81**
Species Frequency	*Cenchrus ciliaris*	0.35	-0.53	-0.25	-0.17	0.17	0.64
	Chrysopogon fallax	-0.50	-0.06	0.13	0.28	0.24	-0.29
	Enneapogon spp.	0.04	-0.12	0.40	0.14	-0.17	-0.20
	Aristida spp.	-0.20	0.17	0.06	0.17	0.01	-0.22
	Heteropogon contortus	-0.62	0.13	0.31	0.32	0.04	0.09
	Fimbristylis dichotoma	0.21	-0.14	-0.23	0.14	-0.06	-0.26
	Brachiaria spp.	-0.03	-0.41	-0.16	0.17	0.62	0.31
	Salsola kalii	-0.31	0.06	0.11	0.28	-0.03	-0.07
	Malvaceae	-0.47	0.15	0.03	0.29	0.38	-0.13
	Waltheria indica	0.16	-0.49	0.04	0.09	0.55	0.48
Species Biomass	*Cenchrus ciliaris*	0.05	-0.41	0.07	-0.17	0.41	0.27
	Chrysopogon fallax	-0.25	-0.04	0.23	0.33	0.33	-0.23
	Enneapogon spp.	0.03	-0.31	-0.23	0.21	0.26	0.58
	Aristida spp.	-0.22	-0.22	-0.07	0.18	0.27	0.55
	Heteropogon contortus	-0.47	0.18	0.20	0.01	-0.02	0.35
	Fimbristylis dichotoma	0.72	0.07	-0.24	-0.21	-0.01	0.12
	Brachiaria spp.	0.10	-0.32	-0.31	0.05	0.50	0.61
	Salsola kalii	-0.14	-0.24	-0.03	0.18	0.18	0.52
	Malvaceae	0.24	0.08	-0.12	0.07	-0.03	0.29
	Waltheria indica	0.24	-0.43	-0.21	0.08	0.54	0.58
	Total Biomass	0.08	-0.39	-0.03	-0.13	0.41	0.41
Primary Productivity		**0.97**	-0.08	-0.29	-0.24	0.25	-0.02

Table 2.3. One-way ANOVA describing temporal variation between collection times and spatial variation between BOTANAL transects for BOTANAL attributes and forage species potentially consumed by wombats. Significance levels: * $p \geq 0.05$; ** $p \geq 0.01$; *** $p \geq 0.001$.

BOTANAL Attribute	Plant Type	Temporal Variation		Spatial Variation	
		$F_{16,101}$	Significance	$F_{6,111}$	Significance
Species Composition	*Cenchrus ciliaris*	5.107	***	9.469	***
	Chrysopogon fallax	3.789	***	8.179	***
	Enneapogon spp.	0.874	N.S.	30.821	***
	Aristida spp.	0.647	N.S.	25.246	***
	Heteropogon contortus	1.070	N.S.	6.742	***
	Fimbristylis dichotoma	1.011	N.S.	25.546	***
	Brachiaria spp.	7.759	***	2.362	*
	Salsola kalii	3.206	***	4.771	***
	Malvaceae	1.774	*	12.539	***
	Waltheria indica	2.199	**	9.126	***
	Total Monocots	9.349	***	3.547	**
	Total Dicots	9.387	***	3.549	**
Species Frequency	*Cenchrus ciliaris*	0.350	N.S.	65.558	***
	Chrysopogon fallax	2.675	**	16.630	***
	Enneapogon spp.	0.965	N.S.	40.311	***
	Aristida spp.	1.481	N.S.	18.788	***
	Heteropogon contortus	1.116	N.S.	11.269	***
	Fimbristylis dichotoma	0.375	N.S.	92.672	***
	Brachiaria spp.	14.641	***	1.676	N.S.
	Salsola kalii	2.250	**	6.824	***
	Malvaceae	2.124	**	9.235	***
	Waltheria indica	1.490	N.S.	25.994	***
Species Biomass	*Cenchrus ciliaris*	6.912	***	8.708	***
	Chrysopogon fallax	3.101	***	8.323	***
	Enneapogon spp.	1.502	N.S.	23.081	***
	Aristida spp.	1.256	N.S.	15.138	***
	Heteropogon contortus	1.145	N.S.	8.614	***
	Fimbristylis dichotoma	2.222	**	10.865	***
	Brachiaria spp.	10.505	***	1.466	***
	Salsola kalii	3.931	***	3.031	**
	Malvaceae	2.339	**	10.771	***
	Waltheria indica	2.174	*	8.149	***
	Total Monocots	9.431	***	6.565	***
	Total Dicots	11.309	***	2.819	***
Ground Cover		10.507	***	4.856	***

Figure 2.11. Bimonthly biomass (kg.ha^{-1} dry matter) of five plant groups. Plant groupings expressed as a mean of the seven BOTANAL transects ± one standard deviation. Data for March/April 1996 were not collected. Note different scales.

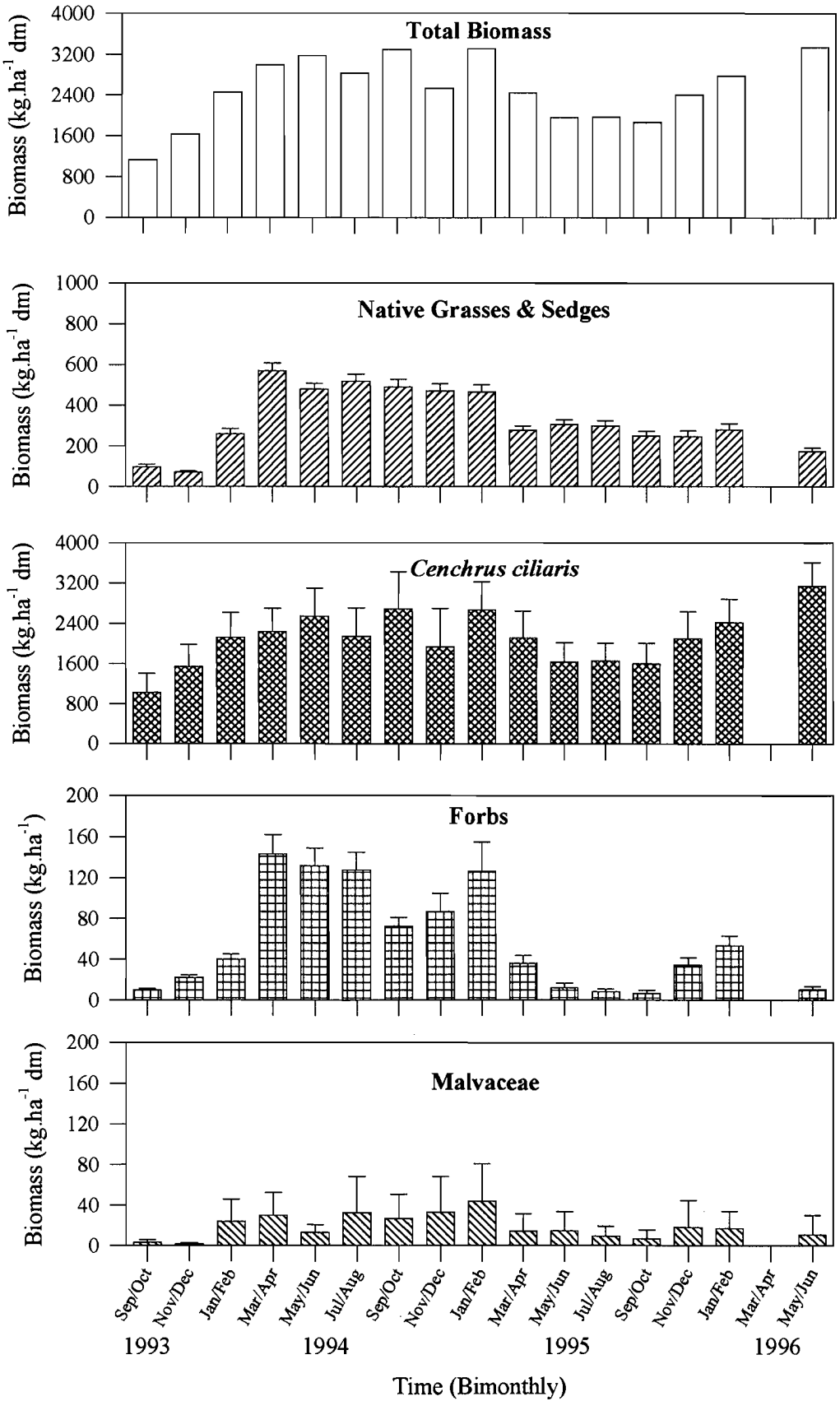


Figure 2.12. Bimonthly species composition for five plant groups. Plant groupings expressed as a mean of the seven BOTANAL transects ± one standard deviation. Data for March/April 1996 were not collected.

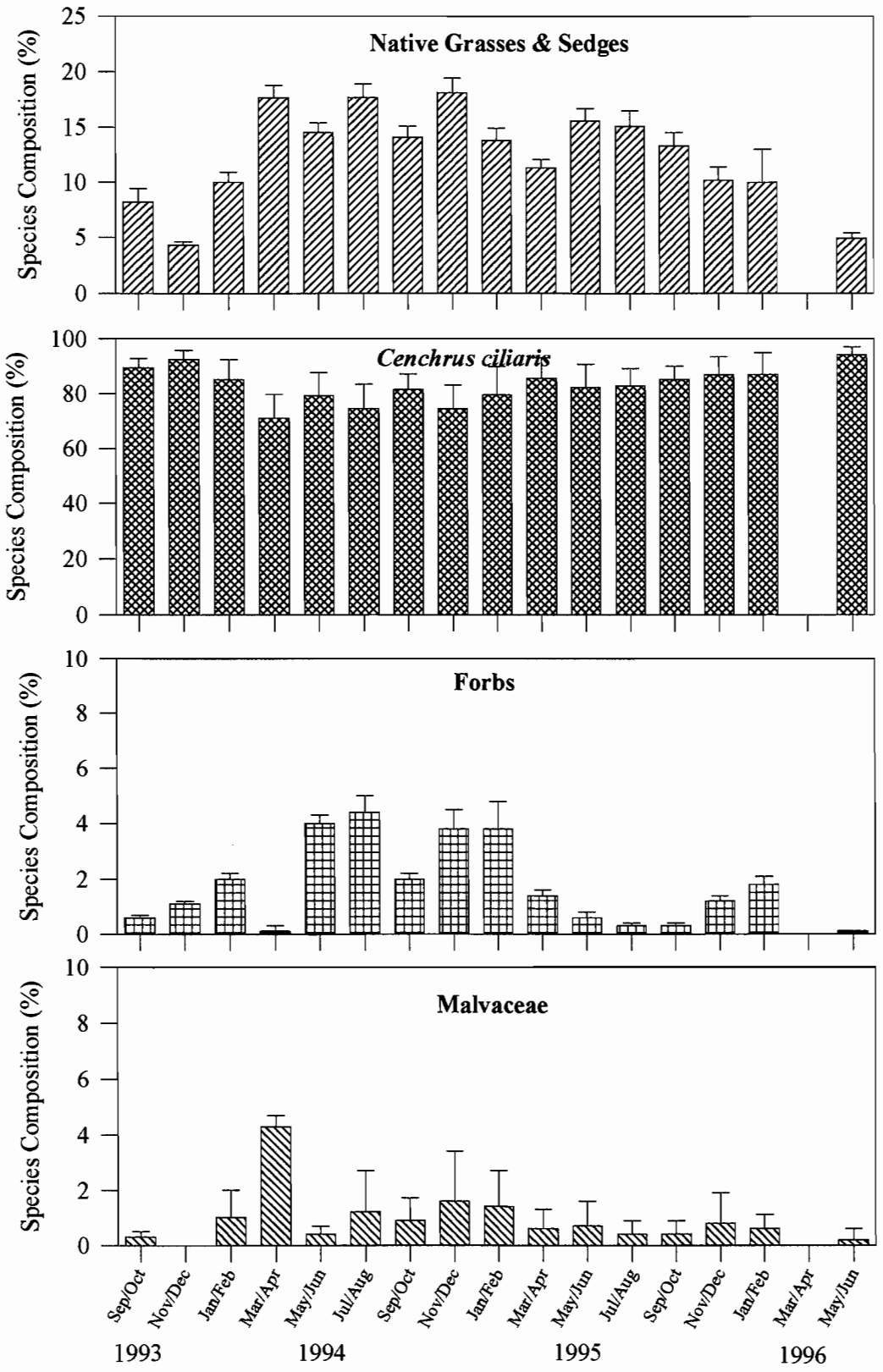
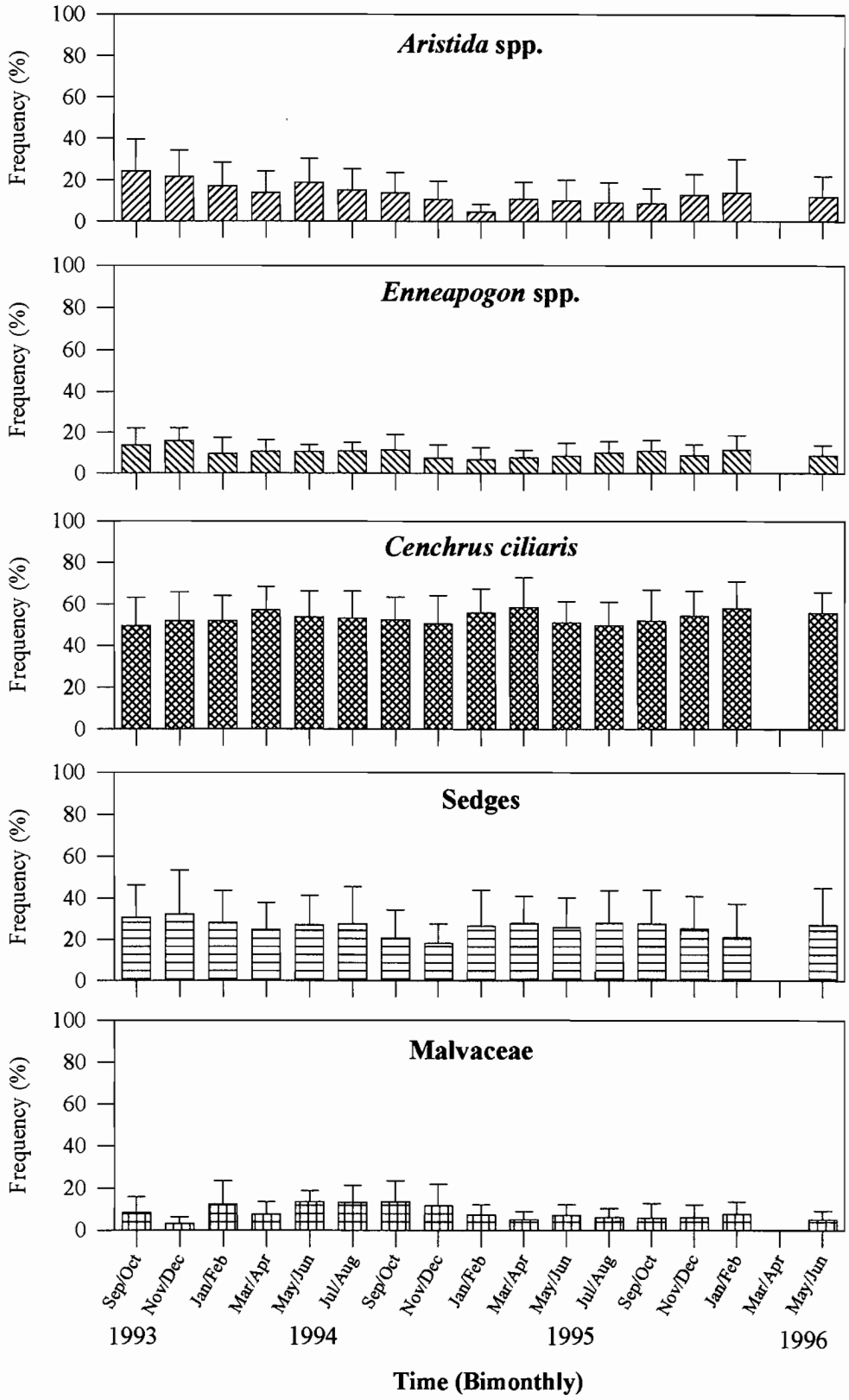


Figure 2.13. Bimonthly frequency of occurrence for selected plant groups, as a mean of the seven BOTANAL transects ± one standard deviation. Data for March/April 1996 were not collected.



species frequencies (Figure 2.13 - category of sedges) but contribute relatively little to the total biomass or species composition due to their slight physical stature. Although these species may be under-represented in measurements of species biomass and composition, the NHN wombat is more likely to encounter these species while foraging.

Forage Availability

Forage choice by the NHN wombat should not be limited by total forage quantity. Total forage biomass was continually above 1000 $kg.ha^{-1}$ (dry matter) throughout any given year of the study, even though the study was conducted in four years of below average rainfall. In comparison with other Australian savanna environments, the minimum total forage biomass at EFNP exceeds that of other studies (e.g. Evans 1992). However, total forage biomass at EFNP was dominated by the introduced *C. ciliaris* with this single species having a frequency of occurrence of up to 58.4% and contributing up to 93.9% of the species composition (Figures 2.11 to 2.13). If the NHN wombat does not consume *C. ciliaris* in quantities equal to or above its frequency of occurrence (i.e. random feeding), the wombat may forage in a non-random manner to select plant species with a lower frequency of occurrence. Therefore, if the wombat does feed in a non-random manner, it is the availability of plant species other than *C. ciliaris* that would influence the wombat's foraging decisions.

Many of the plant species consumed by the NHN wombat (Chapter 3, Flößer 1986, Crossman 1988) differed with time and between transects in their composition, frequency of occurrence and biomass (Table 2.3). Therefore, it seems logical that forage selection could be a direct response to temporal and spatial changes in species composition, frequency and biomass (Table 2.3), as well as changes in forage quality (section 2.4). However, two of the principal species consumed by the NHN wombat, *Aristida* spp. and *Enneapogon* spp., show little temporal variability. This suggests that the NHN wombat may consume plant species that are equally encountered throughout the year, despite changes in their concentrations of nutrients.

2.3.3.2 Primary Production

Primary productivity in the wombat habitat was distinctly seasonal and was strongly correlated with rainfall with pulses of productivity occurring after summer rainfall (Tables 2.3&2.4). Spatial variation between sites approached significance (one-way ANOVA: $F_{4,325} = 2.31$; $p = 0.058$). There was no difference in primary productivity between the three different micro-vegetation communities (one-way ANOVA $F_{2,327} = 0.077$; $p = 0.926$). This suggests that relative primary productivity was even across the habitat surrounding the wombat burrows. Likewise, there was no difference in primary productivity between the fenced or unfenced control (one-way ANOVA $F_{1,328} = 0.556$; $p = 0.456$), suggesting that the impact of vertebrate grazers was low.

2.4 FORAGE QUALITY

2.4.1 Forage Quality and Herbivory

As described in Chapter 1, the diet selection by a herbivore should be in response to forage availability and quality, nutritional requirements and digestive constraints (Illius and Gordon 1992, Illius and Gordon 1993, Kohlmann and Risenhoover 1994, Gordon and Illius 1996, Illius and Gordon 1999). Important attributes of forage quality for the nutrition of grazing herbivores include total nitrogen concentration (e.g. Owen-Smith 1982), digestibility (e.g. Van Soest 1964, 1965, Loeb *et al.* 1991, Tixier *et al.* 1997), and to a lesser extent, the concentration of soluble carbohydrates (e.g. Radojevic *et al.* 1994, Tixier *et al.* 1997).

2.4.2 Near-infrared Spectroscopy: Application to Ecological Studies

Traditional methods of measuring nutritional attributes of forage quality are expensive, time consuming and can be subject to many sources of error. Near-infrared spectroscopy (NIRS) offers the analyst the ability to accurately predict forage quality with a high degree of precision, rapidly and non-destructively.

Principle of Near-infrared Spectroscopy

Molecular bonds between two or more elements are not static connections (Shenk *et al.* 1992) but are continually vibrating. For molecular bonds of the type

Table 2.4. Mean bimonthly primary productivity (kg.ha^{-1} DM) ± one standard deviation across three different micro-vegetation communities surrounding five wombat burrows.

		Buffel grass surrounding wombat burrows		Native grasses (approximately 15 metres from the nearest burrow)		Free growing stands of buffel grass (approximately 30 metres from the nearest burrow)	
Year	Bimonth	Grazed	Exclosed	Grazed	Exclosed	Grazed	Exclosed
1994	Jul/Aug	0	0	0	0	0	0
	Sep/Oct	0	0	0	0	0	0
	Nov/Dec	0	0	0	0	0	0
1995	Jan/Feb	58.8 ± 40.9	61 ± 38.5	31.8 ± 19.2	32.4 ± 14.2	48 ± 59.0	60.2 ± 87.8
	Mar/Apr	4.6 ± 6.3	1.2 ± 1.1	2.6 ± 2.1	13.4 ± 9.8	8.8 ± 11.0	4.8 ± 2.3
	May/Jun	0	0	0	0	0	0
	Jul/Aug	0	0	0	1.8 ± 4.0	0	0
	Sep/Oct	0	0	0.2 ± 0.3	1.2 ± 1.1	0.2 ± 0.2	0.7 ± 0.8
	Nov/Dec	0.6 ± 0.7	2.2 ± 1.9	7.4 ± 9.1	12.3 ± 11.2	3.8 ± 3.6	3.0 ± 2.4
1996	Jan/Feb	10.9 ± 16.1	13.7 ± 10.9	16.7 ± 18.5	43.4 ± 28.8	20.4 ± 22.1	14.3 ± 8.5
	Mar/Arp	-	-	-	-	-	-
	May/Jun	0	0	4.4 ± 4.0	9.8 ± 5.1	1.1 ± 2.5	2.1 ± 2.8

X-H (where X represents carbon, nitrogen or oxygen), hydrogenic stretching, bending or deformation vibrations occur in the near-infrared region - 700 to 2500 nm (Shenk *et al.* 1992, Osborne *et al.* 1993, Shenk and Westerhaus 1994). Similar molecular bond vibrations occur for functional groups including carbon-carbon bonds, carbonyl bonds (CHO) and metal halides (Shenk *et al.* 1992). When monochromatic near-infrared light is impinged on finely ground plant material, the light is reflected, refracted, absorbed, diffracted and/or transmitted (Shenk *et al.* 1992). Therefore quantifying the near-infrared radiation reflected or transmitted (in the form of harmonics or overtones) provides detail on the chemical composition of the ground plant material (Foley *et al.* in press).

Attributes of forage quality, including concentrations of protein, fibre, *in vitro* digestibility and soluble carbohydrates, can be measured by NIRS. Protein, cellulose, lignin and carbohydrates are all classes of organic compounds that can be generalised with X-C bonds. Shenk *et al.* (1992) detail specific bond vibrations (X-C or X=C) and their characteristic near-infrared wavelengths. Further, Shenk *et al.* (1992) generalise the basic characterising near-infrared wavelengths for protein (2180 nm), cellulose (2336 nm), lignin (2270 nm) and carbohydrates (2100). However, in a sample of finely ground plant material the absorbance spectra are not distinct because of a combination of primary absorptions, overtones and scattering of some of the near-infrared light (Foley *et al.* in press). Therefore, interpretation of spectra has to be made by indirectly comparing them with samples of known composition, using multivariate analysis (Foley *et al.* in press).

Multivariate Quantitative Analysis

Several multivariate models have been used in NIRS calibrations including principal component regression (PCR), partial least squares (PLS) and modified partial least squares (MPLS) regression techniques, and are considered standard practice for quantitative NIRS analysis (Anon 1995). Each multivariate model uses wavelength data of the spectra (including location, height and width) to generate predictive equations for forage quality attributes (Shenk *et al.* 1992, Shenk and Westerhaus 1994).

Each quantitative multivariate model requires validation to prevent over-fitting. Two approaches have been used. The first uses external validation using either randomly selected samples from the calibration set or a separate group of samples (Shenk and Westerhaus 1994). Samples selected from within the calibration set can provide estimates of prediction error and detect over-fitting (Shenk and Westerhaus 1994). Samples from a separate group can be used for testing the robustness of the predictive equation but should not be used to estimate prediction error or detect over-fitting (Shenk and Westerhaus 1994). The second approach to validate the predictive model is cross-validation; a form of Monte Carlo simulation. Cross-validation obtains validation errors by partitioning the calibration set into several groups (Shenk and Westerhaus 1994). Then, a calibration is performed on all but one group, with the remaining group serving as the validation group. This procedure is repeated until every sample has been predicted and validated once, generating a standard error of cross-validation (SECV) (Shenk and Westerhaus 1994). By virtue of using all samples in the validation and calibration, the cross-validation procedure generates better estimates of prediction error than external validation and is consequently recommended by Shenk and Westerhaus (1994) as the preferred procedure.

Applications of Near-infrared Spectroscopy

Near-infrared spectroscopy was first used to predict composition of legumes and grass feeds by Norris *et al.* (1976). Attributes of forage quality successfully predicted from NIRS included total nitrogen (N) (and consequently crude protein (CP)), organic matter (OM), neutral detergent fibre (NDF), acid detergent fibre (ADF), acid-lignin (AL), *in vitro* dry matter disappearance (or digestibility - IVDMD), *in vivo* digestibility, intake and digestible energy intake (Ward *et al.* 1982, Brooks *et al.* 1984, Meuret *et al.* 1993, García-Ciudad *et al.* 1993, Aragones 1996). The predictive models for these forage quality attributes have proved to be very accurate (Table 2.5). Since then, the agricultural industry has successfully used NIRS to measure forage quality in both regulated and managed grazing systems, and animal feed production enterprises (Shenk *et al.* 1979; Ward *et al.* 1982; Baker *et al.* 1994).

Table 2.5. Review of validation statistics for studies using NIRS to predict attributes of terrestrial forage quality. Validation statistics include coefficient of determination (r^2) and a measurement of standard error*. Forage quality attributes predicted include Total Nitrogen (N) or Crude Protein (CP), Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF), Acid Lignin (AL), Organic Matter (OM) and *in vitro* Dry Matter Digestibility (IVDMD).

Forage Description and Country of Study	N/CP		NDF		ADF		AL		OM		IVDMD	
	r^2	SE*	r^2	SE*	r^2	SE*	r^2	SE*	r^2	SE*	r^2	SE*
Cultivated crops, USA[1]	0.99	0.74	0.98	2.39	0.96	1.56	0.96	0.80	-	-	-	-
Cultivated grain crops, USA[2]	0.96	0.95	0.95	2.64	0.86	2.31	-	-	-	-	0.89	2.73
Semi-arid rangeland grasses, forbs and browse, USA[3]	0.98	0.37	-	-	0.90	1.26	0.67	0.67	-	-	-	-
Temperate grasses and legumes, USA[4]	1.0	0.03	1.0	0.09	1.0	0.36	0.99	0.23	0.72	1.52	1.0	0.73
Semi-natural grasses and legumes, Spain[5]	0.95	0.56	0.91	1.97	0.87	1.24	0.91	0.47	-	-	-	-
Mature annual legumes, Australia[6]	-	-	0.93	0.31	0.91	0.44	0.79	1.09	-	-	-	-
Trees and shrub foliage, France[7]	0.98	0.11	0.99	1.36	0.97	1.85	0.97	1.04	0.97	0.54	0.99	1.51
Grass silage, UK[8]	-	-	-	-	-	-	-	-	-	-	0.82	2.35
Semi-arid rangeland grasses, Argentina[9]	0.90	0.31	0.94	0.43	-	-	0.93	0.21	-	-	0.97	1.14
Semi-arid rangeland grasses, Spain[10]	-	-	-	-	-	-	-	-	0.88	4.6	-	-

* Standard Error Estimates is Standard Error for Calibration[7,9] (SEC), Standard Error of Prediction[2,,5,6,8,9] (SEP) or Standard Error (SE) of the regression[1,3,4]. SEC is the error due to differences between the laboratory values and the NIR predicted values *within* the calibration set, while SEP is the same error but *outside* the calibration set (Kellaway and Stimson 1993).

Source: 1. Norris *et al.* (1976); 2. Shenk *et al.* (1979); 3. Ward *et al.* (1982); 4. Brooks *et al.* (1984); 5. García-Ciudad *et al.* (1993); 6. Kellaway and Stimson (1993); 7. Meuret *et al.* (1993); 8. Baker *et al.* (1994); 9. Rabotnikof *et al.* (1995); 10. Vázquez de Aldana *et al.* (1996).

The application of NIRS to assess the nutritional quality of forage consumed by non-domestic herbivores is comparatively recent. Brooks *et al.* (1984) successfully used NIRS to assess the quality of forage fed to captive elk (*Cervus elaphus*). They found NIRS provided robust measurements of OM, CP, fibre (NDF, ADF and AL), digestibility (*in vivo* and *in vitro*), intake and average daily dry matter gains or losses for both forage and faeces in captivity. Aragones (1996) extended the application to measure temporal changes in quality of sea grasses in a comprehensive long-term experiment to measure the impact of dugong (*Dugong dugon*) and turtle (*Chelonia mydas*) grazing on seagrass communities. His study again proved the utility of NIRS for measuring quality attributes including N, NDF, ADF, AL, OM and IVDMD. He also used NIRS successfully to measure the concentration of water soluble carbohydrates (WSC).

Many applications of NIRS in agricultural research had only narrow objectives. For example, many applications attempt to explain variation of forage quality within a season, or across several seasons under controlled experimental conditions (e.g. Meuret *et al.* 1993, García-Ciudad *et al.* 1993). Broad-based applications of NIRS to agricultural products are comparatively rare (e.g. Smith and Flinn 1991). Ecological application dictates that NIRS have the ability to predict forage quality between species, within species (plant-parts) and incorporate temporal and spatial variation. The first field-based ecological application of NIRS by Aragones (1996) extended the potential of the technique beyond the bounds of agriculture and animal husbandry. The current study aims to extend the ecological application of NIRS by producing broad-based robust equations to predict forage quality in a heterogeneous habitat and incorporate temporal variation. The products of this study can then be used as a management tool for future forage studies at Epping Forest National Park.

2.4.3 Methods

2.4.3.1 Plant Samples

Whole plant samples were collected at random within the range (300 ha) of the NHN wombat (Section 2.2). Samples were collected bimonthly over a three-year period representing all seasons encountered during this period. Samples

collected represented pasture species potentially consumed by the NHN wombat including grasses, native sedges and native forbs. All samples were cut and oven dried at 60°C for a minimum of 24 hours. Samples were then ground in a cyclone mill (Udy Corporation, Fort Collins, Colorado) to pass a 1-mm screen.

In preparation for collection of NIR spectra, ground samples were placed in a constant temperature (5°C) and relative humidity (15%) environment for a minimum of two weeks so that residual moisture was standardised. Standardising for residual moisture increases the predictive ability and repeatability of NIRS models (Baker *et al.* 1994).

2.4.3.2 Near-infrared Spectroscopy

Collection and Storage of Spectra

One hundred and fifty-four ground samples were scanned with a near-infrared monochromator spectrophotometer (NIRSystems Inc. Model 6500, Silver Spring, Md, USA) with a spinning cup attachment. The instrument was housed at the Bureau of Sugar Experiment Stations, Meringa via Gordonvale, Queensland, and was operated under conditions of constant temperature (22°C) and relative humidity (55%). The instrument operated under ISI software (NIRS 3.11, Infrasoft International, Port Matilda, Pennsylvania, USA), which was also used for subsequent data analysis.

Spectra were collected between 402 nm and 2498 nm at 2 nm intervals. Repeatability of the NIR spectra was measured by deviations in optical data (log 1/R) at each wavelength using an internal ceramic reference tile (Shenk and Westerhaus 1992). The log 1/R function aims to linearise the relationship between the measured reflectance (R) and the absorbance of the substance (Anon 1995). Sub-samples were scanned in duplicate, with the sample being re-packed between samples. Duplicate spectra were averaged and accepted as a spectral data point only if the difference between the two spectra did not exceed a root mean square (RMS) of 50 (as suggested by Westerhaus 1985).

Calibration Set and Calibration Equation Development

A calibration set was identified using the CENTER and SELECT programs of the ISI software. The CENTER program is used to generate an algorithm to rank all spectra according to the Mahalanobis (H) distance from the mean spectrum using principal component scores. The principal component scores were calculated from the principal components (eigenvectors) of the spectra (Shenk and Westerhaus 1992, Shenk and Westerhaus 1994). Mahalanobis distances were then standardised by dividing each value by the mean H to produce 'global H' values (Shenk and Westerhaus 1994). The ranking process of the CENTER-generated algorithm allowed identification of potential outliers or spectra belonging to a different population (Shenk and Westerhaus 1994).

The SELECT program is used to generate an algorithm to identify spectra that are representative of all spectral variability. Selected spectra represented 'neighbourhoods' of common H distances (neighbourhood H), where spectra within the neighbourhood shared similar H distance with its neighbours (or similar spectral variability) (Shenk and Westerhaus 1992). The SELECT-generated algorithm operates on the assumption that one sample is representative of each neighbourhood (Shenk and Westerhaus 1994). A total of 86 samples were identified on the basis of spectral variability. An additional 5 samples were selected so that each species/genus was represented in the calibration set.

To develop the most appropriate predictive model for each forage attribute, a variety of models and mathematical transformations and smoothing functions were explored. Models examined included modified partial least squares (MPLS), partial least squares (PLS) and principle components regression (PCR), with each model cross-validated to prevent over-fitting (Shenk and Westerhaus 1994). The transformations and pre-treatment of the calibration models allowed comparisons of the linearisation process. Model variations included altering the derivatives of log 1/R (e.g. no derivative, first derivative or second derivative), the number of data points over which the derivative is to be calculated, and/or the number of smoothing functions. For example a mathematical transformation and smoothing function of 1,4,4,1 uses the first derivative of log 1/R calculated every 4 data

points (or 8 nm), with the first smoothing occurring across 4 data points and no second smoothing (Shenk and Westerhaus 1992). In addition each calibration model used standard normal variate (SNV) and detrend as a means for scatter correction. Scatter of data "can distort the relationship between the NIR spectrum and the reference value" (Shenk and Westerhaus 1992) and is often caused by variation in particle size and moisture (Shenk and Westerhaus 1991). The SNV and detrend method of correcting for scatter thereby improves the accuracy of the prediction (Shenk and Westerhaus 1991, Shenk and Westerhaus 1992, Baker *et al.* 1994).

A total of twenty-two calibration models were derived for each quality attribute, these were subsequently ranked on the basis of lowest standard error of the cross-validation (SECV) statistic and the highest coefficient of determination (r^2). The model that provided the best predictive capability for each quality attribute was subsequently used to predict the attribute for each species. The relationship between observed and predicted values were assessed by least squares linear regression for each quality attribute. Potential outliers, where the values predicted by the model were significantly different from the observed laboratory value, were identified using principal component scores and the Mahalanobis distance. Laboratory chemistry was repeated on any outliers.

2.4.3.3 Chemical Analysis

Attributes of forage quality measured included organic matter, total nitrogen, neutral detergent fibre, acid detergent fibre, acid lignin, water soluble carbohydrates and *in vitro* dry matter digestibility. Each attribute was measured in duplicate. An estimate of the error of the laboratory procedures (SEL - standard error of the laboratory) was calculated following the method of Smith and Flinn (1991). The SEL provided a measure of precision of the laboratory procedures.

Organic Matter

Samples were dried to a constant mass at 105°C for 24 hours to determine dry matter (DM) content. Organic matter was determined by burning each sample in a muffle furnace at 550°C. Crucibles containing ash were re-weighed at a constant temperature to determine ash weight, and subsequently percent ash

content of DM. The percent of organic matter was calculated subtracting the percentage ash content from one hundred.

Total Nitrogen

Total nitrogen concentration of plant samples were determined by a semi-micro Kjeldahl technique with a selenium catalyst using a Gerhardt Vapodest nitrogen analyser. Recoveries of ammonium were checked against ammonium sulphate standards and were in the range of 99.7% to 101.6% for all analyses.

Fibre - Neutral Detergent Fibre, Acid Detergent Fibre and Acid Lignin

Cell wall constituents were measured as NDF and ADF following the methods of Van Soest *et al.* (1991) but with the filter bag modification described by Komarek *et al.* (1994a&b). This modification, or the filter bag technique (FBT), produces non-significant differences compared to the conventional methods (Table 2.6).

Table 2.6. Comparison of methods of determining the cell wall constituents NDF and ADF. Data are the values of 21 forage types including various types of grasses, manure and commercial feeds. Data are from Komarek *et al.* (1994a&b). Abbreviations are: S.D. - standard deviation; C.V. - coefficient of variation; NDF - neutral detergent fibre; ADF - acid detergent fibre.

Method	Cell Wall Constituent	Mean (%)	S.D.	C.V.
Van Soest *et al.* (1991)	NDF	47.10	0.40	1.30
Komarek *et al.* (1994a)	NDF	46.80	0.70	2.20
Van Soest *et al.* (1991)	ADF	30.28	0.24	1.94
Komarek *et al.* (1994b)	ADF	29.66	0.48	3.69

Between 0.20g and 0.25g of ground plant material was placed in a filter bag and sealed. Twenty-four filter bags per batch were placed in the ANKOM ^{200} Fiber Analyser (ANKOM Technology Corporation, Fairport, NY, USA) and refluxed in either NDF or ADF solution for 60 minutes. Following reflux, five rinses of hot water (80°C to 100°C) removed any solution, with heat stable alpha-amylase added to the first three water rinses of the NDF procedure to solublize starch (Komarek *et al.* 1994a). Two rinses in acetone of five minutes duration removed any chlorophyll from the samples.

Acid lignin was measured on the ADF residue in the filter bags, using the 72% sulphuric acid method described by Van Soest et al. (1991). Seventy-two filter bags containing ADF residue were placed in 1000 ml of 72% sulphuric acid in a 3 litre beaker on an ice bath for 3 hours with occasional agitation. Samples were then washed 3 times in hot water and once in cold water before being air dried.

Like the traditional Van Soest *et al.* (1991) method, the FBT relies upon gravimetric difference. Consequently, a strict protocol of weighing, drying and desiccation was employed during sample preparation and analysis for fibre. All samples (NDF, ADF and AL) were air dried and then dried to a constant mass at 60°C for a minimum of 24 hours. Samples were weighed to four decimal places of a gram in batches of 12, after reaching room temperature in a desiccator.

Water Soluble Carbohydrates

Water soluble carbohydrates were extracted using the method of Radojevic *et al.* (1994). Approximately one gram dry weight of plant sample was extracted once in 8 ml of 80% aqueous ethanol at 80°C for 60 minutes and twice with 8 ml distilled water at 60°C for 60 minutes (Radojevic *et al.* 1994). After each extraction, supernatants were combined. The WSC concentration of the extract was estimated by the anthrone method (Yemm and Willis 1954) and expressed as fructose equivalents.

In vitro Dry Matter Digestibility

The IVDMD determination followed a modification of the Choo *et al.* (1981) method, where samples were placed in the ANKOM filter bags and incubated with acid pepsin solution and fungal cellulase solution at 37°C for 24 hours and 48 hours respectively.

2.4.4 Results and Discussion

2.4.4.1 NIRS Equation Development

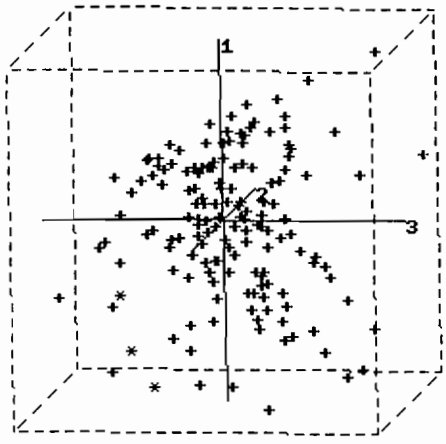
All spectra collected (N = 154) showed an even distribution across three eigenvectors (Figure 2.14) suggesting that all spectra belonged to the same global 'H' (or population) with the possible exception of three outlying spectra. Biologically, the three outliers (one spectra of *Carissa ovata* and two of *Salsola kalii*) are distinct from other forage species considered. *Carissa ovata* should be considered as an 'available' forage resource to browsers but possibly unsuitable for grazers because of its physical defenses (thorns) and the selectivity required for foliage consumption (small leaf). Likewise, the general morphology and growth pattern of *Salsola kalii* is distinct from other forage species considered. It grows rapidly following rain and has thorn like leaves. Once scenesced the whole *S. kalii* plant is wind dispersed, hence its common name 'soft rolly polly'.

Table 2.7 ranks the five best predictive models for each attribute of forage quality out of the twenty-two models explored. The modified partial least squares (MPLS) model explained the maximum variation for each attribute of forage quality. This supports the findings of Shenk and Westerhaus (1994) that MPLS is more stable and accurate than PLS.

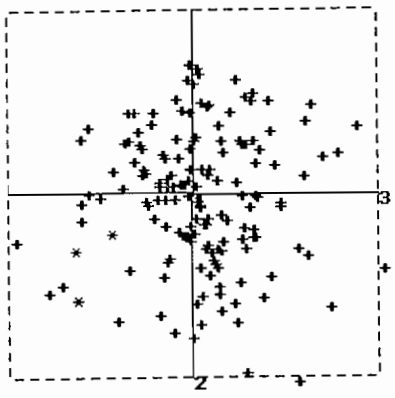
Variation in the mathematical transformations and smoothing functions also improved the predictive model, with the second derivative of log 1/R being most successful in predicting N, OM, IVDMD, NDF and ADF while the first derivative of log 1/R is best for AL and WSC. The optimum gap and the smoothing function for each quality attribute was 5 points (or 10 nm) allowing calculation of second or first derivatives of log 1/R over spectra segments 10 nm in length. The possible exception is for NDF (8 points or 16 nm), however there is little difference between a gap and smooth of 16 nm and 10 nm in the estimates of SEC and r^2. The

Figure 2.14. Symmetry of NIR spectral files. Three dimensional plot (or hypersphere), shown from four perspectives, illustrating the structure and distribution of all spectra. Axes represent the first three principal components. Each spectra is represented by '+' with three global 'H' outliers represented by '*'. Outliers are spectra from the dicotyledons *Salsola kalii* (n=2) and *Carissa ovata*.

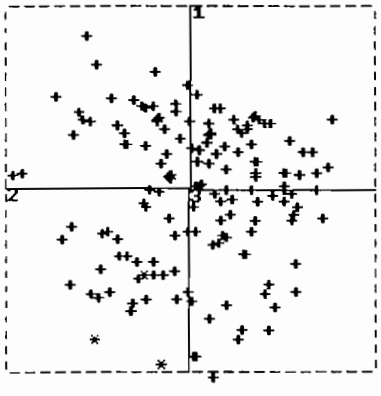
DIAGONAL VIEW



TOP VIEW



RIGHT VIEW



LEFT VIEW

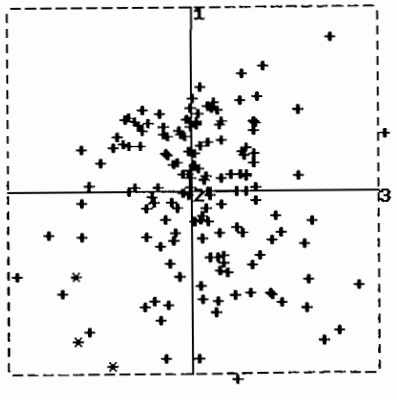


Table 2.7. Rank of five best calibration equations for each attribute of forage quality, ranked according to lowest SECV followed by highest coefficient of determination.

Quality Attribute	Equation Rank	Equation Type*	Derivative	Gap	Smooth	SECV†	SEC‡	r^2
Total N	1	MPLS	2	5	5	0.070	0.043	0.995
	2	MPLS	2	8	8	0.080	0.051	0.993
	3	MPLS	2	4	4	0.091	0.062	0.989
	4	PLS	2	8	8	0.110	0.074	0.984
	5	MPLS	1	4	4	0.112	0.071	0.985
NDF	1	MPLS	2	8	8	2.378	1.485	0.979
	2	MPLS	2	5	5	2.591	1.413	0.982
	3	MPLS	2	4	4	2.605	1.363	0.983
	4	MPLS	1	4	4	2.884	1.979	0.964
	5	MPLS	1	5	5	2.894	1.99	0.964
ADF	1	MPLS	2	5	5	2.850	1.918	0.891
	2	MPLS	2	4	4	2.880	1.828	0.901
	3	MPLS	1	4	4	2.907	2.146	0.877
	4	MPLS	1	5	5	2.910	2.152	0.876
	5	MPLS	0	4	4	3.119	2.459	0.834
AL	1	MPLS	1	5	5	2.284	1.816	0.770
	2	MPLS	1	4	4	2.287	1.813	0.771
	3	PCR	2	5	4	2.334	2.269	0.636
	4	PCR	2	4	5	2.336	2.273	0.634
	5	PCR	2	8	4	2.373	2.292	0.628
OM	1	MPLS	2	5	5	1.705	0.808	0.967
	2	MPLS	2	4	4	1.761	0.812	0.973
	3	PLS	2	4	4	2.130	1.547	0.891
	4	PLS	2	5	5	2.133	1.548	0.891
	5	MPLS	2	8	8	2.156	1.552	0.895
WSC	1	MPLS	1	5	5	0.798	0.588	0.906
	2	MPLS	1	4	4	0.800	0.587	0.907
	3	MPLS	1	10	10	0.865	0.626	0.893
	4	MPLS	1	8	8	0.866	0.612	0.898
	5	MPLS	2	5	5	0.867	0.697	0.862
IVDMD	1	MPLS	2	5	5	3.386	2.184	0.964
	2	MPLS	2	4	4	3.439	2.098	0.967
	3	MPLS	2	8	8	3.615	2.369	0.958
	4	PLS	2	8	8	3.933	3.032	0.931
	5	PLS	1	4	4	4.045	3.215	0.920

*MPLS Modified Partial Least Squares; PLS Partial Least Squares; PCR Principal Components Regression; †SECV Standard Error of Cross Validation; ‡SEC Standard Error of Calibration; r^2 Coefficient of determination. All equations were derived using log 1/R data subjected to standard normal variate (SNV) and detrend method of correcting for scatter (Baker *et al.* 1994). The tabulated results represent the five best equations using permutations of the following mathematical transformation and smoothing functions: 0,4,4; 1,4,4; 2,4,4; 0,5,5; 1,5,5; 2,5,5; 0,8,8; 1,8,8; 2,8,8; 1,10,10; and the statistical methods MPLS, PLS and PCR. Quality attributes assessed include Total Nitrogen (N), Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF), Acid Lignin (AL), Organic Matter (OM), *in vitro* Dry Matter Digestibility (IVDMD), Water Soluble Carbohydrates (WSC).

highest ranked model for each forage attribute produces comparable values for the coefficient of determination and standard error of the calibration to those of other studies (Table 2.5).

The performance of the least-squares regression equations for the values predicted by the NIRS models compared to the laboratory values is dependent on the accuracy of the laboratory analysis. The precision of the analytical technique for each attribute of forage quality is measured by the standard error of the laboratory (SEL) (Table 2.8a). The semi-micro Kjeldahl technique for measuring total nitrogen and water soluble carbohydrate extractions with anthrone determinations proved the most accurate laboratory assays. The greatest source of error in the laboratory analysis was associated with the sequential fibre analysis.

The relationship between the values predicted by NIRS and the values measured in the laboratory (Figure 2.15) was highly significant ($p \leq 0.001$) for all quality attributes. The accuracy of the relationship is indicated by the standard error of the cross validation (SECV), the one-minus the variance ratio (1-VR), and the deviation of the regression line from the equation $y = x$ (where y is the NIRS predicted value and x is the laboratory measured value, with a slope of 1.00 and an axes intercept about the origin). The slopes of the values measured in the laboratory and the values predicted by NIRS do not differ significantly from 1.00 ($p = 0.001$) for all measures of forage quality. This confirms the utility of NIRS to successfully and reliably predict each attribute of forage quality. However, while the slopes of each model were not statistically different from one (Table 2.8), the observed difference from one, with the exception of N, can be primarily attributed to the accuracy of the laboratory assays. Errors in the laboratory analysis, measured by the SEL, are discussed in detail in Section 2.6.

2.4.4.2 Temporal changes in forage quality

The predictive models of each of the attributes of forage quality was applied to each of the 154 samples scanned by the NIR spectrophotometer. The results generated for individual species, incorporating seasonal variation, are tabulated in Appendix 1.

Figure 2.15. Relationship between the attributes of forage quality measured in the laboratory and value predicted by NIR spectrometry. All values are expressed as a percentage of dry matter.

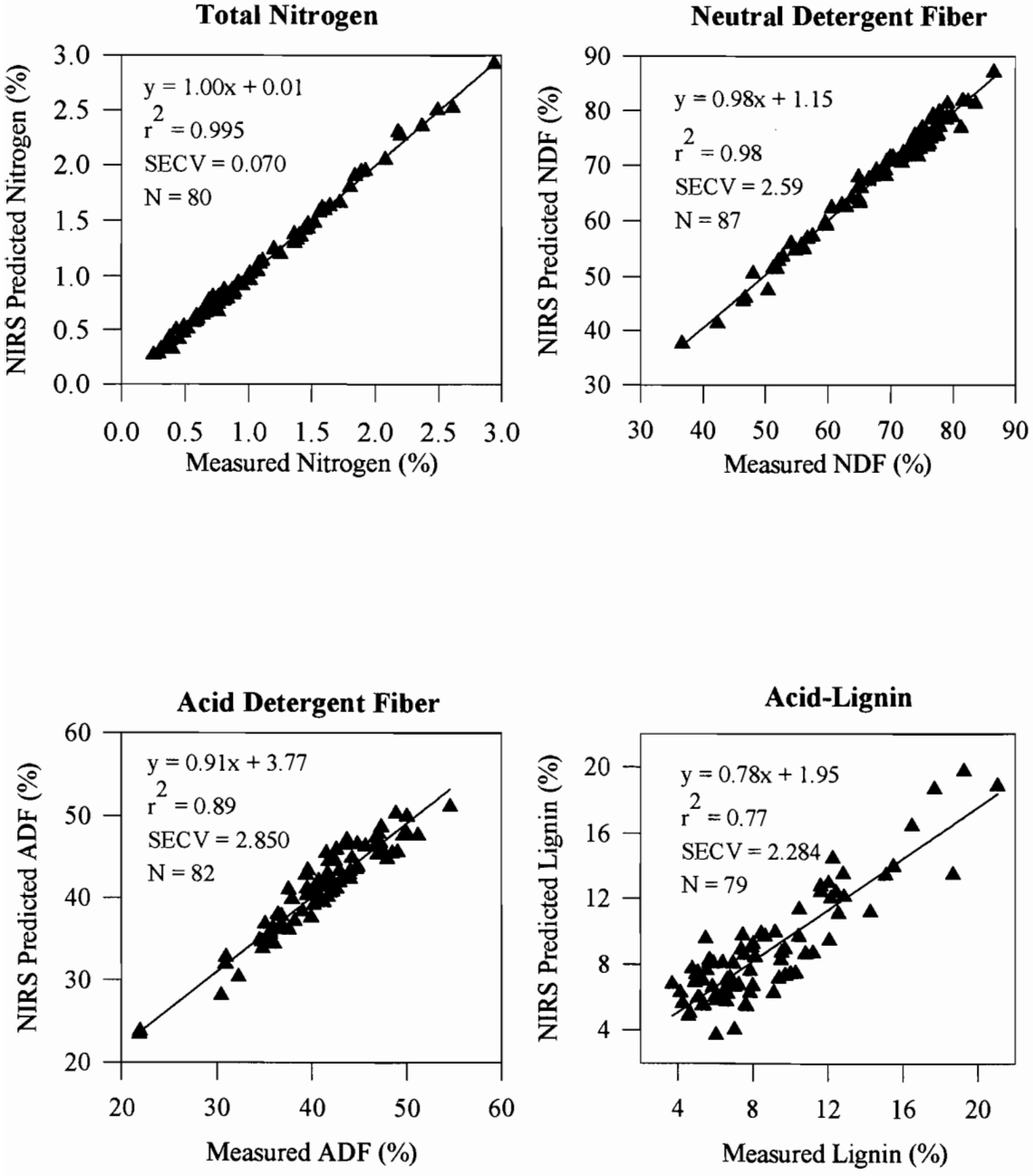


Figure 2.15 (continued). Relationship between the attributes of forage quality measured in the laboratory and value predicted by NIR spectrometry. All values are expressed as a percentage of dry matter.

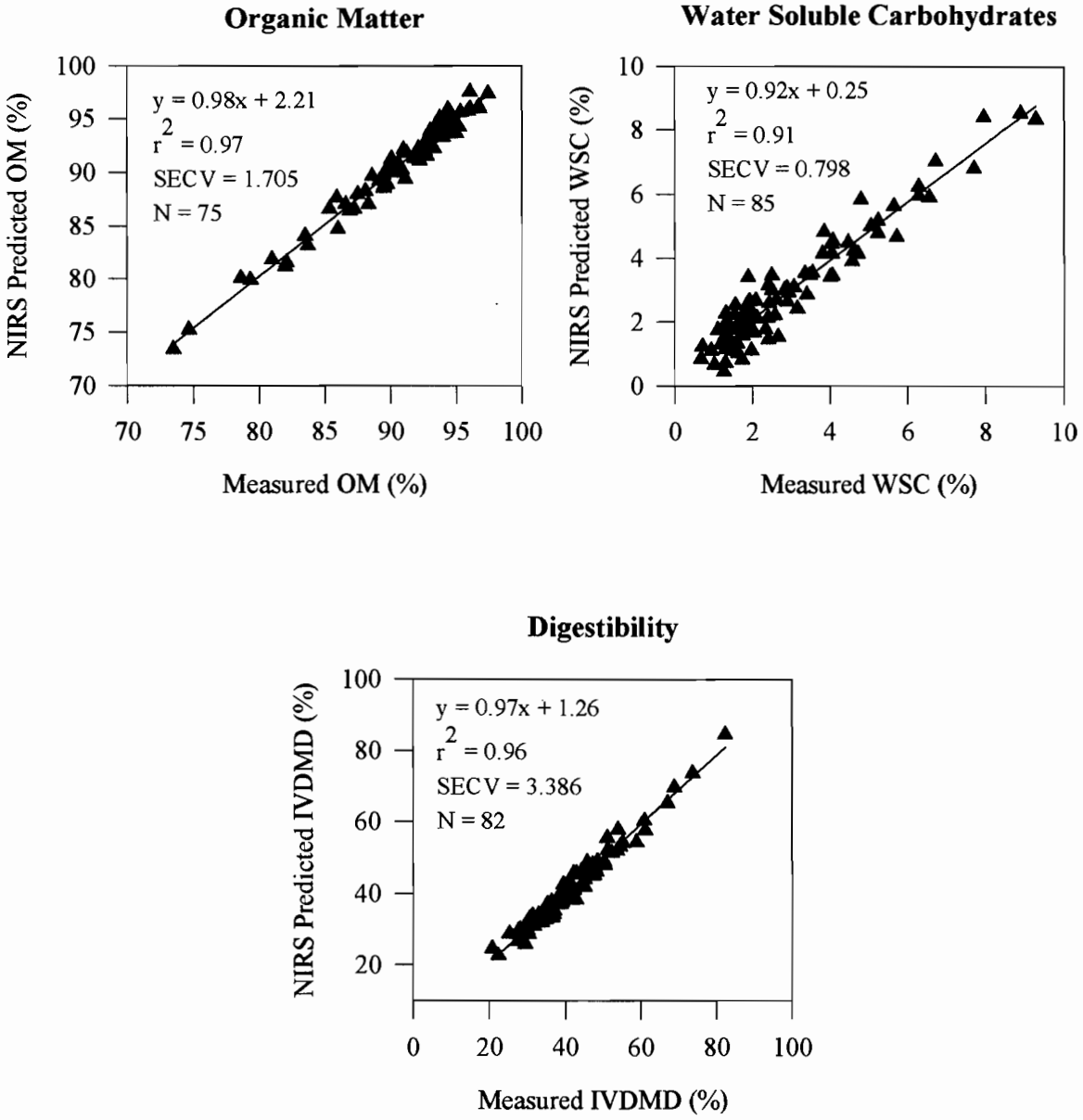


Table 2.8. Performance of both laboratory methods and NIR to measure attributes of forage quality.

a) Laboratory accuracy assessment. '*N*' represents the number of samples in the calibration set. Mean and range of forage quality attributes expressed as percentage of dry matter. Standard Error of the Laboratory (SEL) is: $SEL = \sqrt{(\Sigma(y_1 - y_2)^2 / N)}$ where y_1 and y_2 are duplicates of analyses (Smith and Flinn 1991, Aragones 1996).

b) Least squares linear regression statistics for predicted values for forage quality attributes using NIR spectra, with the appropriate statistical model, versus actual measurements made by laboratory analyses. Error between laboratory values and the values predicted by NIRS is measured by the standard error of the cross-validation (SECV) and 1-VR. The SECV provides an estimate of accuracy (%) and 1-VR (one minus the variance ratio; where the variance ratio is the explained variance divided by the total variance) explains variation (%) in the reference method values explained by NIRS (Shenk and Westerhaus 1992). Validity of the statistical model assessed by the difference of the slope from 1.0. Degrees of freedom (df) equal *N* - 2 for variation in regression coefficient from 1.0. Significance level $t_{0.001} \approx 3.390$ (based on df 100).

	a) Laboratory Accuracy Assessment				**b) Statistical Model Performance Assessment**							
					Regression Equation						*Variation in Slope from 1.0*	
	N	Mean ± SD	Range	SEL	Slope	Intercept	r^2	SECV	1-VR	Significance	*t* value	Different from 1.0
Total N	80	1.07 ± 0.60	0.25 - 2.94	0.02	1.00	0.01	0.99	0.07	0.99	***	-1.25	No
NDF	87	69.20 ± 10.33	36.63 - 86.50	1.42	0.98	1.15	0.98	2.59	0.95	***	-8.06	No
ADF	82	41.13 ± 5.80	21.84 - 54.49	1.16	0.91	3.77	0.89	2.85	0.76	***	-20.46	No
AL	79	8.78 ± 3.79	3.70 - 21.04	1.29	0.78	1.95	0.77	2.28	0.63	***	-11.89	No
OM	75	91.02 ± 4.44	73.48 - 97.44	0.44	0.98	2.21	0.97	1.71	0.86	***	-25.59	No
WSC	85	41.38 ± 11.51	20.73 - 82.23	0.08	0.92	0.25	0.91	0.80	0.92	***	-5.11	No
IVDMD	82	3.01 ± 1.92	0.68 - 9.28	1.34	0.97	1.26	0.96	3.39	0.83	***	-5.94	No

Attributes of forage quality include Total Nitrogen (N), Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF), Acid Lignin (AL), Organic Matter (OM), *in vitro* Dry Matter Digestibility (IVDMD), Water Soluble Carbohydrates (WSC). Coefficient of determination represented by r^2, with significance levels for regression at $p < 0.001$ (***).

To illustrate the temporal variation in forage quality, bimonthly trends over the study period for each attribute of forage quality are graphically represented in Figures 2.16 to 2.22. Forage categories illustrated include native grasses (family Poaceae), the introduced grass *Cenchrus ciliaris*, native sedges (*Fimbristylis* spp.), forbs, and species from the Malvaceae family. While illustrative of temporal trends, these figures present only mean values expressed without a standard deviation because the sample size was not large enough to generate meaningful measures of variability.

Nitrogen

Trends in total nitrogen (N) concentration are illustrated (Figure 2.16). The reference line represents the critical N level (0.8% N or 5% crude protein) for maintenance of cattle feeding upon tropical grasses as defined by Owen-Smith (1982). Native grasses and *C. ciliaris* were, for the majority of the study period, near or below Owen-Smith's (1982) critical value. Consequently, if cattle were to utilise these forage groups as the principal component of their diet, they would presumably enter a negative nitrogen balance and lose body condition. Sedges, forbs and Malvaceae had total N concentrations consistently higher than this critical total N concentration of 0.8%.

Concentrations of total N were positively correlated with rainfall for all forage groups but statistically significant for just *C. ciliaris* and native grasses (Table 2.9). This was most apparent for native grasses, *C. ciliaris* and sedges, with peaks in total N content directly related to rainfall events (Figure 2.16).

Fibre

Native grasses and *C. ciliaris* were generally higher in neutral detergent fibre (NDF) than sedges, and forbs (including Malvaceae) (Figure 2.17). Further, NDF content of both groups of grasses were generally less variable than forbs and sedges with time. Trends in acid detergent fibre (ADF) (Figure 2.18) and acid lignin (AL) (Figure 2.19) did not produce the same quantitative differentiation between the two grass groups and the sedges and forbs but the temporal variation in sedges and forbs remained. However, these results for the components of fibre

are just trends and require a larger sample size and a measure of error to confirm these conclusions.

Fibre did not have a clear relationship with rainfall (Table 2.9) with each forage group responding differently to rainfall and the long term effects of rainfall. This may reflect different growth patterns of each forage group.

Digestibility

The *in vitro* dry matter digestibility (IVDMD) of native grasses was lower than any forage group, including *C. ciliaris* (Figure 2.20). This trend was consistent throughout the study period. The IVDMD of native grasses and sedges were positively correlated with immediate rainfall but not at a level of statistical significance. The IVDMD of sedges and forbs (including Malvaceae) showed the greatest positive correlation with rainfall lagged by six months, again not at a level of statistical significance. The IVDMD for *C. ciliaris* was significantly correlated with rainfall lagged by six month. This suggests different growth patterns of each forage group, particularly in response to rainfall.

Organic Matter

Organic matter contents were as expected for plant material or even a little lower suggesting large amounts of silica or other minerals. All forage groups were high in organic matter (OM) (Figure 2.21), as expected for plant material. Grasses, both native and introduced, and sedges maintained relatively consistent levels of organic matter. Forbs and Malvaceae were more variable in OM content reflecting different morphology between monocotyledons and dicotyledons. Organic matter for each forage group was positively correlated with rainfall lagged between 6 and 10 months (Table 2.9).

Water Soluble Carbohydrates

Concentrations of water soluble carbohydrate (WSC) ranged between 0.16% and 6.79% of dry matter (expressed as fructose equivalents). Native grasses, *Cenchrus ciliaris* and sedges showed little temporal variation in WSC concentration (Figure 2.22). However the concentrations of WSC in both native grasses and *C. ciliaris* were correlated in a positive trend with rainfall with a 2

Table 2.9. Correlation of attributes of forage quality with rainfall, and the long-term effect of rainfall associated with bimonthly time lags ($r = 0.81$; $p \leq 0.05$; 4 df). Significant correlations with rainfall are highlighted in bold underlined text. Fibre values represented by neutral detergent fibre (NDF); IVDMD is *in vitro* dry matter digestibility; OM is organic matter; WSC is water soluble carbohydrates. All forage quality attributes were measured as a percentage of dry matter.

		TIME LAG (MONTHS)					
Correlation Parameters		**0**	**2**	**4**	**6**	**8**	**10**
Total Nitrogen	Native Grasses	**0.91**	-0.24	-0.07	-0.38	-0.22	-0.01
	Cenchrus ciliaris	**0.81**	-0.11	-0.04	-0.29	-0.21	-0.60
	Sedges	0.67	-0.09	0.06	-0.47	-0.41	-0.03
	Forbs	0.40	0.21	0.15	-0.55	-0.29	-0.17
	Malvaceae	0.39	0.23	0.55	-0.35	-0.58	**-0.82**
Fibre (NDF)	Native Grasses	0.03	-0.32	-0.43	0.22	0.28	-0.19
	Cenchrus ciliaris	-0.21	-0.12	-0.62	0.09	0.55	**0.94**
	Sedges	0.51	-0.07	-0.05	-0.59	0.02	0.06
	Forbs	0.38	-0.43	-0.21	0.01	0.29	-0.19
	Malvaceae	0.04	-0.52	-0.53	0.12	0.64	0.55
IVDMD	Native Grasses	0.42	0.09	0.12	-0.34	-0.58	0.45
	Cenchrus ciliaris	0.42	-0.18	0.41	-0.06	-0.35	**-0.95**
	Sedges	0.14	-0.20	0.39	-0.08	-0.57	-0.08
	Forbs	-0.07	-0.03	0.18	0.04	-0.34	-0.23
	Malvaceae	-0.15	0.10	0.73	-0.01	-0.40	-0.74
OM	Native Grasses	-0.08	0.34	-0.10	-0.66	0.23	0.34
	Cenchrus ciliaris	-0.61	0.39	-0.33	0.29	-0.06	0.43
	Sedges	-0.38	-0.01	0.19	0.40	-0.05	0.17
	Forbs	-0.03	0.26	-0.12	-0.24	0.25	0.29
	Malvaceae	-0.19	-0.05	-0.56	0.23	0.54	**0.83**
WSC	Native Grasses	-0.34	0.60	0.01	0.13	-0.07	0.19
	Cenchrus ciliaris	-0.39	0.38	0.30	0.34	-0.23	-0.46
	Sedges	-0.16	-0.30	-0.01	**0.86**	0.09	-0.36
	Forbs	-0.38	0.22	-0.09	0.08	-0.09	0.58
	Malvaceae	0.48	0.35	-0.05	-0.44	-0.41	-0.28

Figure 2.16. Bimonthly trends in total N concentration of plant groups,as a percentage of dry matter predicted by NIRS. Reference line represents critical N level for maintenance of cattle feeding upon tropical grasses - 0.8 % N (Owen-Smith 1982).

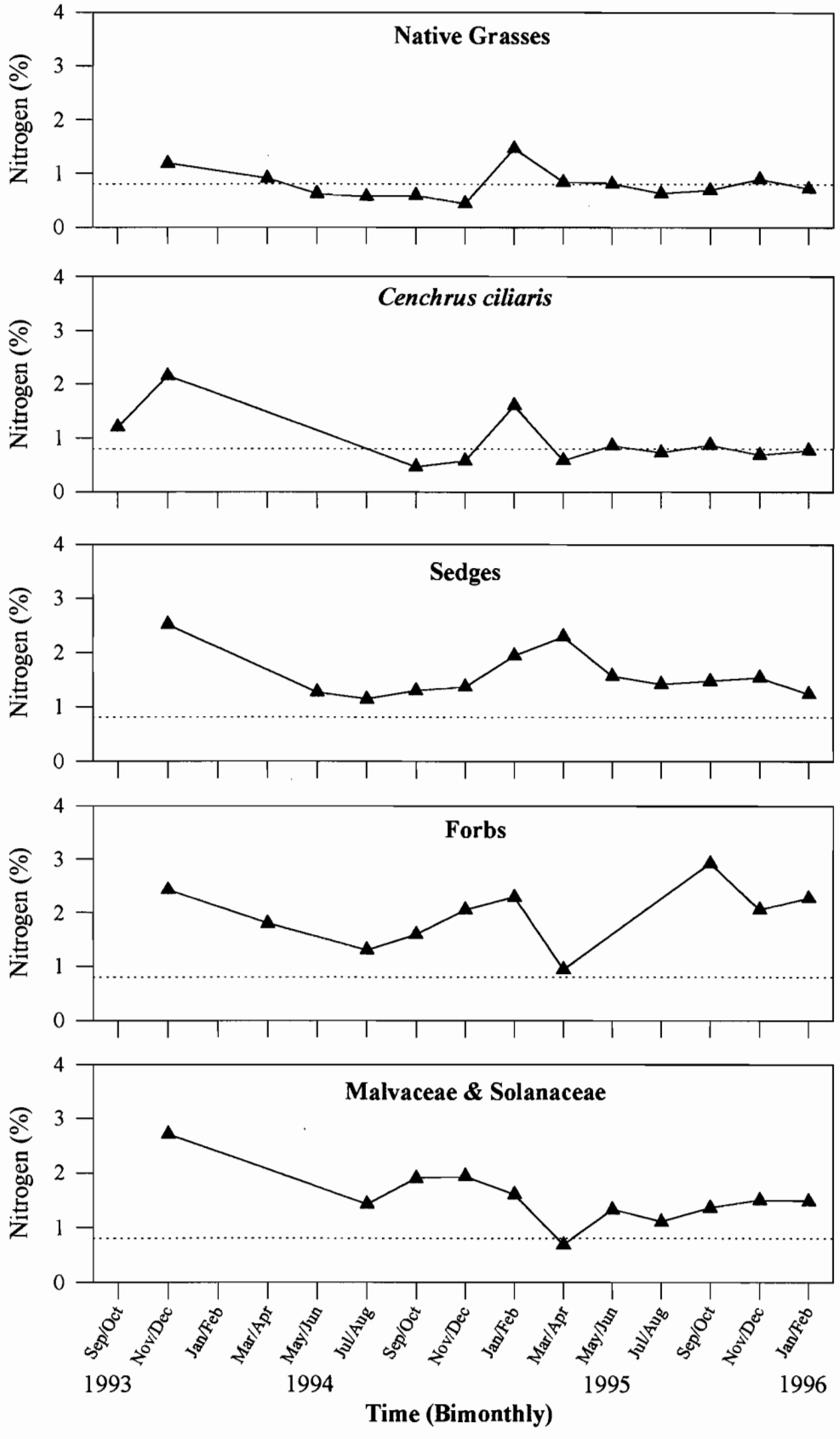


Figure 2.17. Bimonthly trends in Neutral Detergent Fibre (NDF) content of plant groups as predicted by NIR spectroscopy. NDF expressed as a percentage of dry matter.

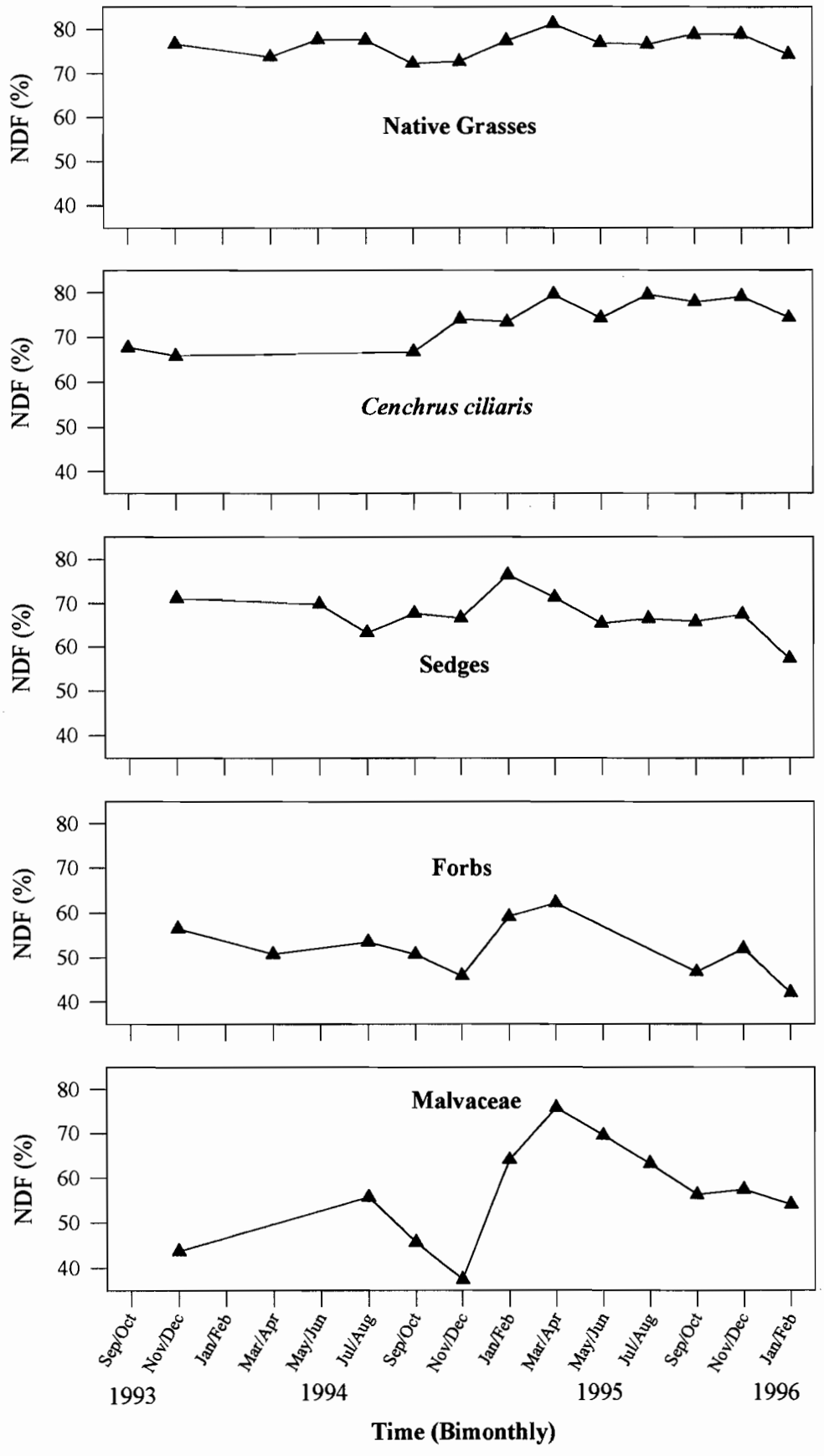


Figure 2.18. Bimonthy trends in Acid Detergent Fibre (ADF) content of plant groups as predicted by NIR spectroscopy. ADF expressed as a percentage of dry matter.

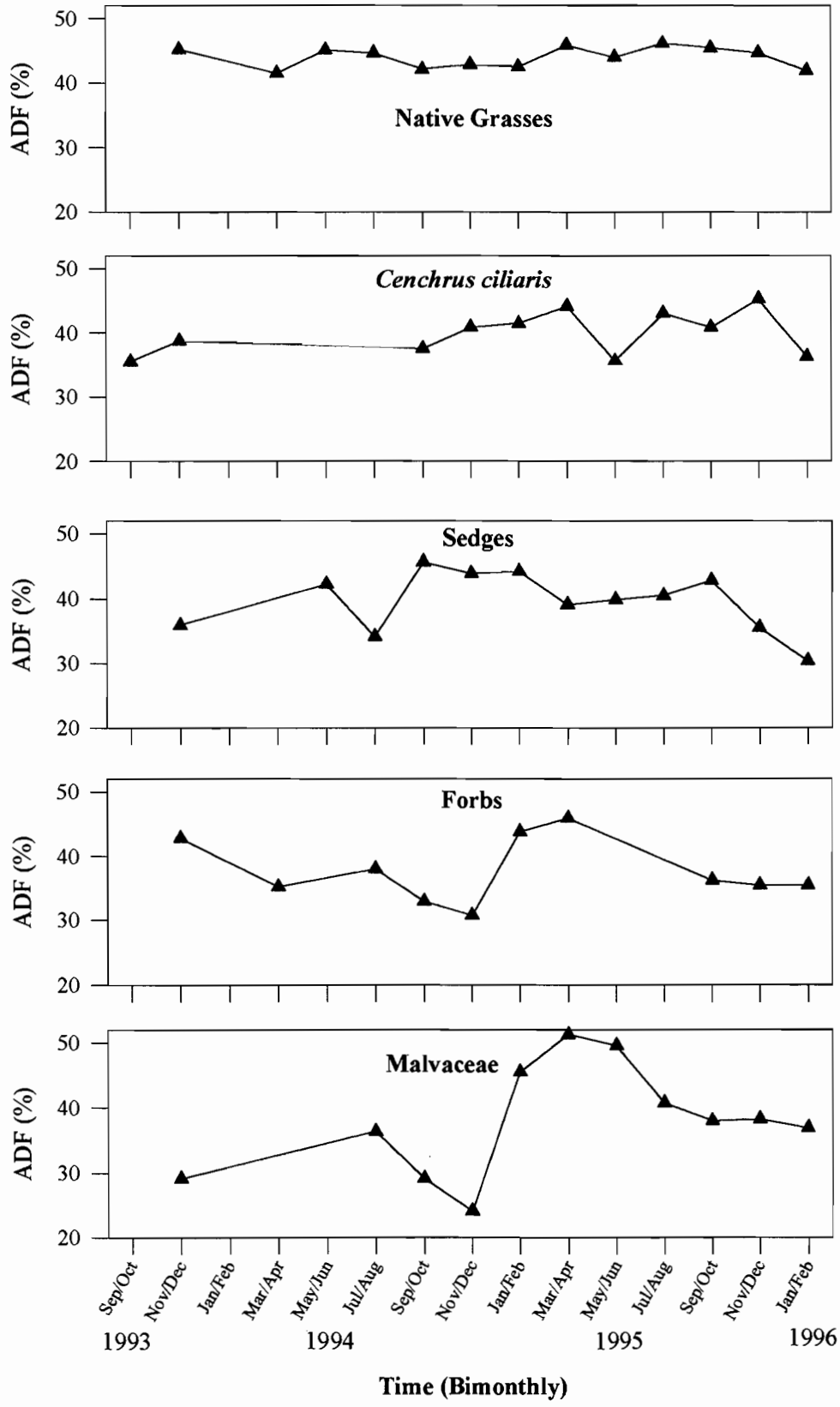


Figure 2.19. Bimonthy trends in Acid Lignin (AL) content of plant groups as predicted by NIR spectroscopy. AL is expressed as a percentage of dry matter.

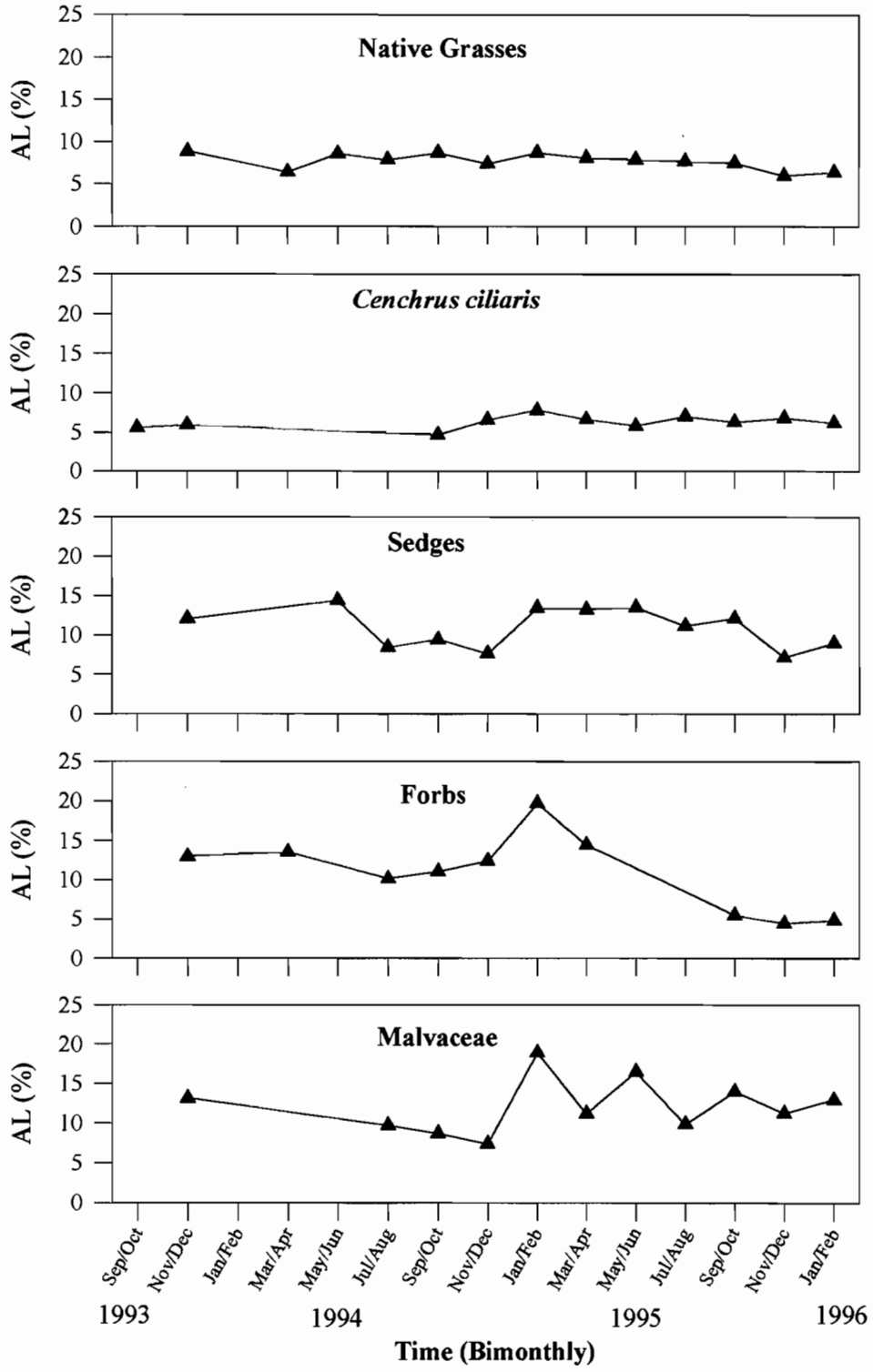


Figure 2.20. Bimonthly trends in *in vitro* dry matter digestibility (IVDMD) of plant groups as predicted by NIR spectroscopy. IVDMD expressed as a percentage of dry matter.

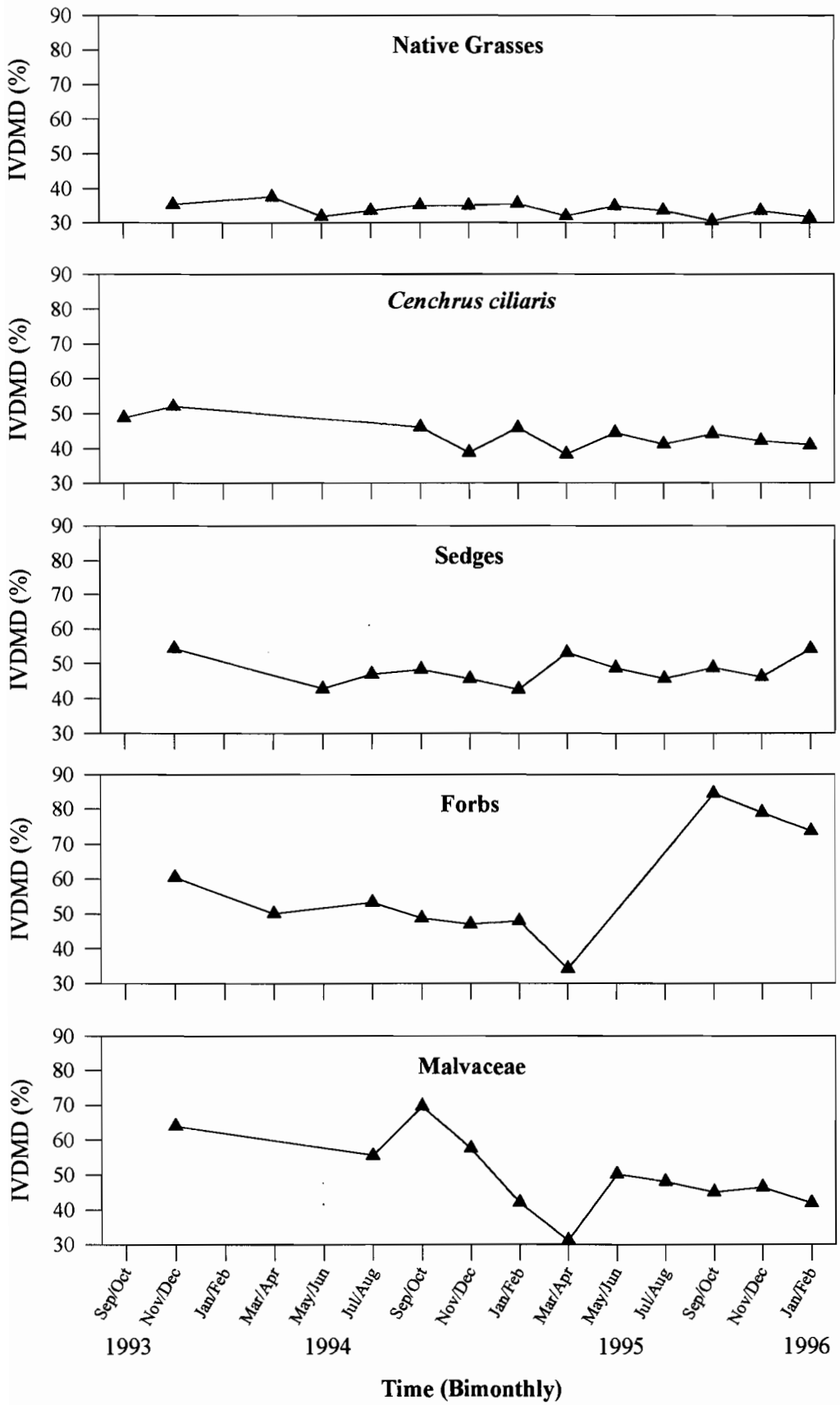


Figure 2.21. Bimonthly trends in total organic matter (OM) content of plant groups, as predicted by NIR spectroscopy. Organic matter is expressed as a percentage of dry matter.

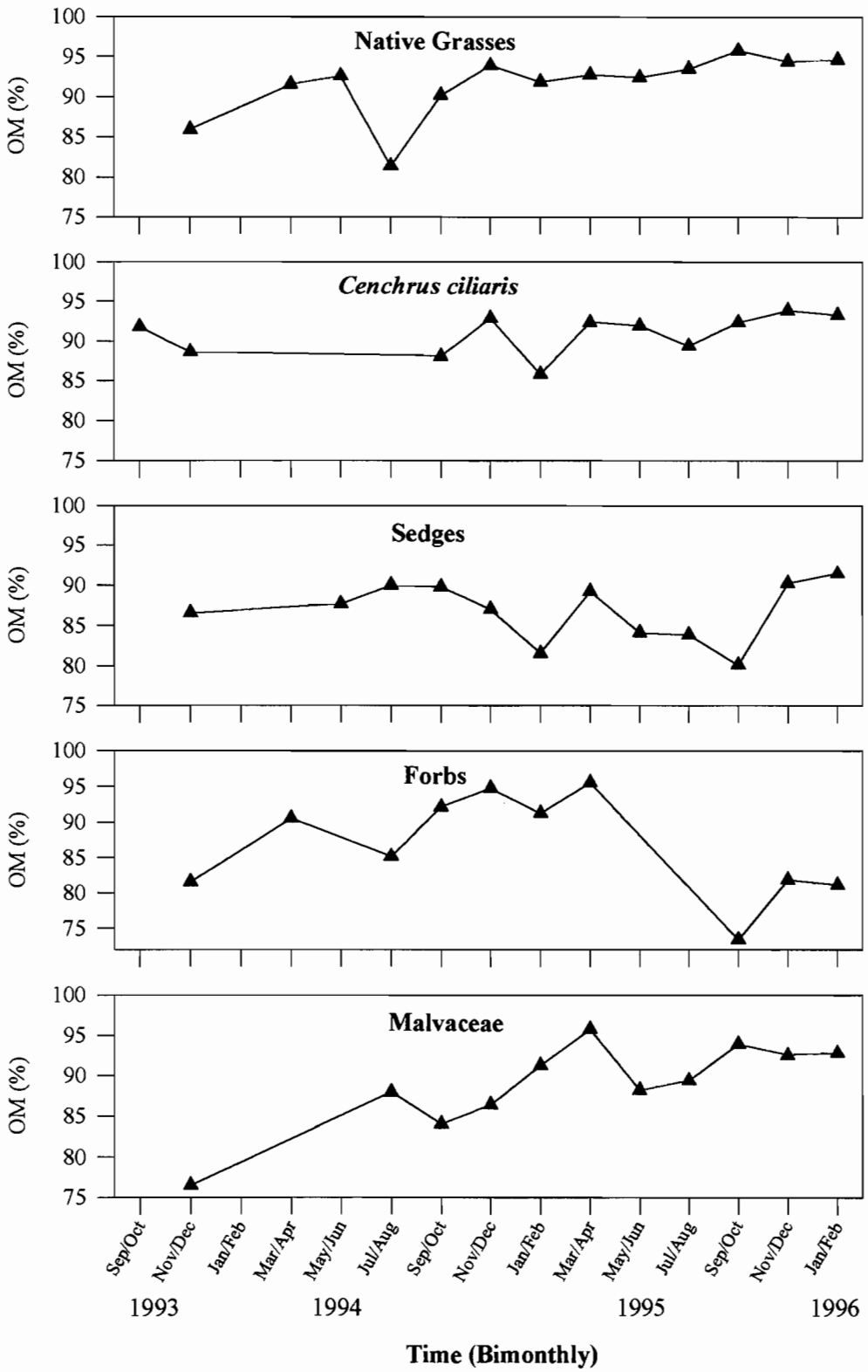
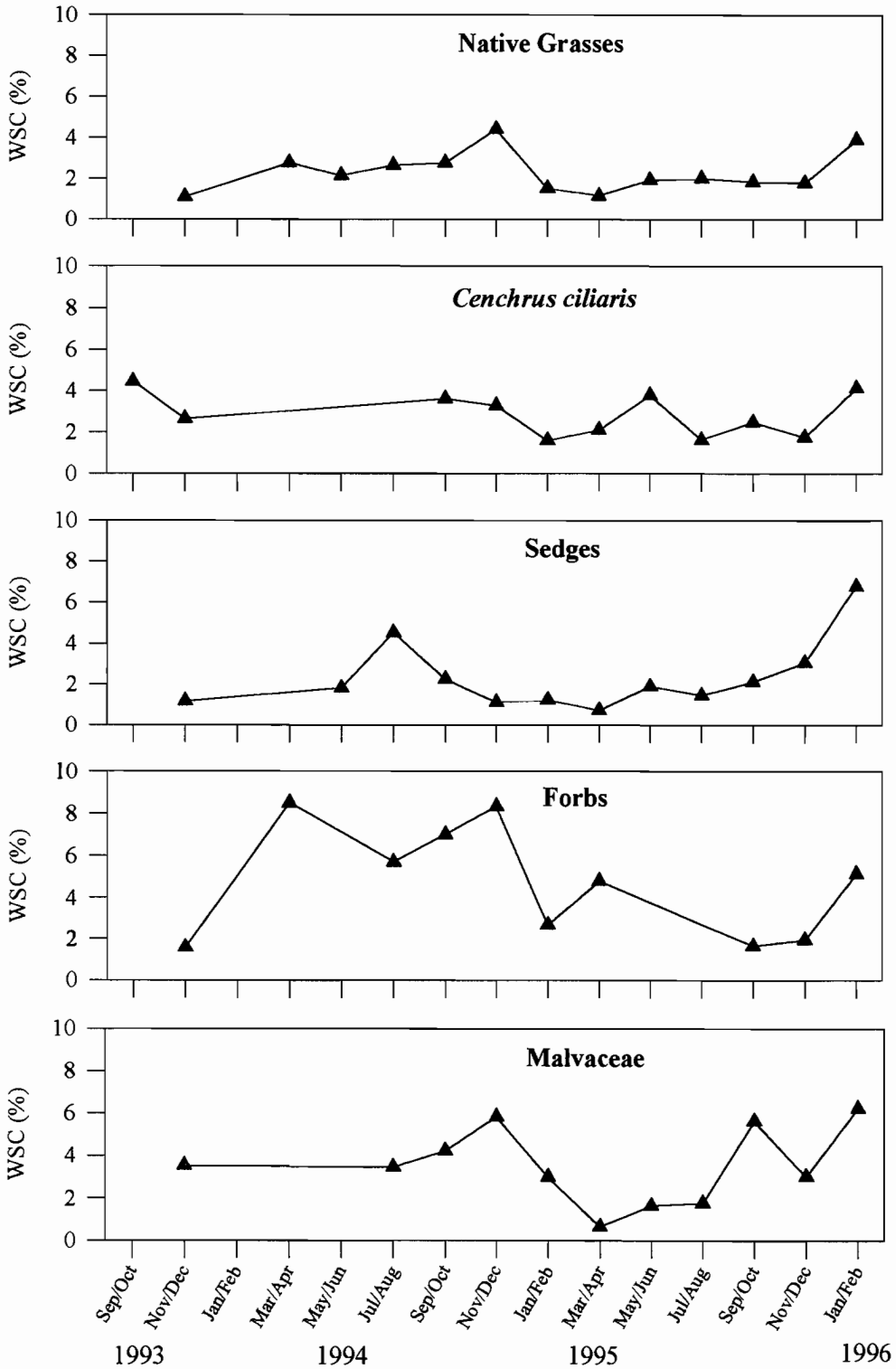


Figure 2.22. Bimonthly trends in water soluble carbohydrate (WSC) concentration of plant groups predicted by NIR spectrometry. WSC expressed as fructose equivalents as a percentage of dry matter.



month lag period (Table 2.9). In comparison, the concentration of WSC in forbs and Malvaceae was more variable (Figure 2.22). The concentration of WSC in Malvaceae correlated in a positive trend following rainfall, whereas forbs showed a similar trend after a lag of 10 months. Again, the variation in the concentration of water soluble carbohydrates between groups and in response to rainfall suggests the different growth patterns and functional significance of WSC's of each plant group.

A Comparison with Other NIRS Studies

Generally, the attributes of forage quality predicted by NIRS in the current study are comparable to other reported studies using NIRS methodologies (Table 2.10). The mean values for total nitrogen content and IVDMD of both monocotyledons and dicotyledons were lower in the current study than the mean values of other studies. Conversely, the mean values for NDF, ADF and AL of monocotyledons were all higher than the mean values of other studies. These suggest that the quality of forage at EFNP is lower than in other published studies. However, the other studies incorporate prepared forages, such as hays and improved pastures, which should be of higher quality than forages growing under natural conditions. Further, the other published studies using NIRS also included temperate growing plants. Temperate C_3 grasses generally have lower fibre content and the minimum concentrations of crude protein are generally higher than tropical C_4 grasses (e.g. Owen-Smith 1982). A comparison of the range for each of the attributes of quality indicates that the forage species of Epping Forest National Park are similar to other reported studies, but maybe slightly higher in fibre and lower in nitrogen.

2.5 FORAGE QUANTITY AND QUALITY: A SYNTHESIS

The NHN wombat is confronted with a dynamic heterogeneous forage resource that varies temporally in quality and quantity. Like other tropical savanna habitats (e.g. McNaughton 1979, McNaughton and Banyikwa 1994) the forage resource at EFNP rapidly responds to rainfall in quantity through primary

Table 2.10. Comparison of attributes of forage quality measured in the current study and values reported in other studies using NIRS. Attributes of forage quality reported include total nitrogen concentration; neutral detergent fibre (NDF); acid detergent fibre (ADF); acid lignin (AL); organic matter (OM); *in vitro* dry matter digestibility (IVDMD); and water soluble carbohydrates (WSC). All attributes of forage quality are expressed as percentages of dry matter. Mean data for the current study is reported with one standard deviation along with the number (N) of samples of each forage type (monocotyledon or dicotyledon). Mean data for the published data is the grand mean of all reported means.

Attribute	Current Study						Published Studies					
	Monocotyledons			Dicotyledons			Monocotyledons			Dicotyledons		
	Range	Mean	N	Range	Mean	N	Range	Mean	Studies	Range	Mean	Studies
Nitrogen	0.27 - 2.52	0.91 ± 0.50	106	0.69 - 2.92	1.72 ± 0.59	28	0.39 - 5.01	1.85	1, 2, 3, 4, 5	0.39 - 4.59	2.25	2, 6
NDF	54.91 - 86.84	74.93 ± 5.53	106	37.38 - 75.89	54.77 ± 9.76	28	23.60 - 78.75	58.08	1, 2, 3, 4, 5	22.5 - 85.0	46.6	2, 6
ADF	30.44 - 56.09	43.01 ± 4.02	106	24.10 - 51.28	38.21 ± 6.45	28	15.8 - 47.0	30.68	1, 3, 4	12.7 - 54.1	31.5	6
AL	3.71 -15.00	9.08 ± 2.18	106	4.01 - 19.71	11.47 ± 4.50	28	2.48 - 15.70	6.01	1, 3, 4, 5	3.0 - 26.9	13.9	6
OM†	75.24 - 99.27	91.46 ± 4.36	106	73.40 - 95.75	88.14 ± 6.70	28	86.9 - 93.2	89.94	7, 8	82.6 - 98.3	93.8	6
IVDMD	22.64 - 65.13	36.83 ± 8.15	106	31.18 - 84.53	53.14 ± 13.26	28	22.2 - 80.2	62.31	2, 3, 5	28.2 - 82.0	65.85	2, 6
WSC	0.16 - 6.79	2.38 ± 1.26	106	0.66 - 9.68	4.24 ± 2.55	28	-	-		-	-	

Studies: 1. Norris *et al.* (1976); 2. Smith and Flinn (1991); 3. Aastveit and Marum (1993); 4. García-Ciudad (1993); 5. Rabotnikof *et al.* (1995); 6. Meuret *et al.* (1993); 7. Adu and Adamu (1982); 8. McIntosh (1966).

† Organic matter has not been examined for monocotyledons using NIRS, hence the tabulated values are from other studies reporting the organic matter content of C_4 grasses.

productivity (Table 2.2) and quality (Table 2.8) producing a variable forage resource for the NHN wombat.

The NHN wombat might be expected to consume forage of greatest nutritional reward (Schoener 1971, Belovsky 1984, Kohlmann and Risenhoover 1994). Such a diet would be high in crude protein, readily available carbohydrates and digestibility but low in dietary fibre (Robbins 1983), and be readily accessible to the NHN wombat. Rarer plant species may require greater foraging time and energy expenditure to find but abundant species may not necessarily be the most palatable.

2.5.1 Foraging Effort - Quantity

The total forage biomass at EFNP is largely determined by rainfall with primary production correlating significantly with rainfall. The total forage biomass does not appear to be a limiting resource for the NHN wombat with the current densities of herbivores. However, the biomass of the preferred leaf fraction of the plants consumed (Chapter 3) may be limited and, although not investigated in this study, warrants further investigation.

Total forage biomass ranged from 1137 kg/ha DM in the dry season of 1993 to 3332 kg/ha DM in the wet season of 1995. This range in biomass would be considered 'good' for animal production in the context of the Australian semi-arid zone, particularly grasslands where summer rainfall is dominant (Wilson 1974) and forage has not been improved by fertilisers (e.g. Evans 1992).

The range of total biomass compared favourably with other tropical savanna ecosystems. For example, in the Serengeti-Mara forage genera are similar to those of EFNP. Biomass ranges from 200 kg/ha in the dry season to inexcess of 2000 kg/ha in the wet season (McNaughton 1979). However, the EFNP habitat lacks the grazing pressure of a substantial biomass of herbivores.

The consistently high biomass of grasses throughout the year has important implications for diet selection by the NHN wombat. In general, the relationship between the rate of intake of forage and forage availability is asymptotic; that is,

when forage availability is moderate to high, the rate of intake of forage is relatively constant but intake declines rapidly as forage availability declines (Trudell and White 1981, Short 1987, Demment and Greenwood 1988). The consistently high biomass of grasses at EFNP throughout the year should therefore enable the NHN wombat to maintain relatively constant rates of intake.

The structure and quality of the forage can greatly influence forage intake (Chacon and Stobbs 1976, Trudell and White 1981, Short 1987, Demment and Greenwood 1988, Hanley 1997). Bite size of a herbivore eating grass may vary with forage biomass, which in turn may correlate with dry-matter intake (Wickstrom *et al.* 1984, Hanley 1997). Bite size is also interrelated with gut-fill, rate of passage and processes of digestion (Hanley 1997). For wombats, small bite sizes will reduce the size of the particles ingested and maximise cellulose digestion, but reduce the rate of dry matter intake. The high availability of forage at EFNP suggests that the NHN wombat could maintain a relatively constant rate of intake of forage throughout the year.

The structure of the forage in NHN wombat habitat at EFNP, although not measure, is variable both temporally and spatially. Forage species vary in height growth stage and density both within and between species. Further, ground cover varies significantly with time and spatially (Table 2.3). How the NHN wombat responds to the heterogenous habitat on a fine scale, such as patches, is unknown. Behavioural responses to changes in forage quantity and quality on a broad scale have been examined by Johnson (1991a) and are further examined in Chapter 5.

The biomass and species composition of the wombat's foraging environment was dominated by *Cenchrus ciliaris* throughout the year (Figures 2.11&2.12). However, this was partly an artefact of the morphology of *C. ciliaris* compared to native grasses, sedges and forbs. *Cenchrus ciliaris* maintains an impressive above-ground biomass with the core of the plant often consisting of scenesced leaves and stems. The dry weight rank estimates of the BOTANAL technique tends to overestimate forage species that maintain high biomass over forage species that maintain less biomass (Tothill *et al.* 1992, Catchpoole and Wheeler 1992). Therefore, the results of this study may overestimate the

contribution of *Cenchrus ciliaris* to the total species composition in comparison to native grasses, forbs and sedges.

2.5.2 Foraging Effort - Quality

Many other studies have shown that grasses contain high concentrations of fibre and are poorly digestible compared to other plants (e.g. Van Soest 1964 & 1965, Olubajo *et al.* 1974). This study found that the clipped grasses were both high in fibre and low in digestibility. Further, the concentration of fibre, N and WSC in grasses varied throughout the year at EFNP. Variation in the nutritional constituents of forage species may result in selective consumption of forage by NHN wombats. If NHN wombats choose forages with high N, IVDMD and WSC but with low fibre, then forbs may be eaten if they have low concentrations of secondary chemicals. However, the sampling procedures in this study examined whole plants rather than differentiated analysis of plant parts. The interpretation of the effect of plant quality on foraging decisions by the NHN wombat may therefore be subjected to some level of error. This is because the perceived nutritive value of the forage by the wombat may be different to the measured nutritive value of the whole plant if the wombat is specifically selecting plant parts.

Of all the attributes of forage quality, trends in total nitrogen concentration are considered the most important in tropical savannas (Owen-Smith 1982). The range of N concentration of grasses at EFNP is 0.32% to 2.28% with pronounced seasonal variation (Figure 2.16). This range is comparable to other tropical grasses (e.g. Owen-Smith 1982, Georgiadis and McNaughton 1990), although peak N concentrations do not reach the 4.90% N recorded by Georgiadis and McNaughton (1990).

The minimum N concentrations were sometimes below the critical value of 0.8% N (Owen-Smith 1982), where the metabolic losses of nitrogen by grazing cattle would exceed intake. This may be of concern for the NHN wombat, but Barboza *et al.* (1993) demonstrated that wombats have one of the lowest known dietary nitrogen requirements. Further, Barboza (1989) found that wombats began to enter negative N balance and slowly lost body mass on diets of 0.6% N. The minimum N requirements for wombats were 201 $mg.kg^{-0.75}.d^{-1}$ for a 30kg SHN

wombat at an intake of $30g.kg^{-0.75}.d^{-1}$ or a projected minimum dietary concentration of 0.67% N (Barboza *et al.* 1993). The nitrogen concentration of the clipped forage at EFNP falls below even this low level. While the minimum requirements for dietary nitrogen for NHN wombats are unknown, it is likely they are similar to those reported by Barboza *et al.* (1993) for SHN wombats and common wombats. Therefore, the potential exists that the NHN wombat may enter nitrogen deficit if the nitrogen concentration of principal grass species consumed by the NHN wombat falls below a critical concentration. The potential impact on body condition and body composition of the NHN wombat are investigated in Chapter 4.

A further consideration of diet choice by the NHN wombat is the digestive quality of the forage. Hirakawa (1997) defines digestive quality as "the digestible energy content per gross intake per digestive time". The digestive quality incorporates ingestive behaviours (e.g. search time, bite rate, chewing rate) and digestive processes (Demment and Greenwood 1988). A key consideration in Hirakawa's (1997) concept of digestive quality is the fibre content of the forage, which will affect both ingestion (Stobbs 1973a&b, Demment and Greenwood 1988) and digestion processes (Hume 1989, Illius and Gordon 1993).

Barboza (1989) examined forage intake and fibre digestion in both the SHN wombat and the common wombat fed a captive diet of barley straw and maize with nutritional supplements. He found that neither wombat species altered rates of intake of dry matter or rates of passage when the fibre (NDF) concentration of the forage increased from 44% to 59%. However, the dry matter digestibility of forages with low fibre concentration (44% NDF) was higher than forages with high fibre concentration (59% NDF). Barboza's (1989) category of 'high fibre' (59% NDF) was probably an underestimate in comparison to forage available to SHN wombats and common wombats in their natural habitat (Table 2.11). Indeed, Barboza (pers. comm.) experimented with diets containing of up to 73% NDF and found similar results despite some problems in both making the pellet and feeding the pellets to *Lasiorhinus latifrons*. Even so, Barboza (1989)

Table 2.11. Comparison of seasonal variation in attributes of forage quality for grass species consumed by the three wombat species.

Attribute (% of Dry matter)	NHN Wombat			SHN Wombat		Common Wombat		
	⇐SEASON ⇒							
	Wet Season	Early Dry	Late Dry	Growing Season	Senescent	Summer	Autumn	Winter
Organic Matter	92.2	92.3	89.5	93.0	90.2	87.9	90.4	85.7
Neutral Detergent Fibre	75.7	77.1	75.6	66.5	79.8	61.7	69	57.8
Acid Detergent Fibre	42.9	43.8	43.7	30.8	53.3	32.3	38.6	32.2
Acid Lignin	7.3	7.7	7.7	4.5	8.0	1.9	4.5	1.8
in vitro Digestibility	35.7	34.5	34.9	-	-	-	-	-

Northern Hairy-nosed (NHN) wombat data: this study; Southern Hairy-nosed (SHN) wombat and Common wombat data from Barboza (1989).; Data presented by Barboza (1989) for the Common wombat is from McIllroy (1973).

clearly demonstrated that the Vombatidae have comparatively higher rates of digestion of high fibre diets than other colonic fermenters (e.g. Orton *et al.* 1985).

The level of complexity related to the foraging decisions of the NHN wombat, increases when nutritional variation within a plant is considered. For example *Cenchrus ciliaris* leaves show elevated concentrations of total nitrogen and higher levels of IVDMD compared to sheath and stems (Appendix 1). Conversely the stem of buffel grass can have considerably higher concentrations of water soluble carbohydrates than leaves and sheaths. Also the stems of *C. ciliaris* are higher in organic matter, a likely consequence of the high cellulose in the stem or the lack of siliceous hairs on the sheath protected stem. This trend is consistent with the plant part nutritional variation encountered by SHN wombats (Dierenfeld 1984).

2.5.3 Conclusions

Forage selection by the NHN wombat should not be limited by the total biomass of available forage. Instead, the nutritional quality of the forage and limited quantity of native forage species are likely to be determinants of forage choice by the NHN wombat. The biomass, species composition and species frequency are dominated by *Cenchrus ciliaris*, with native monocots and forbs being comparatively less frequent. Consequently, selectivity for preferred species (other than *C. ciliaris*) could result in temporal variation in ranging activity as quality and quantity of the forage environment changes. Selection of plant species is most likely to be influenced by the nutritional quality and morphology of the plant. Within the plant, specific parts may be favoured due to nutritional variability of plant parts. Overall, the nutritional quality of the forage plants available to the NHN wombat is poor. The potential for the NHN wombat to be as efficient as the SHN wombat and common wombat in digestive function, nitrogen maintenance, energy and water conservation would be vital adaptations for foraging on such a temporally variable forage resource. These adaptations are particularly significant for all wombat species because they can not easily migrate to areas of higher forage quality when forage resources become limiting.

2.6 NEAR INFRARED SPECTROSCOPY: ECOLOGICAL APPLICATION

This study has demonstrated that broad-based NIRS predictive equations can be used to predict attributes of forage quality. Further, the predictive equations generated in this study have immediate benefits as a management and research tool to be used in future pasture studies at Epping Forest National Park. More importantly however is the potential of NIRS technology and the application of broad-based predictive equations to ecological studies examining plant ecology or plant-herbivore interactions.

This study has further demonstrated that robust predictive equations for attributes of forage quality (N, OM, NDF, ADF, AL, IVDMD and WSC) can be developed and applied to a natural habitat of high species diversity. This is distinct from other studies using NIRS where the forage assessed has either been from a semi-natural habitat or cultivated forages (Table 2.5). The application of predictive equations developed in this study has allowed successful measurement of forage quality for twenty-five plant species, representing perennial grasses, annual grasses, sedges and forbs encompassing a three year sampling regime across each season. Further, the predictive equations can predict the nutritional quality of plant parts (e.g. attributes of forage quality for Leaf, Stem and Sheath of *Cenchrus ciliaris*; Appendix 1).

2.6.1 Cost Benefit Analysis

The benefits of NIRS have been long been known among agricultural scientists (Shenk *et al.* 1992, Shenk and Westerhaus 1994). There are two major benefits of the application of NIRS. The first benefit is the reduction of analytical cost and time without compromising precision or accuracy. Secondly, once the predictive equations have been developed, large numbers of samples can be processed. For example, an experienced operator can scan between 120 to 140 ground plant samples in duplicate per day. Applying the appropriate predictive equation will then allow rapid prediction of the attributes desired. In comparison, a technique such as NDF analysis, even with improved methods, may be able to process 48 samples in duplicate per day at best (not including weighing and drying

time). The reduction in time spent in the laboratory and reduced analysis cost could then allow ecological experimentation or ecological monitoring of a single population on a broader scale than conventional methods.

Another major benefit of NIRS is the measurement itself. Since data for each sample are recorded as spectra, the measurement is non-destructive. Therefore there are no expensive chemicals or hazardous by-products in measurements. In addition, spectra can be measured on relatively small samples (e.g. seagrass samples weighing less than 5 g dry matter; Aragones 1996), allowing many attributes to be measured on samples for which only one or two attributes could be measured by conventional methods.

2.6.2 Statistical Robustness of Equations

The collection of spectra are subjected to two major influences: the environment during collection (temperature and humidity) and the particle size of the samples (Shenk and Westerhaus 1991, Shenk and Westerhaus 1992, Baker *et al.* 1994). To obtain repeatable and comparable spectra, standardising the collecting environment becomes critical. By standardising samples prior to measurement and the collecting environment, potential problems associated with sample moisture are reduced (Baker *et al.* 1994). Likewise, by applying standard normal variate and detrend to correct for scatter, the effect of variation in particle size is reduced and statistical models become more robust (Shenk and Westerhaus 1991, Shenk and Westerhaus 1992, Baker *et al.* 1994).

The multivariate techniques of PCR, PLS and MPLS are the recommended standards for quantitative analysis techniques for NIR spectra (Anon 1995). Each of these multivariate techniques aim to reduce the spectra to linear functions of the sample absorptions (Anon 1995). Modified partial least squares produced the best predictive models for each forage quality attribute (Table 2.7), supporting the findings of Baker *et al.* (1994) and the recommendations of Shenk and Westerhaus (1991 & 1994). The MPLS technique appears to be advantageous as most of the spectral data are reduced into relatively few linear combinations (Baker *et al.* 1994).

The MPLS model for each forage quality attribute was validated by cross-validation producing a measurement of the standard error of the cross-validation (SECV), standard error of the calibration (SEC) and a measurement of precision (either the correlation coefficient and/or 1-VR). Estimates of SECV, SEC, r^2 and 1-VR compare favourably to other studies (Table 2.5, Shenk and Westerhaus 1992). The values of SECV and SEC increase as the model's ability to predict the forage quality attribute decreases and the slope of the observed laboratory data versus values predicted by the NIRS moves away from one.

The predictive model for each attribute of forage quality varies in precision. The predictive model for total nitrogen is very precise because of the high occurrence of nitrogen-carbon bonds in plant material. Other attributes such as AL are comparatively low in precision but can still predict more than 77% of the variation. The decrease in precision for an attribute such as AL is in part due to the relatively complex structure of cellulose compared with the simple X-H bonds as described by Shenk *et al.* (1992). Further contributing to the reduced precision of AL is the sequential fibre analysis used as the reference method. Potential errors in the sequential method will be magnified in AL compared to NDF. However, the reduction of precision is not significant and the predictive ability is still very useful in broad-scale ecological studies.

The precision of the predictive models can be constantly refined and improved. Increasing the sample size of the calibration set will improve each model by reducing the effect of outliers. Further, increasing the sample size will improve the ability of the model to predict a diverse range of species and temporal variation. Consequently, NIRS models can be continuously improved to suit the function and purpose of the model.

2.6.3 Future Application

The potential application of NIRS to aspects of ecological research seems limitless, particularly in plant-herbivore systems. As well as studies of forage quality like this one and others (Table 2.5), NIRS has been applied to other aspects of grassland studies including measurement of water use-efficiency estimated from $\delta^{13}C$ discrimination (e.g. Clark *et al.* 1995) and measurement of botanical

composition of mixed species hay harvests (e.g. Coleman *et al.* 1985, García-Ciudad *et al.* 1991, García-Ciudad *et al.* 1993). NIRS has also been applied to animal production studies to predict rates of intake (e.g. Flinn *et al.* 1994, Poppi 1996), and estimate botanical composition of diet from oesophageal extrusa (e.g. Volesky and Cleman 1996) or faeces (e.g. Lyons *et al.* 1995). Future broad-scale applications may include measuring botanical composition of free-ranging herbivore diets calibrated by techniques such as $\delta^{13}C$ discrimination or long-chain alkanes, estimating rates of intake of free-ranging herbivores, with excellent potential for application in the field of herbivore-antifeedants and quantification of secondary metabolites produced by plants (Foley *et al.* in press).

The products of this study can be specifically applied to the management and improvement of the forage environment of the NHN wombat. Near infra-red spectrometry offers the means to assess in real time changes in forage quality with minimal interference to the forage and with minimal cost. Implementation of potential pasture improvement programs, using NIRS in conjunction with a species composition, frequency and biomass monitoring technique such as BOTANAL, will be able to rapidly assess the success of future forage improvement programs under controlled experimental designs. Active evolution of the NIRS equations will improve the predictive ability of equations developed in this study.

CHAPTER 3

HERBIVORE DIET ANALYSIS: THE NORTHERN HAIRY-NOSED WOMBAT

3.1 INTRODUCTION

3.1.1 The diet of the NHN wombat

One of several key issues relating to the ecology of the NHN wombat is diet selection, which is the focus of this chapter. Norbury and Sanson (1992) describe diet selection as the "animal's choice of food from an array of different food items". Chapter 2 demonstrated that the 'array of different food items' available to the NHN wombat varies in quality and quantity in both time and space. Therefore, it seems likely that diet selection may differ between individual NHN wombats and with time. Quantification of the diet selected by the NHN wombat has important implications for understanding its feeding strategy and practical applications for conservation management.

Flößer (1986) and Crossman (1988) have examined the diet of the NHN wombat, but in limited detail and with differing results. The differences between these studies in their estimation of diet composition in the NHN wombat is of some concern, especially since there was overlap between them in the dates when faeces were collected (Table 3.1). This may represent either spatial heterogeneity between the collection sites in the two independent studies or potential errors in the analytical methods. Both explanations require further investigation.

Flößer (1986) demonstrated that the diet of the NHN wombat consisted primarily of native grasses, with *Eragostris lacunaria*, *Chrysopogon fallax*, *Heteropogon contortus*, *Fimbristylis* spp., and *Aristida* spp. being the principal species of the diet (Table 3.1a). Flößer (1986) concluded that although the NHN wombat preferred specific plant species, it was not a specialist feeder. This study

Table 3.1. Diet of the Northern Hairy-nosed wombat from two independent studies using histological diet analysis techniques. The study by **a)** Flößer (1986) quantifies the mean botanical composition of NHN wombat faeces from seven burrows distributed throughout the NHN wombat habitat. The study by **b)** Crossman (1988) describes only the frequency of occurrence of plant species in twenty NHN wombat faecal pellets from two collection sites and two collection times, but does not quantify botanical composition or frequency. For example *Aristida* spp. was present in eleven of the twenty faecal samples examined. Both studies represent only one sampling period.

Plant Species	**a)** Flößer (1986) Dec '82 to Feb '83 Species Composition (%)	**b)** Crossman (1988) Nov '82 and Apr '83 Species Frequency (%)
Aristida spp.	4.29	55
Bothriochloa ewartina	0.83	-
Brachiaria spp.	1.11	-
Cenchrus ciliaris	0.89	20
Chrysopogon fallax	5.37	-
Dactyloctenium radulans	0.60	-
Dicanthium sp.	-	0
Digitaria brownii	0.34	-
Enneapogon spp.	1.74	50
Enteropogon ramosus	1.23	25
Eragostris lacunaria	14.14	0
Heteropogon contortus	3.97	80
Panicum sp.	-	5
Paspalidium sp.	-	-
Perotis rara	1.46	-
Seteria australiensis	0.20	-
Sporobolus caroli	-	50
Tragus sp.	-	0
Sedges	-	10
Fimbristylis spp.	5.46	-
Unidentifiables	58.37	-

was limited to one sampling period (December 1982 to February 1983) and a limited number of burrows (seven), and did not follow temporal variation in plants consumed by the NHN wombat. Further, Flößer's (1986) conclusion that the NHN wombat was a generalist feeder cannot be confirmed because the availability of grasses was not quantified. This requires further investigation.

The study by Crossman (1988) described the frequency of occurrence of selected plant species in the diet (Table 3.1b), and concluded that native grasses comprise the bulk of the NHN wombat's diet. However, these data differ markedly from those of Flößer (1986), despite similarities in the methods used to quantify the diet. By examining total nitrogen, ash and fibre content of NHN wombat faeces between August 1980 and October 1983, Dierenfeld concluded that the quality of diet changed considerably during this period (unpublished correspondence with Department of Environment). Chapter 2 demonstrates that total nitrogen, ash and fibre vary with season and rainfall. Therefore, it is unclear if the temporal variation in the quality of the consumed forage is representative of temporal changes in the species composition of the diet or nutritional variability of the available forage. This warrants further investigation.

Neither study investigated spatial variation in the diet of the NHN wombat across its range. Flößer (1986) collected NHN wombat faeces from seven burrows located in the centre of the range. Likewise, Crossman (1988) collected twenty faecal samples from two locations (10 samples at each) in November 1982 and April 1983. Although the diets described by Flößer (1986) and Crossman (1988) probably represent forage species consumed by a number of individual NHN wombats, the spatial heterogeneity of the habitat potentially presents individuals with different choices of forage species. By quantifying the species consumed across the range of the NHN wombat the importance of spatial heterogeneity and forage selection can be resolved.

Because the NHN wombat is critically endangered, a comprehensive account of the botanical composition of its diet is necessary particularly for conservation management. One of the objectives of the recovery process is to

establish an independent and geographically-separate population. The selection of a suitable introduction site would be greatly aided by determining the principal foods consumed by the NHN wombat at Epping Forest, variation in forage selection between individuals and temporal variation in the species composition of the diet. Further, the species composition at Epping Forest National Park is constantly changing (Figure 2.8) and it is therefore important to examine how the NHN wombat responds to these changes.

This chapter assesses three different techniques for analysis of diets of the NHN wombat and, in less detail, that of the eastern grey kangaroo, *Macropus giganteus*. Chapter 6 describes the diet of the eastern grey kangaroo in detail and estimates dietary overlap between the NHN wombat and eastern grey kangaroo.

3.1.2 Review of techniques

Quantifying the diet of the NHN wombat has implications far-reaching beyond this study, particularly in conservation management. Clearly some techniques for forages consumed by a herbivore are not practical for a critically endangered species. These include invasive techniques such as examining mouth contents, fistula contents or stomach contents. The nocturnal habits of the NHN wombat, coupled with its low stature and the prevailing vegetation structure of its habitat make direct observations of feeding very difficult. Also, the impact of NHN wombat and other herbivore grazing on the forage at Epping Forest National Park is minimal (Chapter 2) reducing the effectiveness of the utilisation approach which compares reduction of biomass over time and attributes this reduction to grazing activities.

All species of wombat deposit groups of approximately five individual faecal pellets, combined with secretions from the cloacal glands, in the immediate vicinity of the burrow as a means of marking (Triggs 1988, Hamilton 1997). These deposits are obvious and make faecal pellet analysis the preferred option for quantifying the ingested herbage by wombats because they are easy to find and collect. Nonetheless, although several methods are available to determine diet from faecal samples, all suffer from biases due to differential digestibilities of plant species (Stewart 1967, Norbury and Sanson 1992; see Appendix 1 for IVDMD).

There are three principal techniques to estimate the composition of the diet from faecal residues: stable isotopes; microscopic techniques; and the use of long-chain alkanes. The following sections discuss the merits of these techniques and where appropriate, describes the influence of differential digestibilities on these techniques.

3.1.2.1 Stable Isotopes

Stable carbon isotopes can be used to trace the flow of energy in an ecosystem (Tieszen *et al.* 1983). Differences in the photosynthetic carbon reduction pathways result in distinct differences between the ratio of ^{13}C to ^{12}C (termed $\delta^{13}C$) in C_3 and C_4 plants (Tieszen *et al.* 1983, Tieszen and Boutton 1988). In tropical savanna ecosystems, C_4 plants are represented by grasses and some forbs, and C_3 plants by tropical legumes, forbs and woody plants (browse) (Jones *et al.* 1979, Tieszen *et al.* 1979, Ambrose and DeNiro 1986). From an examination of the $\delta^{13}C$ of faecal material or body tissue, the contribution of C_3 and C_4 plants to the diet can be established (DeNiro and Epstein 1978, Tieszen *et al.* 1983).

Since plant cell-wall material forms the major source of carbon intake by herbivores, measuring $\delta^{13}C$ in faeces may underestimate the contribution of plants with higher digestibilities. The alternative source of carbon, body tissue, offers a source of assimilated carbon. However, measurements of $\delta^{13}C$ from body tissue underestimate the contribution of consumed plants of low digestibility.

While animal tissue presents some bias in dietary studies of herbivores, it should be the preferred source of carbon for $\delta^{13}C$ analysis. Some types of body tissue are analytically more representative of the ingested $\delta^{13}C$ value than others depending on the metabolic activity of the tissue (e.g. hair samples are metabolically less active than lipid samples Tieszen *et al.* 1983). However, destructive sampling or biopsy samples have clear limitations for endangered species. Hair provides an alternative but problems may arise because of fractionation and turnover of the isotopes of carbon. The process of hair growth enriches $\delta^{13}C$ by as much as 1.0‰ relative to a diet consisting solely of C_4 plants.

Further, hair has a relatively long growth period. This results in a lag time of up to two months from when the carbon was consumed to its appearance in hair samples (Tieszen *et al.* 1983). Apart from these few relatively minor shortcomings and the issues of differential digestibility, $\delta^{13}C$ remains the only accurate means of diet analysis, but its precision is reduced to distinguishing the contribution of C_3 and C_4 plants to the diet (assuming Crassulacean Acid Metabolism plants are absent from the plant-herbivore system or are not consumed by the herbivore).

3.1.2.2 Microscopic Techniques

Analyses of herbivore diets have generally been based on histological methods to examine of epidermal fragments of either stomach contents (e.g. Ellis *et al.* 1977, Norbury 1988a, Vernes 1995) or faecal samples (e.g. Stewart 1967, Dawson and Ellis 1979, Copley and Robinson 1983, Hollis *et al.* 1986, Ellis *et al.* 1992, Dawson *et al.* 1992, Horsup and Marsh 1992, Evans 1992, McIllwee 1994). Faecal samples are usually easier to obtain and their collection is not harmful to the animal. The principle of histological analysis is described in detail by Holechek *et al.* (1982). Briefly, it relies on the identification of plant tissue, usually plant epidermis, in a prepared sample of faeces or stomach contents. The diet of the herbivore can then be described by the relative proportions of identifiable epidermal fragments of each species or plant group. However this creates a problem since the proportion of identifiable epidermal fragments is not consistent with the proportion of unidentifiable fragments. Coupled with differential digestibilities, an unavoidable bias is always present in histological techniques (Barker 1986a&b, Norbury 1988b). Subtle variations to the methods used to score the relative abundances (e.g. frequency of occurrence; number of occurrence; and the area covered) have all been used in studies to improve the estimate of the composition of the diet (Norbury 1988b), but still cannot fully overcome this problem. The main advantage of histological techniques over other techniques is that it is relatively simple and does not require expensive equipment. However, it is very tedious and time consuming.

3.1.2.3 Long-chain Alkanes

The use of plant wax alkanes as a method to examine the botanical composition of consumed herbage is relatively recent (Dove and Mayes 1991, Dove and Mayes 1996, Newman *et al.* 1995, Dove and Moore 1995, Dove *et al.* 1996). Currently, the technique is being applied to several ecological studies of free-ranging herbivores such as moose, mountain hare and deer (Taylor 1994).

In brief, the alkane method takes advantage of the taxonomic differences in the concentrations of long-chain alkanes in the epidermal wax of flowering plants (e.g. Eglinton *et al.* 1962, Dyson and Herbin 1968, Herbin and Robins 1969, Dyson and Herbin 1970). Long-chain alkanes, with carbon chain lengths ranging from C_{25} to C_{37}, are relatively indigestible by herbivores as they are not part of the digested matrix of ingested plant material (Dove and Mayes 1991). Although not fully recovered in faeces, the rates of recovery are high and are dependent on chain-length (Mayes *et al.* 1986, Dove and Mayes 1991). Rates of recovery can be adjusted by correction factors. Further, the rates of recovery of long-chain alkanes generally do not differ between plant species making them ideal as quantitative internal markers (Mayes *et al.* 1986, Dove and Mayes 1991). Gas chromatography can quantify long-chain alkanes in faeces and the plant species on offer. Estimation of the composition of the diet is based on least-squares optimisation procedures which find that combination of plant alkane fingerprints which best explain the alkane concentrations observed in the faeces (Dove and Moore 1995).

This study of the diet of the NHN wombat offers an ideal opportunity to test the long-chain alkane method on a free-ranging herbivore in order to estimate the composition of the diet of the NHN wombat.

3.1.3 Assumptions for Herbivore Diet Analyses

As with any study of diet selection, it is necessary to begin with a number of assumptions. The first assumption is that forage availability measured by a technique such as BOTANAL (Chapter 2) is the same as that perceived by the herbivore (Norbury and Sanson 1992). This includes the assumption that all parts of the plant are equally available. However, for herbivores, such as wombats, that uses olfaction as the dominant form of social interaction and spatial assessment of

their environment, grass inflorescences may play a role in forage selection. If so, there may be differences in the measured availability of forage by an observer and perceived availability by the wombat (Norbury and Sanson 1992).

The second assumption is specific to faecal analysis techniques where the herbivore deposits more than one faecal pellet in a defecation event. It assumes that all faecal pellets in a pellet group are representative of one foraging bout, and the botanical composition of one pellet is the same as every pellet in the group. This is a gross generalisation, with mixing of ingesta occurring along the digestive tract (Stevens and Hume 1996). Therefore one faecal pellet may represent a number of foraging bouts. Further, there may be non-uniform mixing of ingesta and differential digestion resulting in variability of epidermal residues from one faecal pellet to another in a single defecation. This is however unlikely because of the retention time of ingesta is relatively long but random flow may cause irregular mixing through the release small pockets of ingesta (Barboza 1989).

The final assumption is specific to histological analysis. It assumes that the identified epidermal fragments of each plant species are encountered in the same proportion as the unidentified fragments of each species. This assumption does not consider that histological analysis is based on identification of non-digested plant material consumed by the herbivore. Further, differential digestibility of plant material, particularly of different parts of the plant reduces the probability that the identifiable epidermal fragments occur in the same proportion as the unidentifiable fragments. Although invalid, this assumption does however allow results to be used in the calculation of selection indices where the estimated composition of the diet needs to be summed to unity (or 100%).

3.2 METHODS

3.2.1 General Methods

Collection of faeces

Selection of NHN wombat faeces for analysis was based on two criteria: burrow activity ranking and distribution within the habitat. Burrows were allocated a rank based upon activity surveys of burrows (Horsup, Johnson and Crossman

unpublished data). Activity surveys score each burrow as either active, visited or inactive based on evidence of activity by NHN wombats. To determine 'core' burrows, each burrow was given a rank of between zero and six based upon the number of 'active' scores from the six most recent activity surveys available at the time of calculation. A score of six represented a burrow that was scored as 'active' for each of the six activity surveys sampled, and zero represented inactivity. Ranks of six and five were deemed 'core' burrows that were almost continually occupied or at least visited by NHN wombats regularly. Consequently, faecal samples were first selected if they came from a core burrow and then so that they encompassed the entire range of the NHN wombat.

Northern hairy-nosed wombat faecal pellet groups were collected at bimonthly intervals for three years (May 1993 to June 1996) from 'core burrows. Faecal pellet groups were collected if they appeared to be recently deposited, characterised by a green appearance and/or the presence of the sticky cloacal secretion. Samples were placed in paper bags, and allowed to dry-out. Upon return to the laboratory samples were oven-dried at 60°C for 24 hours and stored at -20°C until analysis.

Preparation of Faeces

In general, six NHN wombat faecal pellet groups and two eastern grey kangaroo faecal pellet groups were used for analysis for each bimonthly collection. Each pellet group, consisting of between four to ten faecal pellets, was sub-divided so that there was one pellet for carbon isotope analysis, one for microscopic analysis and the remaining pellets for alkane analysis. Faecal pellets and reference plant samples used for carbon isotope analysis and alkane analyses were ground in a cyclone sample mill (Udy Corporation, Fort Collins, Colorado) to pass a 1-mm screen. Microscopic analysis was performed on the maximum number of samples able to be efficiently processed (based upon time and budget) and consequently focused upon the faecal material collected in the first twelve months of field sampling.

3.2.2 Carbon Isotope Analysis

Carbon isotope analysis was carried out at CSIRO Division of Tropical Agriculture and the Department of Botany, University of Queensland using the methods described by LeFeuvre and Jones (1988). One-way ANOVA was used to examine differences in $\delta^{13}C$ between sampling periods.

3.2.3 Conventional Microscopic Analysis

Plant Reference Collection

A comprehensive reference collection of the epidermal patterns of forty plant species available to the NHN wombat and eastern grey kangaroo was prepared. Within each plant species, epidermal samples were taken from as many different parts of the plant as possible - generally 3 or 4 for each species. The categories include leaf, stem and sheath. These were often sub-divided, depending upon plant morphology, to include leaf sheath, stem sheath, the stem inside the stem sheath, and the base of the leaf blade.

Preparation of the epidermal reference collection followed the method of Evans (1992) with slight modification. Briefly, epidermal samples were prepared by separating epidermal cells from the underlying mesophyll cells by heating small sections of plants in solution of 10% chromic acid (50%) and nitric acid (50%). Separated epidermal fragments were then washed in distilled water and stained with gentian violet and finally mounted semi-permanently in a glycerol-based mounting medium on a microscope slide with coverslip (Evans 1992). Photomicrographs were then taken at varying magnifications to best illustrate epidermal patterns present in each sample.

Faecal Material

Faecal material was prepared following a modification of the procedure of Evans (1992). Faecal pellets were broken into pieces and soaked overnight in a solution of commercial bleach (20% sodium hypochlorite). The bleach was then removed and the faecal material rinsed with distilled water over a 2 μm filter under gentle vacuum. All faecal material was retained. Two subsamples were then stained with gentian violet and evenly spread across microscope slides in glycerol-based mounting medium.

Microscopic Analysis

Analysis of faecal material followed a modification of Barker (1986b). The slides were examined at 100X magnification using an Olympus BH-2 compound light microscope. Systematic sampling occurred along three longitudinal transects per slide at 5 mm intervals (equating to just greater than one field-of-view) and vertical transects separated by 5 mm. A total of 24 fields-of-view were scored per slide and two slides analysed per faecal sample. Systematic sampling is preferred to random sampling as it is more precise (Norbury 1988b). Within each field of view, fragments were categorised into proportions. This allowed faster quantification of epidermal residues without compromising precision (Barker 1986b). Barker's (1986b) original categories of none (zero), a little (one), some (two), or a lot (three) were modified to none (zero), a contribution of epidermal fragments of a particular species that consists of less than 25% of the field of view (one), between 25% and 50% of the field-of-view (two), between 50% and 75% of the field-of-view (three), and above 75% of the field-of-view (four). Category scores were summed, with the observed composition of the estimated diet described as a proportion of all scored fragments. Any error associated with the estimation of one species will therefore be reflected in all species (Barker 1986).

In conjunction with describing the species composition of the epidermal residues, plant-parts were also quantified. Each identifiable epidermal fragment was allocated to a specific part of the plant, including a category of 'unknown'. The contribution of each plant-part within a species was expressed as a proportion of the total epidermal fragments identified for only that species.

One-way ANOVA was used to assess whether there was temporal variation in the diet of the NHN wombat using the microscopic technique. The October 1993 sampling period was used to asses differences in the diet consumed by individual NHN wombats. Faecal samples were collected from twenty-one NHN wombat burrows, encompassing the entire range of NHN wombat. Variation between collection sites (burrows) was assessed by ANOVA. To estimate accuracy of the microscopic technique, two independent observers scored the same faecal

samples for the October 1993 sampling period. One-way ANOVA was used to test whether differences between the two observers was important.

Selection Index

Selection indices, or electivity indices, have been used to quantify the degree of selection a herbivore has for specific food items (Lechowicz 1982, Norbury and Sanson 1992). The value and problems associated with several indices have been comprehensively reviewed by Lechowicz (1982). Norbury and Sanson (1992) suggest that an index should "enable quantitative comparisons between differing levels of food availability and for a variety of different food types". By using a selection index to describe the food preferences of the NHN wombat (and eastern grey kangaroo in Chapter 6), comparisons between the Crossman (1988) study and the current study can be drawn.

Crossman (1988) described the dietary preference of the NHN wombat, eastern grey kangaroo and cattle by using Ivlev's Forage Ratio (Ivlev 1961):

$$E'_i = r_i/p_i \qquad \text{(Equation 3.1)}$$

where E'_i is Ivlev's Forage Ratio; r_i is the relative utilisation in the diet; and p_i is the availability of food types in the habitat. This index aims to determine whether food choice is non-selective, with a value of 1.0 for true non-selective feeding; less than 1.0 the food item is avoided; and above 1.0 through to infinity a preference is exhibited for the food item. Crossman (1988) found that NHN wombats, eastern grey kangaroos and cattle all selected grasses. Since E'_i for preferred items does not fit within a predefined range, comparable levels of 'preference' can not be easily made. Consequently, to make a comparison of preference between the two studies a different selection index should be used.

Lechowicz (1982) suggested that the Vanderploeg and Scavia (1979) Relativised Electivity Index as the best performing selection index. Further, it would provide a better comparison between the Crossman (1988) study and the current study than E'_i because data can be standardised. The Relativised Electivity Index is calculated using the following equations:

$$E^*_i = [\alpha_i - (1/n)]/[\alpha_i + (1/n)] \quad \text{(Equation 3.2)}$$

and:

$$\alpha_i = [r_i/p_i]/[\Sigma_i r_i/p_i] \quad \text{(Equation 3.3)}$$

where E^*_i is the Relativised Electivity Index; α_i is Chesson's alpha (Chesson 1978); and n is the number of different food types. The Relativised Electivity Index provides a measure of selection from -1.0 (representing total avoidance of food type) to 1.0 (representing total preference for food type) with a value of 0.0 representing random feeding.

To compare the study of Crossman (1988) and the current study the values of r_i and p_i need to be comparable. Consequently the calculation of E^*_i for NHN wombats and eastern grey kangaroos in both studies can only successfully be calculated for the vegetation groupings of grasses, forbs, browse, and Malvaceae and Solanaceae, and only for cases where diets have been measured by microscopic analysis. In addition, the calculation assumes that the sum of the diet is 100%. Therefore, any unidentifiable epidermal fragments are assumed to occur in the same relative proportion as identifiable epidermal fragments.

3.2.4 Long-chain Alkane Analysis

Long-chain alkane analysis was performed in collaboration with Dr H. Dove, CSIRO Plant Industry, Canberra, ACT. The method described below is that detailed in Mayes *et al.* (1986), Dove (1992), and Dove *et al.* (1996), and descriptions from S. Hoebee (CSIRO Plant Industry pers. comm.).

Extraction of Alkanes

In an alkane-free McCartney bottle, 1.2 g of ground plant or faecal material was added along with 0.2 g of an internal standard solution. The internal standard solution consisted of the alkanes C_{24} and C_{34} (approximately 500 μg of each) of known concentration. Alkanes were extracted by heating samples in 10 ml of 1 M ethanolic KOH at 85°C for three hours with occasional gentle agitation. Separation of crude-extract into two phases occurred by adding 8 ml of *n*-heptane and 5 ml of

distilled H_2O. The top phase (non-aqueous) containing *n*-alkanes was removed and placed in a glass vial and the phase separation repeated with a further 5 ml of *n*-heptane. The solvent was then evaporated from the glass vial to remove residues of ethanol and the residue re-dissolved in *n*-heptane before elution through silica gel (Kieselgel 60, 70-230 mesh, Merck, Darmstadt, Germany) to remove long-chain alcohols and pigment breakdown products (Mayes *et al* 1986).

Alkane Analysis

Alkane concentrations in purified samples were assayed by gas chromatography using a Varian 3400 gas chomatograph fitted with a Varian 8034 autosampler (Dove *et al.* 1996). Peak areas were calculated using the software package DAPA (DAPA Scientific Software, Kalamunda, WA) (Dove *et al.* 1996). The peak areas were then used to calculate alkane concentrations, by reference to the internal standards. Corrections allowed for incremental changes in the calculation of C_{24}: C_{34} in a sample. The known weight and concentration of the internal standard, and the dry matter (DM) weight of the sample, allowed alkane concentrations to be expressed as g/kg DM of sample.

To prepare the data for the least-squares optimisation procedures and estimation of the composition of the diet, the alkane concentrations of plant species on offer were corrected for simulated recovery following digestion. Alkane concentrations were corrected using the assumption that recoveries for the alkanes C_{25} to C_{36} ranged from 90% to 97% respectively, based on unpublished data for alkane recoveries in pigs (H. Dove, CSIRO Plant Industry, pers. comm.). Pigs were selected over other animals such as sheep because, like wombats, they have a monogastric digestive tract. However, two attempts were made to measure the recovery rates of long-chain alkanes during gut passage for wombats (as part of the validation experiment, described below). Both attempts were unsuccessful because the captive southern hairy-nosed wombats were housed in sand enclosures rather than metabolism cages. Consequently, values of food intake and faecal output could not be precisely quantified.

Evaluation Experiment

An evaluation experiment was conducted to assess the suitability of the long-chain alkane technique for analysis of NHN wombat and eastern grey kangaroo faecal pellets. The experiment used of faecal material collected from twenty-four NHN wombat burrows in October 1993 and plant material from ten plant species covering eight genera. Alkanes were extracted in triplicate using the methods described. Estimation of the composition of the diet used the simultaneous equation method described by Dove and Mayes (1991). In the absence of data to the contrary, alkane recoveries for the evaluation experiment were assumed to be similar to those of sheep (Dove pers. comm). Similarities or otherwise between the patterns of alkane concentrations of each species were compared by canonical variates analysis (CVA).

The twenty-four samples were the same as the samples used in the microscopic analysis for the same collection period. This allowed comparison of the two methods by ANOVA. Analysis of variance was also used to determine spatial variation in diet composition between burrows.

Mathematical Analysis

Solutions to estimate the composition of the diet of the NHN wombat were calculated by the "EatWhat" program (Dove and Moore 1995). This program uses least-squares optimisation procedures to predict the optimum solution from the alkane concentrations of the faecal samples compared to the alkane concentrations in the plant samples on offer. This method of analysis is considered statistically more robust than the simultaneous equation method of Dove (1992), as it is constrained to produce only positive solutions.

Principal component analysis (PCA) was used to identify plant species of similar alkane concentrations and the alkanes with the most variability in non-replicated samples. However, in the evaluation experiment, samples were replicated and CVA was the more appropriate analytical technique.

The least squares optimisation procedure of 'EatWhat' estimates the composition of the diet in terms of proportion of each plant species in the diet.

Each estimation of composition of the diet is summed to unity (or 100%). Further, the estimated diet is limited to the number of plant species on offer, provided they do not exceed the number of alkanes. Hence, there are several possibilities for the estimation of the composition of the diet depending on the number and diversity of plant species on offer. To provide the best possible estimate of the herbivore diet, three estimation procedures were used:

1. All plant species and all alkanes were used in the EatWhat prediction.

2. PCA was used to identify plants with similar patterns of alkane concentrations (see Appendix 2) as determined by the position of their first two principal components, with the axes about the origin used as an arbitrary divider (M. Lorimer, University of Adelaide, pers. comm.). Species with similar values for principal component (PC) 1 and PC 2 were then grouped. Only one species from each 'group' could be used in the least-squares optimisation procedure. The species chosen to be representative of the group was determined by its contribution to the diet measured by microscopic analysis. This approach is similar to that used by Salt *et al.* (1994).

3. In a manner similar to that described in (2), plant species for the 'EatWhat procedure were selected if their patterns of alkane concentrations were distinct from other plant species, and if similar they were also grouped together. However if a PCA group was represented by a C_3 plant or a plant that was considered rare in its contribution to the diet (as determined by microscopic analysis), it was eliminated from the least-squares optimisation procedure.

One major advantage of the least-squares optimisation procedure is that it can use more alkanes than plant species on offer. This allows more variability of alkane patterns to be considered than the simultaneous equation method. Determining which alkanes to include in the least-squares optimisation procedure is by PCA. Results of the principal component analyses for each bimonthly sampling period are shown in Appendix 2.

ANOVA was used to determine the differences between the alkane and histological methods in estimating the contribution of 27 plant species to the diet of the NHN wombat. Since the alkane solution sums to 100%, the plant species identified in the histological estimation of the composition of the diet were also summed to 100%, assuming that the unidentifiable fragments were in an equal proportion to the identifiable fragments. However, because alkane analyses were restricted to limited sets of plant species on offer, comparison only of species present in both alkane and histological analyses was possible.

3.2.5 Validation Experiment

To assess the validity of carbon isotope analysis, microscopic analysis and long-chain alkane analysis a validation experiment on captive southern hairy-nosed (SHN) wombats was performed. Whole plant samples of five grass species (*Cenchrus ciliaris, Bothriochloa ewartina, Heteropogon contortus, Chrysopogon fallax,* and *Aristida* spp.) that are present at EFNP and potentially consumed by the NHN wombat were collected from Fletcherview Station (James Cook University's experimental research property near Charters Towers, approximately 300 km to the north of Epping Forest National Park) in October 1995. The rainfall and temperature in the Charters Towers district are similar to that of EFNP. The species were collected from paddocks that had not been grazed by cattle for at least two years. *Cenchrus ciliaris*, *B. ewartina* and *H. contortus* were collected with a reciprocator mower and *Chrysopogon fallax* and *Aristida* spp. were hand cut. A total of 102.2 kg of plant material was collected (53.8 kg, 28.15 kg, 10.35, 2.5 kg and 7.4 kg respectively). Plant material was chopped in a hammer mill (10 mm screen), mixed in a paddle mixer and then pelleted. Assuming the pelleted food would have a low protein content as the plant material was collected in the late dry season (October), the pellet mix was supplemented by casein. Molasses, crushed wheat and hot water were added to ensure a pellet could be formed. Wheat was chosen as the binding agent because of its low alkane content (Dove pers. comm.). The final mix consisted of 42.9% *Cenchrus ciliaris*, 22.4% *B. ewartina*, 8.2% *H. contortus*, 2.0% *Chrysopogon fallax*, 5.9% *Aristida* spp., 2.9% molasses, 4.3% casein and 11.4% wheat. Alkane concentrations were estimated for each species using the methods described above.

The pellets were fed to four captive SHN wombats housed at Monash University, Victoria, for a period of nine days. Barboza (1989) demonstrated that the mean retention time for particles of a diet of high fibre concentration was 52 ± 5 hours. Therefore, faecal pellets were collected 6 to 9 days after SHN wombats began eating the pelleted feed. These faecal pellets produced by the SHN wombats were deemed to be representative of the prepared diet.

Faecal pellets were analysed by carbon isotope, microscopic and long-chain alkane methods (described above) by observers who were unaware of the composition of the prepared diet, and in the case of microscopic analysis, the observer believed the faecal pellets were produced by NHN wombats. Further, the observer performing the microscopic analysis was already familiar with the plant species in the prepared diet, from the Epping Forest National Park reference collection.

The validation experiment was repeated twice, and two problems were encountered. The first was in relation to the attempt to estimate faecal recoveries. The primary objective of the validation experiment was to produce faecal material of known species composition for blind microscopic and long-chain alkane analyses. However, the condition of the captive housing was not suited to accurate measurements of food intake and faecal output because of the sand covered floor. Consequently, the estimation of alkane recovery rates for SHN wombats could not be measured.

The second problem was cause by the addition of wheat because the wheat in the pellet mix inadvertently masked the alkane contributions of the grass species (Dove pers. comm.). Consequently, the least-squares optimisation procedure could not determine the correct composition of the prepared diet. Even altering part of the laboratory procedure, by increasing the alkane extraction time from three hours to six hours, could not improve the estimates of botanical composition. Therefore, the validity of the alkane method could not be demonstrated in this study. Instead, published calibration studies of Mayes *et al.* (1986), and Dove and Mayes (1991) serve as confirmation of the long-chain alkane method's validity. The validity of

the microscopic method was successfully examined using the methods described above.

3.3 RESULTS

3.3.1 Carbon Isotope Analysis

Validation Experiment

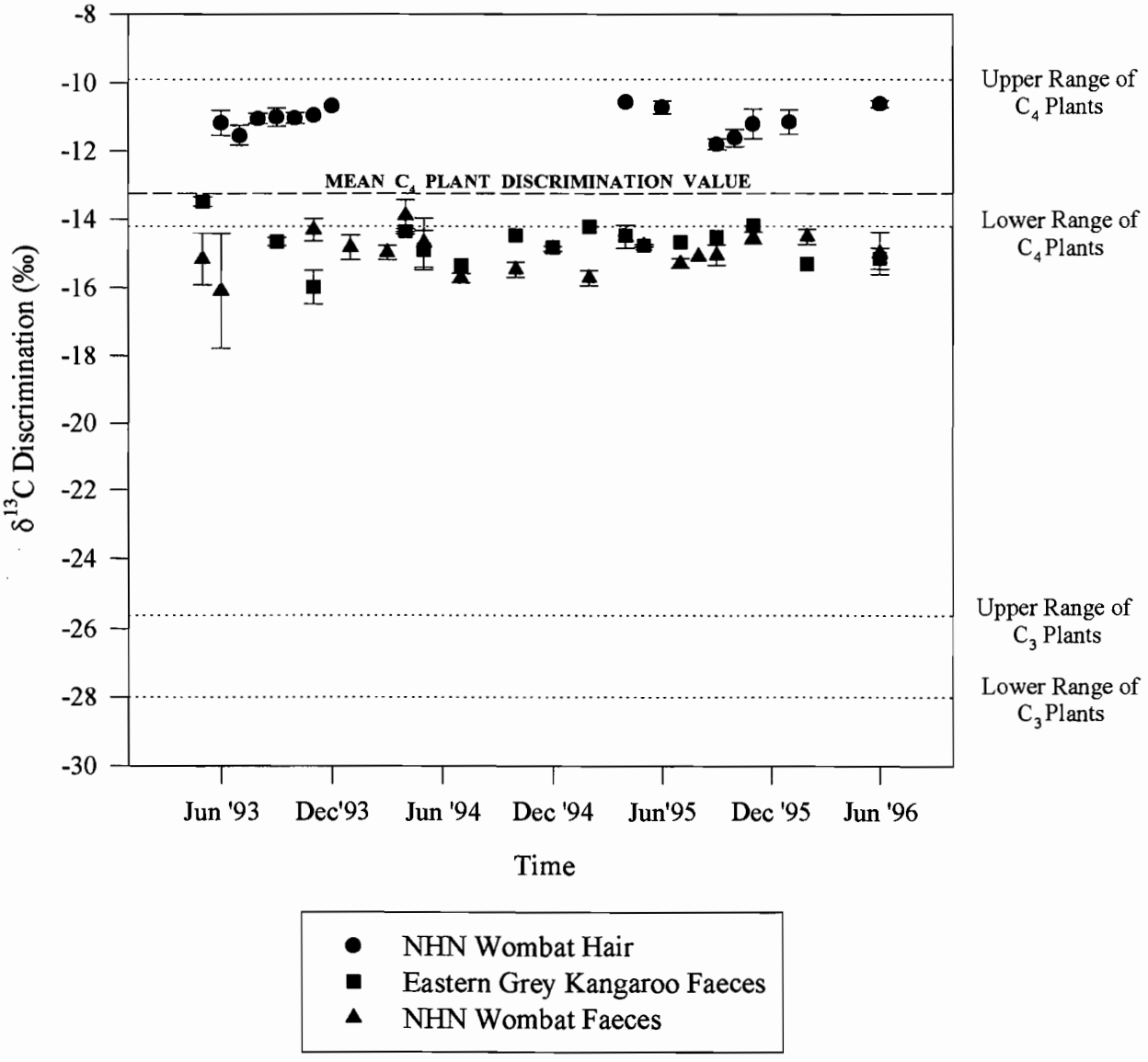
The $\delta^{13}C$ value of the pelleted food (-14.21‰) was influenced by the C_4 grasses (*C. ciliaris*: -12.54‰, *B. ewartina*: -13.8‰, *H. contortus*: -13‰, *C. fallax*: -12.2‰, *Aristida* spp.: -12.8‰, this study and A. Ash, CSIRO Division of Tropical Agriculture, unpublished data) but the contribution of C_3 wheat (-21.8 ± 0.25‰; Tieszen *et al.* 1983) enriched the value slightly. Little variation in $\delta^{13}C$ occurred between the pelleted food (-14.21‰) and faecal output (mean faeces $\delta^{13}C$ value: -14.88 ± 0.26‰, n = 4) suggesting gut flora have limited influence on enriching the $\delta^{13}C$ value of faeces. Therefore, $\delta^{13}C$ value of NHN wombat faeces is likely to be a true representation of forage eaten at the time of sampling.

Field Results

The diets of both NHN wombats and eastern grey kangaroos were dominated by C_4 plants, although deviation of the $\delta^{13}C$ value beyond the range of C_4 plants suggests that occasional C_3 plants were consumed (Figure 3.1). The diet of the NHN wombat exhibits temporal difference in the contribution of C_4 and C_3 plants represented by carbon in hair (ANOVA $F_{5,43} = 4.550$; $p = 0.002$) or faecal residues (ANOVA $F_{5,65} = 3.335$; $p = 0.010$). This is despite the $\delta^{13}C$ value for hair will always be enriched by more than 1‰ because of fractionation (Tieszen *et al.* 1983). Therefore, these results suggest that the bulk of the diet of the NHN wombat is C_4 plants but possibly with the occasional consumption of C_3 plants.

A similar pattern of plant consumption was exhibited by eastern grey kangaroos. No seasonal difference in $\delta^{13}C$ was evident in the diet of eastern grey kangaroos from faecal residues (ANOVA $F_{5,16} = 0.251$; $p = 0.933$) and hair samples were not collected.

Figure 3.1. Carbon isotope analysis measuring the mean contribution (± S.D.) of C_4 and C_3 plants in the diet of NHN wombats and eastern grey kangaroos from faecal samples and body tissue samples (NHN wombat hair).



A single swamp wallaby, *Wallabia bicolor*, faecal sample was analysed by carbon isotope discrimination analysis. It confirmed the browsing habits of the swamp wallaby (Hollis *et al.* 1986), with an exclusive C_3 diet illustrated by a $\delta^{13}C$ value of -25.407‰.

3.3.2 Conventional Microscopic Analysis

Field Results

The results of the microscopic analysis are in agreement with the carbon isotope analysis, with both NHN wombats and eastern grey kangaroos consuming primarily grasses. The preference for grasses is generally non-random for both NHN wombats (Figure 3.2a) and eastern grey kangaroos (Figure 3.2b) with similar preference trends to those found by Crossman (1988). One major difference between the two studies is the value of p_i: the proportion of grasses in the diet ranged between 61% and 85% in Crossman's (1988) study and 84.8% to 99.1% in the current study. The high occurrence of grasses in the foraging environment of the NHN wombat results in low values of p_i for forbs and Malvaceae, and if forbs are consumed even in low quantities the electivity index becomes positive. However, with such high values of p_i, if the consumption of grasses is near or above the value of p_i both the NHN wombat and the eastern grey kangaroo prefer grasses as their principal forage type.

The mean diet of the NHN wombat is dominated by *Aristida* spp., *Enneapogon* spp. and *Cenchrus ciliaris* along with a considerable proportion of unidentifiable fragments (Table 3.2). The accuracy of the analysis was confirmed by the agreement of two independent observers scoring the same twenty two duplicate samples in October 1993 (Table 3.3; ANOVA $F_{1,52} = 0.00$; $p = 1.00$). There was no significant variation in the mean diet with time (ANOVA $F_{9,260} = 0.000$; $p = 1.00$). This trend was repeated when considering the diet of the NHN wombat across its range. Faecal pellets collected from individual wombat burrows, sampling the entire range of the NHN wombat, show no difference in the species composition of the diet (October 1993 samples, ANOVA $F_{20,462} = 0.002$; $p = 1.00$). This suggests that the NHN wombat maintains a consistent diet composition throughout the year with little variation in diet between individuals.

Figure 3.2. Vanderploeg and Scavia's (1979) Electivity Index for three broad classes of forage available to **(a)** NHN wombats and **(b)** eastern grey kangaroos. Data are diets analysed by histological techniques from Crossman (1988) and the current study.

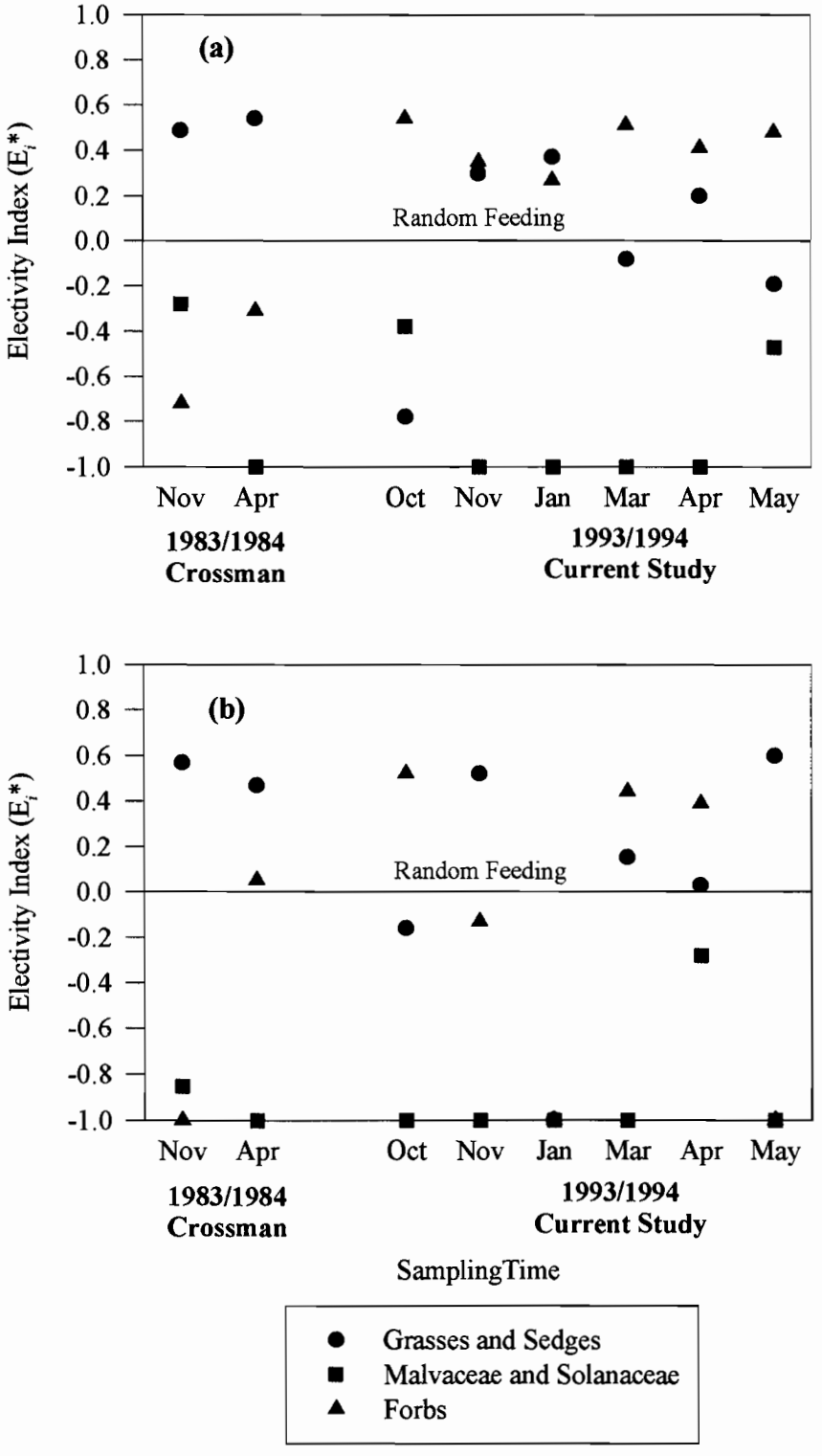


Table 3.2. Temporal trends in diet composition of the NHN wombat as a percentage determined by conventional microscopic analysis. Data for October 1993 compare the findings of two independent observers.

Plant Species	May-93	Jun-93	Jul-93	Sep-93	Oct-93 Observer 1	Oct-93 Observer 2	Nov-93	Jan-94	Mar-94	Apr-94	May-94	Mean (%)	S.D.
Fimbristylis dichotoma	5.2	3.5	0.6	0.5	3.5	1.6	0.1	2.4	0.3	1.7	1.1	1.9	1.6
Aristida spp.	15.0	13.7	22.5	19.1	18.1	10.4	24.7	22.2	25.1	29.9	26.5	20.6	6.0
Bothriochloa ewartina	0	0	0	0	0	0.1	0	0	0	0	0	0	0
Brachiaria spp.	0	0	0	0	0.1	0	0	0	0	0	0	0	0
Cenchrus ciliaris	11.0	8.0	20.2	17.0	6.2	9.3	19.1	22.4	17.1	17.7	11.2	14.5	5.5
Chrysopogon fallax	0	0	0	0	0	0.1	0	0	0.3	0.1	0	0	0.1
Dactlyoctenium radulans	0	0	0	0	0	0	0	0	0	0	0	0	0
Enneapogon spp.	10.6	9.7	15.4	14.0	24.4	9.6	23.7	13.0	25.7	20.2	23.0	17.2	6.3
Enteropogon ramosus	0	0.1	0	0	0.7	0.5	0	0	0.1	0.1	0	0.1	0.2
Eragostris lacunaria	0	0	0	0	0	0.2	0	0	0.2	0	0	0	0.1
Eragostris schultzii	0	0	0	0	0.2	0	0	0	0.2	0.2	0	0	0.1
Heteropogon contortus	0	0	0	0	0	0	0	0	0	0	0	0	0
Perotis rara	0	0	0	0	0.2	0	0	0	0.2	0	0	0	0.1
Themeda triandra	0	0	0	0	0.1	0	0	0	0.1	0	0	0	0
Tragus australianus	0	0	0	0	0.3	0	0	0	0	0	0	0	0.1
Triraphis mollis	0	0	0	0	0.2	0	0	0	0	0	0	0	0
Carissa ovata	0	0.4	0.1	0	0.4	0	0	0	0.1	0	0.2	0.1	0.2
Helichrysum sp.	0	0.1	0	0	0.3	1.2	0	0.2	0	0	0	0.2	0.4
Cleome viscosa	0	0.1	0	0	0.3	0.1	0	0	0.2	0	0	0.1	0.1
Salsola kalii	0	0	0	0	0	0	0	0	1.1	1.8	0.3	0.3	0.6
Evolulus alsinoides	0	1.6	0	0	4.3	0.2	0	0	1.4	3.4	9.4	1.9	2.9
Abutilon sp.	0.1	0	0	0	0.1	1.2	0	0	0.1	0	0	0.1	0.4
Sida sp.	1.1	0.1	0.1	0.2	0	0	0	0	0	0	0	0.1	0.3
Waltheria indica	1.7	3.4	4.3	6.4	2.5	0.1	0.7	0.6	1.2	0.1	0.7	2.0	2.0
Tribulus terrestris	0	0	0	0	0.4	0.2	0	0	0.1	0	0	0.1	0.1
Unidentifiable Monocots	49.5	58.8	34.6	38.4	33.6	60.2	31.0	34.3	25.1	24.5	26.2	37.8	12.8
Unidentifiable Dicots	5.7	0.4	2.1	4.4	4.5	5.1	0.7	1.8	1.4	0.2	1.4	2.5	2.0

Table 3.3. Percentage contribution of plant parts of species consumed by the NHN wombat. The relative proportion of identifiable fragments (one decimal point) represents the number of identified fragments for each species as a proportion of the total fragments identified (N = 13328).

	Leaf	Stem	Sheath	Unknown	Relative Proportion of Identifiable Fragments
Fimbristylis dichotoma	100	0	0	0	2.8
Aristida spp.	56.6	20.1	23	0.13	34.8
Bothriochloa ewartina	0	0	0	100	0.0
Brachiaria spp.	0	0	100	0	0.0
Cenchrus ciliaris	17.7	15.5	66.4	0.2	25.8
Chrysopogon fallax	0	14.2	85.7	0	0.1
Cymbopogon refractus	0	0	0	0	0
Dactyloctenium radulans	0	0	100	0	0.0
Enneapogon spp.	29.4	44.4	25.7	0.32	29.1
Enteropogon ramosus	9.5	0	90.4	0	0.2
Eragostris lacunaria	20	60	20	0	0.1
E. schultzii	12.5	62.5	25	0	0.1
Perotis rara	50	50	0	0	0.0
Themeda triandra	66.6	0	33.3	0	0.0
Tragus australianus	0	0	100	0	0.1
Triraphis mollis	66.6	0	33.3	0	0.0
Carissa ovata	100	0	0	0	0.2
Helichyrsum sp.	95.8	0	4.1	0	0.2
Cleome viscosa	100	0	0	0	0.1
Salsola kali	100	0	0	0	0.3
Evolvulus alsinoides	100	0	0	0	2.1
Abutilon sp.	100	0	0	0	0.2
Sida sp.	100	0	0	0	0.3
Waltheria indica	100	0	0	0	3.4
Tribulus terrestris	100	0	0	0	0.1
Mean Consumption	42.6	24.1	33.1	0.2	

Table 3.4. A comparison between two independent observers and their scores for plant parts expressed as percentages. The three plant species (*Aristida* spp., *Enneapogon* spp. and *Cenchrus ciliaris*) are the three species occurring most often in the NHN wombat diet from the October 1993 sampling period.

	Plant Part							
	Leaf		Stem		Sheath		Unknown	
Observer ⇒	1	2	1	2	1	2	1	2
Aristida spp.	64.4	60.1	13.9	11.7	18.0	28.0	3.4	0.0
Enneapogon spp.	34.9	26.6	12.2	44.4	50.3	28.8	2.5	0.0
Cenchrus ciliaris	21.2	76.9	6.1	22.5	67.2	0.4	5.3	0.0

The NHN wombat appears to select leaf over sheath and stem as the preferred part of the plant (Table 3.3). This is only a representative trend with the NHN wombat consuming different parts of the plant for different species. However, caution should be used in the interpretation of the results. For many plant species differential digestibility of the different parts of the plant may influence the proportion of fragments observed in the faeces. Similarly, interpretation differences between observers may lead to a bias of one part of the plant over another depending on the quality and quantity of the reference photomicrographs. This is evident in a comparison between the two independent observers and their scores for the three major plant species consumed by NHN wombats (*Aristida* spp., *Enneapogon* spp. and *Cenchrus ciliaris*) in the October 1993 sampling period (Table 3.4). While there was good agreement for *Aristida* spp. and *Enneapogon* spp., there was a clear discrepancy in *C. ciliaris* and the allocation of fragments into the four plant part categories.

Validation Experiment

Of the five plant species in the prepared diet only two were identified by the observer (Table 3.5). This highlights potential sources of error in the histological technique. The relative contribution of dicots also emphasises potential errors in

the histological technique. Assuming the relative proportion of unidentifiable fragments varies consistently with plant species, an estimate of biomass can be made. The original total biomass was 102.2 kg. Therefore, an estimate of biomass of dicots in the prepared forage, based on the microscopic analysis, would be 4.1 kg. Even if some dicots were inadvertently collected and then processed through the hammer mill (effectively a screening process) a 4.23% contribution to the diet was very unlikely, especially as *Carissa ovata* and *Salsola kalii* were not observed at the collection site.

Table 3.5. Comparison between actual composition of a prepared diet and the diet measured by the conventional microscopic technique.

Diet Composition		Prepared Diet* (%)	Microscopic Analysis (%)
Additives	Molasses	2.9	-
	Casein	4.3	-
	Wheat	11.4	-
Plant Species	*Aristida* spp.	5.9	24.0
	Bothriochloa ewartina	22.4	0.0
	Cenchrus ciliaris	42.9	12.8
	Chrysopogon fallax	2.0	0.0
	Heteropogon contortus	8.2	0.0
	Fimbristylis dichotoma	-	0.1
	Enneapogon spp.	-	26.5
	*Carissa ovata**	-	0.2
	*Cleome viscosa**	-	0.1
	*Salsola kalii**	-	0.1
	*Evolvulus alsinoides**	-	0.2
	*Waltheria indica**	-	0.1
Unidentifiables	Monocots	-	32.3
	Dicots*	-	3.6

* The prepared diet contained some dicots. This was a direct consequence of the broad scale harvest technique using to collect plants, but their contribution was small. *Carissa ovata* and *Salsola kalii* were not observed at the collection site.

The distinction between epidermal patterns of monocots and dicots is clear, because monocots exhibit a regular pattern of epidermal cells compared to a random arrangement of cells in dicots. An observer should easily identify a dicot in a monocot-dominated diet because of the distinctive difference in epidermal patterns. This in turn leads to overestimation of dicots.

Of more concern is the lack of detection of *Bothriochloa ewartina*, *Chrysopogon fallax* and *Heteropogon contortus*, the underestimation of *Cenchrus ciliaris* and overestimation of *Enneapogon* spp. and *Aristida* species. Again this is likely to be in part due to observer error but also due to a shortcoming of the technique. While the reference collection of photomicrographs was as comprehensive as possible, the many combination of distortions that epidermal cells can experience through digestion could only be replicated artificially but at considerable expense. This leads to the observer 'interpreting' the epidermal fragments, and while agreement between two observers can occur (Table 3.2), a fundamental problem of 'interpretation' exists.

3.3.3 Long-chain Alkanes

Evaluation Experiment

The concentration of alkanes found in the plant samples was high in comparison to temperate plants, perhaps because of the role of alkanes in the water economy (H. Dove pers. comm.). Canonical variates analysis determined that the pattern of alkane concentrations of each plant species were well distinguished except for *Eragostris lacunaria* and the two *Enneapogon* spp. which were similar in terms of the first three canonical variates. The predicted botanical composition by alkane analysis was not different to that examined by microscopic analysis (ANOVA $F_{1,10} = 0.837$; $p = 0.382$) (Table 3.6). There was no difference between individual NHN wombats in their selected diets (ANOVA $F_{23,144} = 0.000$; $p = 1.00$). The success of the evaluation experiment prompted large-scale application of the long-chain alkane method.

There were negligible differences in a comparison between the simultaneous equation and least-squares optimisation procedure to predict the diet composition of the NHN wombat and the eastern grey kangaroo (Table 3.7).

Table 3.6. Comparison of the NHN wombat diet measured by long-chain alkane analysis and microscopic analysis for faecal samples collected in October 1993. Predicted diets represent a mean botanical composition as a percentage (± one standard deviation) of 25 faecal samples collected from burrows throughout the known range of the NHN wombat.

Plant Species	Alkane	S.D.	Microscopic*	S.D.
Fimbristylis dichotoma	3.33	4.17	2.48	3.12
Aristida sp.	14.87	9.6	15.81	3.63
Cenchrus ciliaris	31.29	10.37	7.71	4.50
Enneapogon sp.	9.37	4.58	18.90	3.81
Eragostris sp.	6.45	7.31	0.24	0.44
Malvaceae	9.2	7.11	0.62	0.92
Salsola kalii	25.2	22.97	-	-
Waltheria indica	-	-	1.48	1.33
Monocots	-	-	44.86	6.08
Dicots	-	-	6.23	4.91

* Plant species recorded by microscopic analysis but in quantities of less than 1% include *Bothriochloa ewartina, Dactyloctenium radulans, Enteropogon ramosus, Perotis rara, Themeda triandra, Triraphis mollis, Carissa ovata, Helichrysum sp., Cleome viscosa, Evolvulus alsinoides, Tribulus terrestris* (Table 3.3).

Therefore, the results of the preliminary experimentations could be compared with the field results.

Field Results

The estimation of the composition of the diet of the NHN wombat by long-chain alkane analysis differed depending upon what estimation procedure was used in the least-squares optimisation procedure (Table 3.8). None of the estimation procedures statistically differed from the mean microscopic predicted diet (ANOVA $F_{3,92} = 0.353$; $p = 0.787$) despite clear differences between the estimations of diet (e.g. *Fimbristylis dichotoma*). This was likely to be an artefact

Table 3.7. Comparison of the best solutions for long-chain alkane analysis using the simultaneous equation (SIM) method of Dove (1992) and the least-squares optimisation (LSO) of Dove and Moore (1995). Analysis was performed on two data sets. In data set A, canonical variates analysis could not distinguish between the alkane concentrations of the two species of *Aristida*, and likewise the two species of *Enneapogon* in the reference samples and are therefore represented as genera. In data set B, the two species of *Aristida* are considered independent, likewise so are the two species of *Ennepogon*. Faecal samples were collected for NHN wombats at burrows labelled B100, B78 and B69; and for the eastern grey kangaroo on the second BOTANAL transect, approximately 150 metres from the centre of the transect. Diet solutions are expressed as a percentage.

Data Set	Plant Species	Herbivore Identification							
		Wombat B100		Wombat B78		Wombat B69		Kangaroo T2-150E	
		SIM	LSO	SIM	LSO	SIM	LSO	SIM	LSO
A	*Aristida* spp.	27.7	25.1	3.9	7.4	18	11.4	15.7	14.2
	Cenchrus ciliaris	47.8	46.4	42.3	40.3	39	38.9	37.8	37.9
	Enneapogon spp.	12.5	12.2	5.4	5.4	9.3	11.5	9.9	10.3
	Eragostris lacunaria	0	0	22.4	21.1	0	0	0	0
	Fimbristylis dichotoma	0	0	0	0	8.3	1.5	7	3.6
	Abutilon sp.	4.5	5.5	0	0	0	2.2	0	0
	Sida sp.	4.5	4.6	26	23.9	3.3	10.1	2.9	4.7
	Salsola kalii	3.1	6.2	0	1.8	22	24.4	26.7	29.3
B	*Aristida* sp. A	0	0	0	0	0	0	0	0
	Aristida sp. B	22.1	21.2	6.7	8.7	20.1	12.4	18.9	18.9
	Cenchrus ciliaris	47.6	47.2	41.2	40.2	39.6	39.1	38.2	38.3
	Enneapogon sp. A	0	0.6	5.5	5.6	1	0.5	9.3	9.2
	Enneapogon sp. B	11.9	11.2	0	0	7.5	10.5	0	0
	Eragostris lacunaria	2.4	2.4	22.4	21.9	0	0	0	0
	Fimbristylis dichotoma	1.4	1.6	0	0	11.3	4.5	7.4	7.4
	Abutilon sp.	2.4	2.3	0	0	0	0	0	0
	Sida sp.	5.1	5.6	24.2	23.7	1	10	0	0
	Salsola kalii	7.1	8	0	0	19.5	23	26.2	26.1

Table 3.8. A estimation of the composition of the diet of the NHN wombat by histological analysis and three variations of the least-squares optimisation procedure for alkane analysis (see text for descriptions of the three estimation procedures). Dashes represent plant species not included in the analysis. Histological analysis incorporates twelve months of sampling with 10 collection periods. Alkane analysis incorporates thirty-three months with 15 collection periods.

Plant Species	Microscopic Analysis	ALKANE		
		Estimation Procedure 1	Estimation Procedure 2	Estimation Procedure 3
Fimbristylis dichotoma	1.85	14.39	17.75	29.07
Aristida spp.	20.64	14.85	26.63	16.20
Bothriochloa ewartina	0	0.27	0.00	-
Brachiaria spp.	0.01	8.08	10.12	-
Cenchrus ciliaris	14.46	1.76	7.05	7.87
Chrysopogon fallax	0.04	7.75	6.01	13.17
Dactyloctenium radulans	0	3.73	-	-
Enneapogon spp.	17.21	19.02	21.72	26.02
Enteropogon ramosus	0.12	-	-	-
Eragostris lacunaria	0.03	8.34	3.74	7.67
Eragostris schultzii	0.04	-	-	-
Heteropogon contortus	0	1.33	0.69	-
Perotis rara	0.02	2.03	-	-
Themeda triandra	0.01	-	-	-
Tragus australianus	0.02	-	-	-
Triraphis mollis	0.01	0.25	-	-
Carissa ovata	0.1	0.00	0.02	-
Helichrysum sp.	0.15	2.25	2.26	-
Cleome viscosa	0.06	-	-	-
Salsola kalii	0.29	2.20	-	-
Evolvulus alsinoides	1.85	-	-	-
Malvaceae		1.63	1.48	-
Abutilon sp.	0.12	5.31	0.13	-
Sida sp.	0.14	1.03	0.00	-
Waltheria indica	1.96	5.45	2.40	-
Tribulus terrestris	0.05	0.35	-	-
Unidentifiable Monocots	37.83			
Unidentifiable Dicots	2.51			

of the statistical analysis. *Aristida* spp. and *Enneapogon* spp. were again the principal species, with significant contributions of *Fimbristylis dichotoma*, *Chrysopogon fallax*, *Brachiaria* spp., *Eragostris lacunaria* and *Cenchrus ciliaris* depending on what species and/or alkanes were used in the least-squares optimisation procedure. Temporal trends using the alkane methodology are difficult to describe, primarily because of difficulties in harvesting all plant species on offer, especially as perceived by herbivores, and temporal variation and significance of alkane concentrations. Despite these difficulties, temporal trends in the diet of the NHN wombat and the eastern grey kangaroo, using the three variations of the least-squares optimisation procedure were observed and are shown in Appendix 3.

3.4 DISCUSSION

3.4.1 Diet of the NHN wombat

3.4.1.1 Botanical Composition

The NHN wombat is a grazer, consuming more than 90% monocotyledons. The principal components of its diet include native grasses, such as *Aristida* spp. and *Enneapogon* spp., the sedge *Fimbristylis dichotoma* and the introduced grass species *Cenchrus ciliaris*. The short-term composition of the diet (during the current study) showed little temporal variation suggesting that changes in forage quantity (demonstrated in Chapter 2) are not accompanied by changes in diet selection. Similarly, despite spatial heterogeneity of the habitat, the diet composition did not vary between individuals.

The estimated diets determined in this study were slightly different to those of Flößer (1986) and Crossman (1988). Table 3.9 ranks plant species considered to be palatable, identified in the previous two studies on the diet of NHN wombat and the current study, and those species known to be palatable to cattle grazing in a similar environment. Although the palatability ranks do not meet Petrides (1975) classification for principal foods or preferred foods, it does provide an indication of the NHN wombat's diet. Species such as *Aristida* spp. are consumed by NHN wombats but not by cattle, whereas *Enneapogon* spp. is consumed with similar preference to both NHN wombats and cattle. This suggests that in contrast to

cattle, grass species with little leaf and a coarse stem like *Aristida* spp. are consumed by the NHN wombat.

Table 3.9. Species palatability ranks. Studies 1 (Flößer 1986) and 2 (Crossman 1988) are previous studies on the diet of the NHN wombat. Study 3 (A. Ash CSIRO Tropical Crops and Pastures, pers. comm.) is an estimation of the palatability of forage for cattle foraging in a similar environment. Study 4 is the current study highlighting palatability of the diet determined by microscopic analysis (micro) and the three alkane estimation procedures.

Plant Species	Study						
	NHN Study 1 Flößer	NHN Study 2 Crossman	Cattle Study 3 Ash	NHN Study 4 Micro	NHN Study 4 Alkane 1	NHN Study 4 Alkane 2	NHN Study 4 Alkane 3
Fimbristylis dichotoma	2	8	6	5	3	3	1
Aristida spp.	4	2	10	1	2	1	3
Brachiaria spp.	9	n.r.	4	20	5	4	n.r.
Cenchrus ciliaris	10	7	1	3	13	5	5
Chrysopogon fallax	3	n.r.	3	15	6	6	4
Enneapogon spp.	6	3	5	2	1	2	2
Eragostris spp.	1	n.r.	n.r.	13	4	7	6
Heteropogon contortus	5	1	2	22	15	11	n.r.
Salsola kalii	n.r.	n.r.	11	7	11	n.r.	n.r.
Abutilon spp.	n.r.	n.r.	8	10	8	12	n.r.
Sida spp.	n.r.	n.r.	7	9	16	10	n.r.
Waltheria indica	n.r.	n.r.	9	4	7	8	n.r.
Number of Plants Ranked	14*	12	11	25	19	13	6

n.r. Plant species not included in the ranking.
* Only monocots ranked.

Of interest are the temporal trends in the contribution of *Heteropogon contortus* and *Cenchrus ciliaris* to the NHN wombat's diet. *Heteropogon contortus* was the fifth-ranked species consumed by the NHN wombat during Flößer's (1986) study, the most preferred species consumed by the NHN wombat during Crossman's (1988) study and a preferred forage species of cattle (Table

3.9). In the current study, the NHN wombat consumed very little *H. contortus* (less than 1.5%). Rather than reflecting a long-term change in forage preference of the NHN wombat, the differences in the species composition of diet between the current study and Flößer (1986) and Crossman (1986), represent long-term changes in the species composition of the pasture community. Specifically, the frequency of *H. contortus* has declined from 16.7% in 1987 to 1.7% in 1996. Likewise, the frequency of occurrence of *C. ciliaris* had increased substantially from 1987, and in 1996 represented up to 60% of the forage available to the NHN wombat.

In contrast to the palatability ranks for cattle, *Aristida* spp. and *Enneapogon* spp. rank higher than *C. ciliaris* for the NHN wombat (Table 3.9). However, the true contribution of *C. ciliaris* may be greater than these values due to potential errors in the analysis (see discussion below). In October 1993, the mean contribution of *C. ciliaris* to the total composition of the diet of the NHN wombat was estimated to be 31.3% using the alkane method (Table 3.7) compared to the annual mean value of 14.5% for histological analysis (Table 3.8). Regardless of the specific contribution of *C. ciliaris* to the NHN wombat diet, it has become a significant component of the diet of the NHN wombat. Rather than being considered an ecological 'problem', *C. ciliaris* should be regarded as an important forage species for the NHN wombat.

The differences in the diet composition of the NHN wombat in this study compared to the studies of Flößer (1986) and Crossman (1988) supports Flößer's (1986) conclusion that the NHN wombat is a non-specialist forager, which can alter the composition of its diet in response to changes in species composition in the pasture community.

3.4.1.2 Diet Selection

The diet of the NHN wombat exhibits little seasonal variation in composition or in the selection of plant parts. This is in marked contrast to other groups of grazing herbivores, such as ungulates. Often, ungulates exhibit distinct seasonal variability in the composition of their diets (e.g. Dailey *et al.* 1984, Van Rooyen 1992). This variability often extends to seasonal differences in the

contribution of monocotyledons and dicotyledons in the diet (e.g. Van Rooyen 1992) and/or the selection of plant parts (e.g. Gwynne and Bell 1968). There might be two explanations for the lack of seasonal variability in the diet of the NHN wombat. First, diet selection may not be as important to the wombat as it is to ungulates. Instead the conservation of energy is the critical factor of the wombat's biology. The NHN wombat therefore maintains a conservative foraging regime where the time spent above ground is more important than the plants selected. This coincides with the ability of the wombat to maximise the retention of ingesta and its low energy and nitrogen requirements for maintenance. Second, temporal differences in diet selection were not detected by the methods used. The methodology is discussed below.

Both the NHN wombat and the eastern grey kangaroo clearly select grasses as the preferred forage (Figures 3.1 & 3.2). The dietary overlap of these two sympatric herbivores is discussed in Chapter 6. This was maintained both in the short-term (during the current study) and long-term (comparing the current study to the studies of Flößer (1986) and Crossman (1988)), even though the species composition of the pasture community had changed (Figure 2.8).

The benefit of the electivity index is that it should allow a quantitative comparison between the current study and the earlier study of Crossman (1988). However, the limitations outlined by Lechowicz (1982) and Norbury and Sanson (1992) need to be considered. The mean value of the electivity index for grasses consumed by NHN wombats during this study was -0.03, suggesting that NHN wombats consume grasses in proportion to their availability (i.e. the NHN wombat feeds randomly). During the Crossman (1988) study, the mean value of +0.52 indicated a preference for grasses was non-random. However, the mean proportion of monocots consumed by NHN wombats was 98% in Crossman's (1988) and 92.8% in the current study. The different values of the electivity index between the two studies reflect fundamental differences in a key variable used in the calculation of the electivity index: grasses as a proportion of total of forage. Temporal differences in the species composition of the forage between 1982/83 and 1993/94

significantly alter the value of the electivity index even though the general composition of the diet has not changed in the same period.

The Vanderploeg and Scavia (1979) electivity index has been used successfully to describe dietary selection of the bridled nail-tail wallaby (*Onychogalea fraenata*), the black-striped wallaby (*Macropus dorsalis*), the allied rock-wallaby (*Petrogale assimilis*) and domestic cattle (Dawson *et al.* 1992, Evans 1992, Horsup and Marsh 1992). Dawson *et al.* (1992) used the electivity index to describe the preferred diet of *O. fraenata*, and used values of the electivity index to conclude that the habitat was sub-optimal for the wallaby. While each of these studies considered broad groupings such as grasses, forbs, browse and Malvaceae, Evans (1992) extended his analysis to calculate an electivity index for individual species consumed by the black-striped and bridled nail-tail wallabies. The benefits of assigning a scale of preference for each species consumed are clear, particularly from a management perspective. However there are inherent problems in the use and interpretation of selectivity indices (Lechowicz 1982), such as the sensitivity of the index to variations in the relative abundance of food types. As sensitivity increases, caution should be exercised in interpreting selectivity indices. For example, an important forage species such as *Cenchrus ciliaris* has a negative electivity index suggesting that it is a 'sub-optimal' forage species although it contributes more than 14% of the NHN wombat's diet. However, with its high standing biomass, *C. ciliaris* may enable the NHN wombat to maintain rates of intake if the availability of other preferred species is limited. In addition, rare species such as some of the forbs may be misinterpreted as being 'optimal' or preferred forage species, even though they contribute negligible quantities to the diet. Hence the value of individual electivity indices for each forage species, or indeed the broad categories in this study, is contentious.

3.4.1.3 Plant Parts

The NHN wombat consumed leaf (42.4%) in preference to stems (24.0%) or sheath (32.9%) (Table 3.2). This trend is similar to other herbivores (e.g. the black-striped wallaby, *Macropus dorsalis*; Evans 1992) in habitats similar to EFNP. However, these estimates for the consumption of plant parts must be used

only as a guide. Differential digestibility can markedly influence the contribution of one or more of the components. For example, the *in vitro* dry matter digestibility of *Cenchrus ciliaris* ranges from 42.7% to 51.9 % for leaf; 38.0% to 46.0% for sheath; and 28.2 to 36.2% for stem (Appendix 1). The clear difference in the digestibility of each plant part will affect the observed proportion in the diet. To confound the estimation of plant parts using histological analysis, individual interpretation can influence results (Table 3.3)

Jarman (1994) postulated that some macropod species, particularly those of small body size (> 10 kg), may consume seedheads in response to the low digestibility and low concentrations of extractable nutrients in monocotyledons. The consumption of seedheads may provide pulses of high-quality food that may otherwise not be available (Jarman 1994). Conventional microscopic analysis did find the occasional seedhead in the diet of the NHN wombat, but they were in such low abundances that consumption was considered low and incidental rather than deliberate. However, the issue of differential digestibility may affect the passage of seedheads through the gastrointestinal tract. Dove *et al.* (1996) investigated the alkane patterns of six pasture species and found that the inflorescences were characterised by the presence of alkenes (aliphatic hydrocarbons with one or more double bonds), in significantly higher quantities than other parts of the plant (leaf, sheath, base and stem). The presence of C_{31} alkene and C_{33} alkene in the faeces of some NHN wombats may suggest that the consumption of seedheads occurs irregularly, and not by every individual.

3.4.1.4 Grazing - Grasses as a Food Resource

Grasses present a food resource that is generally high in fibre, low in crude protein and low in digestibility (Chapter 2, Robbins 1983, Owen-Smith 1992). The quality of the forage available to the NHN wombat correlated with rainfall and associated plant growth (Chapter 2). The NHN wombat maintains a relatively consistent composition of plant species in its diet throughout the year with no clear seasonal trends. This trend is consistent among individual NHN wombats with little variation in diet composition between individuals.

The clear preference for grasses is consistent with the diets of both the southern hairy-nosed (SHN) wombat (Wells 1973, Dierenfeld 1984, Kean 1990) and the common wombat (McIlroy 1973, Mallet and Cooke 1986, Triggs 1988, Rishworth *et al.* 1995a&b). The common wombat is more flexible in its choice of forage, consuming sedges, rushes, bark, roots, various dicotyledon species and pine needles (in pine plantations) when grasses are limiting (McIlroy 1973, Mallet and Cooke 1986, Rishworth *et al.* 1995a). The foraging activities of the SHN wombat also depend on forage availability. Living in a semi-arid habitat with a high drought frequency, coupled with grazing pressure from European rabbits, sheep and native herbivores (*Macropus* spp.), the SHN wombat often experiences low availability of grasses. When the availability of preferred species of grass becomes limited, the SHN wombat switches in its diet choice to less preferable plants such as forbs, other dicots and chenopod shrubs (Dierenfeld 1984).

The current study period encompassed four years of below-average rainfall and regional drought in the Epping Forest district. However, forage quantity during this period was sufficiently high that grasses were available throughout the year (Chapter 2). It is possible that if the biomass of grasses at Epping Forest becomes significantly reduced, the NHN wombat would exhibit similar levels of flexibility in diet selection as the other two wombat species. However, tropical grasses usually have rapid periods of growth following summer rainfall. In contrast, the growth of temperate grasses generally follows winter and spring rainfall and generally achieve less standing biomass than tropical grasses. Therefore, it is likely that forage quality, rather than quantity is the main determinant of diet selection for the NHN wombat under non-drought conditions.

This chapter has demonstrated that, unlike other herbivores (e.g. the bridled nail-tail wallaby, Evans 1992), the NHN wombat does not change the composition of its diet from grasses to forbs or browse in response to changing forage quality or quantity. Further, since the NHN wombat is dependent on its burrow (Wells 1973, 1978 a&b, Triggs 1988, Johnson 1991a) it also cannot easily move to a dry season forage refuge where the quantity and quality of the forage may be greater (*cf.* the eastern grey kangaroo, Hill 1982). The diet of the NHN

wombat does not change in the short term suggesting that changes in forage quality and quantity do not evoke detectable changes of forage choice. However, changes in forage quality and quantity may effect other aspects of the NHN wombat's physiology and behaviour. These aspects are investigated in Chapters 4 and 5 respectively.

3.4.2 Evaluation of techniques

The quantification of food types consumed by herbivores has received considerable attention and the merits of common methodologies have been reviewed by several authorities (e.g. Holechek *et al.* 1982, Barker 1986a, Norbury and Sanson 1992, Dove and Mayes 1996). The following section adds to the discussion on herbivore diet analysis, focussing specifically on the techniques used to quantify the diet of the NHN wombat.

3.4.2.1 Carbon Isotope Analysis

The use of stable isotopes, both carbon and nitrogen, is becoming a useful and common means of determining diet and habitat selection in ecological studies. Both Tieszen *et al.* (1979) and Ambrose and DeNiro (1986) used stable carbon isotopes successfully to discriminate between grazing mammals and browsing mammals in African savanna grasslands. Horsup and Marsh (1992) used stable carbon isotopes to partition the diet of the allied rock-wallaby, *Petrogale assimilis*, into grasses (C_4 plants) and forbs and browse (C_3 plants). McIllwee (1994) used stable isotopes of both carbon and nitrogen to partition the diets of three herbivorous/mycophagus marsupials (the northern brush-tailed bettong, *Bettongia tropica*, the rufous bettong, *Aepyprymnus rufescens* and the northern brown bandicoot, *Isoodon macrourus*) and determine dietary overlap in a sympatric range.

These studies have demonstrated the success of stable isotopes to partition the diet of herbivores into C_3 and C_4 components. However, if faecal samples are used in the analysis, the issue of differential digestibility must be considered. Further, $\delta^{13}C$ analysed in the faeces represent excreted carbon rather than assimilated carbon. Stable carbon isotope analyses of body tissue provide a

representation of assimilated carbon as long as the findings of Tieszen *et al.* (1983) regarding fractionation and, again, differential digestibility are considered.

The current study clearly demonstrates that the diet of the NHN wombat consists almost entirely of tropical grasses or C_4 plants, with potentially a limited but seasonal contribution of C_3 plants. There were clear differences in the $\delta^{13}C$ values of both body tissue samples (hair) and faecal residues. However, the differences can be explained if $\delta^{13}C$ enrichment is considered for hair samples (Tieszen *et al.* 1983). Similar discrepancies between the $\delta^{13}C$ value of hair and pasture were reported by Minson *et al.* (1975) for cattle grazing on *Heteropogon contortus* pasture. They found that the $\delta^{13}C$ value for cattle hair was -12.1‰ when cattle grazed on forage with a $\delta^{13}C$ value of -14.0‰ in comparison to the current study that recorded a $\delta^{13}C$ value for wombat hair of -11.07‰ grazing on forage with a $\delta^{13}C$ value of -13.24‰. Although the ratios for cattle and the NHN wombat compare favourably, quantification of the actual level of $\delta^{13}C$ enrichment for wombats (and marsupials in general) would be useful.

3.4.2.2 Conventional Microscopic Analysis

Conventional microscopic analysis is the most common method to quantify and provide an inventory of plant species consumed by herbivores (Stewart 1967, Holechek *et al.* 1982, Norbury 1988b). The histological method of analysis describes the diet based on proportions of identifiable epidermal fragments in the faeces. The merits of specific techniques to quantify the fragments have been reviewed in detail by many authorities (e.g. Stewart 1967, Hansson 1970, Dearden *et al.* 1975, Fitzgerald and Waddington 1979, Holechek *et al.* 1982, Barker 1986a& b, Norbury 1988b, Norbury and Sanson 1992). Regardless of the subtle variations in the methodologies used, two fundamental problems exist and will always influence the described diet. They are the effect of digestion on the forage, and the effect of human error.

The influence of digestion

The process of herbivore digestion affects plant species and parts of the plant differentially, depending on their chemical composition. Stewart (1967)

highlighted the fact that the epidermis of some plant species is completely digested or epidermal fragments can be so small that they are unidentifiable. This problem is encountered in every study describing the diet of a herbivore using histological techniques.

Besides differential digestibility, the other main problem affecting histological techniques is the proportion of unidentifiable epidermal fragments. To reduce the influence of unidentifiable fragments some studies categorise the diet into groups such as grasses, forbs and other (e.g. Vernes 1995). This approach is cost effective, but a more accurate account of diet composition could be gained with the use of stable isotope analysis if there are trophic differences between the plant groups and categorisation of the diet is sufficient (e.g. McIllwee 1994). Alternatively, Hansson (1970) attempted to develop correction factors to account for the unidentifiable proportion. This approach has been used in several other studies (e.g. Dearden *et al.* 1975, Norbury 1988b, Orell 1989, Kean 1990) but with limited application to field studies.

In an experiment to examine the effect of digestion of different plant species, Norbury (1988a) fed experimental diets to captive eastern grey kangaroos. He found that the botanical composition of both stomach contents and faeces did not reflect the same dry weight proportions as the original diet. However when a factor correcting for unidentifiable fragments, as described by Norbury (1988b), was applied the proportions of the diet of both the stomach contents and faeces showed agreement with the ingested composition. Norbury (1988a) suggests that rather than differential digestibility causing problems of misinterpretation, the reliance on epidermal fragments for identification and the large proportion of unidentifiable fragments are the principal causes of error when estimating the composition of the diet using histological techniques. Norbury (1988b) encourages the use of a correction factor. However, to calculate the correction factor a knowledge of the proportion of identifiable tissue for each species is required, including the various growth stages encountered by the herbivore. In controlled experimental conditions calculating the conversion equation may be practical but

its application in field studies where the forage environment is spatially and temporally diverse may be limited.

Human Error

The influence of human error has received little attention in critical reviews of the merits of diet analysis by histological techniques (e.g. Holechek *et al.* 1982, Norbury and Sanson 1992). Norbury and Sanson (1992) suggest that carelessness in sample preparation and observer inexperience are the primary human factors causing potential error. However, personal interpretation of epidermal fragments in the faeces needs to be considered as a major source of error in histological analyses.

When a prepared diet of known composition was fed to captive SHN wombats only two of the five species were correctly detected by an experienced observer (but one who was unaware of the source of the samples), and in very different proportions to the actual composition (Table 3.5). This error is not an observer error, rather an interpretation error. Table 3.2 highlights agreement between two independent and experienced observers for the October 1993 sampling period using the same prepared slides and reference collection of photomicrographs. The error in the validation experiment (Table 3.4) reflects how epidermal fragments are interpreted. The reference collection of epidermal fragments was as comprehensive as possible, sampling various parts of the plant from many different species, including all species in the validation experiment. However, ingestion and digestion of the plant epidermis by the herbivore may result in the epidermal fragments appearing completely different to those in the reference collection. This may result in the allocation of the digested epidermal fragment to a different plant species or plant part than was actually consumed by the herbivore. This potential source of error needs to be considered in all studies describing the diet of herbivores using histological techniques.

The validation experiment also highlighted biases in overestimating rare components when their epidermal pattern is distinctive from the majority of epidermal fragments. In the validation experiment, the prepared composition was assumed to include only monocots. A small fraction of dicots may have been

accidentally incorporated into the mix as a consequence of sampling such a large mass of natural vegetation. Although the composition of dicots was only a fraction of a percent, the distinct epidermal pattern of dicots allowed easy identification leading to overestimation of the contribution of the dicots.

The potential error associated with observer interpretation should to be considered when describing the diet of the NHN wombat, and indeed other studies using histological analysis. If these potential errors are not considered, particularly for a threatened species, the resulting management decisions may be erroneous. In the current study, results have implications for the management and conservation of the NHN wombat's foraging environment. A species such as *Cenchrus ciliaris* is considered a 'problem' species for management because it is increasing in biomass and frequency of occurrence at a disproportionate rate compared to native forage species. The mean annual contribution of *C. ciliaris* to the NHN diet was 14.5% of the total identifiable fragments (or 24.2% of the diet assuming an equal proportion of species in the unidentifiable categories). However, since this species was underestimated in the validation experiment by approximately three times, the actual contribution of *C. ciliaris* may range from 14.5% to 43.4% (even up to 72.7% if unidentifiable fragments are considered) due to potential errors resulting from observer interpretation. These potential errors will occur for every species but are particularly important for the preferred species such as *Aristida* spp. and *Enneapogon* spp., both overestimated in the validation experiment, and *C. ciliaris* which may be considerably underestimated. This is clearly an area that warrants further investigation, with very few studies of herbivore diets validating their findings.

3.4.2.3 Long-chain Alkane Analysis

The use of plant wax components in herbivore studies is becoming an accepted technique to determine herbage intake (e.g. Mayes *et al.* 1986, Laredo *et al.* 1991, Vulich *et al.* 1991, Vulich *et al.* 1995) and more recently, botanical composition of herbivore diets (e.g. Dove and Mayes 1991, 1996, Newman *et al.* 1995). Measuring herbage intake takes advantage of the relatively high recovery rates of long-chain alkanes in the faeces so that long-chain alkane patterns in the

faeces are almost identical to the long-chain alkane patterns of the ingested herbage. Dove and Mayes (1991) provide a detailed description of how the high recovery rates of long-chain alkanes and their taxonomic variation in plant epidermal wax (e.g. Eglinton *et al.* 1962, Dyson and Herbin 1968, Herbin and Robins 1969, Dyson and Herbin 1970) can be exploited to determine the botanical composition of consumed herbage. With development and refinement of the analytical methods and statistical models (e.g. Newman *et al.* 1995, Dove and Moore 1995), estimation of the composition of the diet of grazing animals using long-chain alkanes is becoming a practical alternative to other techniques (Dove and Mayes 1991, Dove 1992, Salt *et al.* 1994, Dove and Moore 1995).

The advantage of the long-chain alkane method over conventional microscopic analysis is two-fold. First, the accuracy of the long-chain alkane technique is high, because it is based on accurate analytical techniques rather than human interpretation. Second, the predicted diet does not have a component of unknowns as does the microscopic technique. The summing of the proportions of plant species in the diet to unity thereby provides a more realistic estimate of the contribution of each species to the diet, despite only including a limited number of plants in the analysis. These advantages were illustrated in the evaluation experiment.

Evaluation Experiment

In the evaluation experiment, ten individual plant species were collected to be used as the reference plants. These species were chosen because they represented the most abundant forage species at the time of collection and/or they had previously been documented in the diet of the NHN wombat. Analysis of canonical variates of duplicate samples revealed that species of the same genera (e.g. *Aristida* spp. and *Enneapogon* spp.) shared similar patterns of concentrations of alkanes, as did the two *Enneapogon* species and *Eragostris lacunaria*. However, even considering the similarity of plant species within the same genera and *Enneapogon* spp. and *Eragostris lacunaria*, the botanical composition of the NHN wombat diet was successfully partitioned into eight genera.

Statistical analysis demonstrated that the composition of the diet determined by microscopic analysis did not differ significantly from the prediction derived from long-chain alkane analysis. Comparisons of individual species, such as *Enneapogon* spp., demonstrated a high level of agreement between the two methods, (Table 3.6), despite the potential for over/underestimation of the species composition by the microscopic technique. Further, the contribution of *Cenchrus ciliaris* estimated by the long-chain alkane method may be a better representation of the level of consumption by the NHN wombat. This also applies to *Eragostris* sp., deemed to be the key forage species consumed by the NHN wombat in Flößer's (1986) study.

Of key interest in the comparison between the alkane method and the microscopic method is the contribution of *Salsola kalii*, a C_4 dicot characterised by needle-like leaves and a rapid growth phase following rain. The needle-like leaves make sampling for epidermal fragments very difficult. *Salsola kalii* also has a comparatively high total nitrogen content (up to 2.92%), is highly digestible (up to 84.5% IVDMD) (Appendix 1) and is consumed by other vertebrate grazers (e.g. sheep in New South Wales; H. Dove pers. comm.). Consumption of a species like *S. kalii* may remain undetected by microscopic analysis (because of high IVDMD) and $\delta^{13}C$ (it is a C_4 plant), but the alkane method clearly identifies it as a primary forage species of the NHN wombat. However, there is some variation in its consumption by individual NHN wombats. This species may be an important seasonal forage species for the NHN wombat, particularly as a source of high protein.

Potential Problems

The merits of using alkanes to estimate the diet of a free-ranging herbivore has yet to receive close scrutiny because of the infancy of the technique. The initial problem of the alkane method is in deciding which species are consumed and hence which species are to be used in the analysis. Dove and Mayes (1991) emphasise that there are considerable differences in alkane concentrations between species but also within species. For example, Dove *et al.* (1996) demonstrated that there is considerable variation in the patterns of alkane concentration between plant

species. They found that between the six pasture plants examined, 85% of the total variance in alkane concentration could be explained by using odd-numbered alkanes from C_{25} to C_{33}. Similarly, Laredo *et al.* (1991) demonstrated that alkane concentrations differ with season and part of the plant. For example, Laredo *et al.* (1991) harvested *Brachiaria deumbens* in summer (March) and winter (June) and examined the patterns of alkane concentrations in both the leaf and stem fractions. In summer, the sum of the alkane concentrations from C_{27} to C_{36} for the leaf fraction was 487 $mg.kg^{-1}$ DM compared to the winter value of 349 $mg.kg^{-1}$ DM. In comparison with the leaf fraction collected in summer, the sum of the alkane concentrations was much less for the stem fraction (129 $mg.kg^{-1}$ DM). These trends were repeated for other tropical grass and legume species examined by Laredo *et al.* (1991) and temperate pasture plants by Dove *et al.* (1996).

Ideally, field sampling should not only focus on potential plant species consumed by the herbivore but also the preferred part of the plant and/or growth stage of the plant. Sampling should also include replication to establish variation within species, particularly if spatial variation is a consideration. However, the application of the alkane method to free-ranging herbivores on a large scale (such as in the current study) imposes several limitations. The prime issue is the high cost of analyses, both financially and in time. Sampling protocols should therefore focus on which outcomes are of most importance. If determining botanical composition of the diet is the goal then sampling should focus on species assumed to be consumed by the herbivore (an inventory of species consumed may be obtained through other techniques such as microscopic analysis, direct observation etc.). However this may sacrifice determination of other factors such as spatial variability in the alkane concentrations of the reference collection or examining the diet at the level of parts of the plant. In the current study, plant species were chosen by its contribution to the diet estimated by microscopic analysis. Each plant species was harvested as a whole plant in duplicate from within the habitat of the NHN wombat. Therefore, the issues of spatial variability in the alkane concentrations of plant species on offer or variation in the parts of the plant consumed by the NHN wombat were ignored. Further, replicating the analyses for determining the alkane concentrations of plant species on offer was not possible

(hence PCA compared to CVA) and sampling focussed on whole plant sampling rather than specific parts or growth stages of the plant.

The long-chain alkane method is currently limited by the number of alkanes (C_{25} to C_{36}), and hence plant species, able to be included in the least-squares optimisation procedure. For a free-ranging herbivore, this may restrict the use of long-chain alkanes as the sole technique to estimate the composition of the diet, particularly if the diversity of plant species available to the herbivore is high. Instead a combination of techniques may be preferable. In the current study, the use of histological analysis identified the principal components of the diet of the NHN wombat and both long-chain alkane analysis and histological analyses were used to estimate the composition of the diet. A similar approach using long-chain alkanes with other techniques has been used successfully in other studies (e.g. Salt *et al.* 1994) and should be considered for studies of free-ranging herbivores.

For long-chain alkane analysis, the summing of the diet to unity needs to be considered in the design of the study. Both the least-squares optimisation and simultaneous equation procedures sum to unity the solution that best estimates the botanical composition of the diet (Table 3.7). Rather than a problem with the long-chain alkane methodology, this is an unavoidable consequence of the mathematical procedure. Although summing the solution to unity may affect the composition of the estimated diet, by combining histological analysis and long-chain alkane analysis the effect of this problem is considerably reduced. For example, in the PCA-derived methods (estimation procedures 2 & 3), two plant species may share similar alkane signatures. However, histological analysis may indicate that the NHN wombat may consume one of the species but not the other. By including in the least-squares optimisation procedure only plant species that are known to be consumed by the NHN wombat, the best possible estimate of the composition of the diet is generated.

Finally, alkane recoveries are not complete. Dove and Mayes (1991) demonstrated that the recovery of alkanes are dependent on chain length: C_{27} is less recoverable than C_{36}. In the ruminant (sheep) the faecal recoveries range from 0.59 for C_{27} to 0.92 for C_{36} (Dove and Mayes 1991), and 0.91 to 0.97 for the same

alkanes in an animal with monogastric digestion (e.g. a pig; Dove pers. comm.). Although a simple correction factor can overcome this problem, it still should be considered as a potential source of error. Specifically for wombats, the recovery of alkanes requires validation because the assumption that recoveries are more comparable to pigs than sheep may be an overestimate. This is because the long exposure of ingesta to microbes and the reductive is comparable to rumenal fermentation (P. Barboza, pers. comm.).

3.4.3 Summary

This study confirms the conclusions of Flößer (1986) that the NHN wombat is a generalist rather than a specialist in its food choice.. The NHN wombat consumed more than 25 plant genera as measured by conventional microscopic analysis). The generality of the diet is also confirmed with the bulk of the diet consisting of *Aristida* spp., *Enneapogon* spp., *Cenchrus ciliaris* and *Fimbristylis dichotoma*. These species represent the most common species, or from the NHN wombat's perspective, the most frequently encountered species (Chapter 2). No clear seasonal changes in diet composition were evident suggesting that forage choice was not influenced to any great extent by temporal changes in forage quality or quality. However, the techniques used to quantify the diet had many potential errors and subtle responses to temporal changes in forage quality and quantity may not have been detected.

In a comparison between the two previous studies of the diet of NHN wombat and the current study, the botanical composition of diet had changed considerably. Rather than reflecting a change in forage choice, the changes in the species composition of the NHN diet reflect long-term changes in the availability of forage between the previous two studies and the current studies. In the short-term (covered by this study), diet choice by the NHN wombat was relatively constant with little temporal variation or variation between individuals.

Each technique used to describe the diet of the NHN wombat in this study has its own advantages and disadvantages. Stable carbon isotope analysis does not measure diet composition. It is limited to partitioning the assimilated diet or faecal residues into C_3 and C_4 components. Histological analysis is effective in providing

an inventory of plant species consumed but potential errors associated with human interpretation and large proportions of unidentifiable components reduce the precision of the technique. Studies using microscopic analysis to describe the species composition of the herbivore diet (*cf* plant groupings - grasses, forbs etc) should only be used as a guide rather than the definitive solution. Similarly, when the number of forage species available to the herbivore exceeds the number of alkanes able to be used in the analysis, long-chain alkane analysis should only be used as a guide to describe the relative proportion of diet components. The potential of long-chain alkane analysis for describing herbivore diets clearly exists, but the technique requires considerable refinement particularly when investigating free-ranging herbivores grazing in a heterogenous forage environment. Finally, because both $\delta^{13}C$ and histological techniques use faecal residues to estimate the diet, they are subject to the influence of differential digestibility. Although long-chain alkane analysis also uses faecal residues, the issues of differential digestibility are of reduced concern (Dove and Mayes 1991).

The methods used to examine the diet of the NHN wombat (and the eastern grey kangaroo, Chapter 6) were as comprehensive as possible, and therefore provide the best possible account of the diet of the NHN wombat. However, this description of the diet of the NHN wombat is only valid for the forage composition described in Chapter 2. As the forage environment at EFNP changes, the diet composition of the NHN wombat will need to be continually monitored. Further, with the development of the long-chain alkane technique and the development of other methods (e.g. near infra-red spectroscopy), the accuracy for measuring the botanical composition of the diet of herbivores can only improve.

CHAPTER 4

BODY COMPOSITION AND BODY CONDITION OF HAIRY-NOSED WOMBATS

4.1 INTRODUCTION

4.1.1 Background

Body composition and body condition are terms used to describe the wellbeing of vertebrates. Body composition is defined as the quantification of ingesta-free components of the animal: water, fat, protein and ash or minerals (Robbins 1983). An accurate measurement of body composition can be obtained only through direct carcass analysis but it can also be estimated indirectly. The definition of body condition is more problematic. It relies on the assumptions that an attribute of 'condition' exists in an individual and can be measured (Krebs and Singleton 1993), that body condition is a good indicator of nutritional status (Anderson *et al.* 1990, Batzli and Essecks 1992) and that individuals of higher body condition have increased fecundity, lower mortality and are better able to cope with whatever environmental conditions are encountered (Brochu *et al.* 1988; Krebs and Singleton 1993). Body condition can also refer to the physical appearance of the animal. For the purpose of this study, body condition is defined as variation in body composition from a mean value.

Most studies on body composition and condition focus on body fat and assume that fat is directly related to the overall energy balance of the animal. Also, fat storage is considered to be favoured by natural selection, particularly if the organism is likely to experience food shortages (Batzli and Essecks 1992).

Measuring body composition and condition is a valuable tool for monitoring physiological and physical response of an organism to changes in climate (Aggrey 1982), environmental quality (Gales *et al.* 1994) and life cycle stages. Changes in body

composition also allow identification of critical times or events in an animal's life cycle (Farley and Robbins 1995). These have particular management implications since body composition of an organism may directly influence growth, reproduction, survival, and/or the overall dynamics of a population. Measuring body composition may also allow an understanding of physiological adaptive strategies for survival (Barnett *et al.* 1979).

Body composition and condition measurements of the NHN wombat have added significance because of the wombats' endangered status, behavioural and physiological ecology, and habitat. The food resource of the wombat is highly variable, with both seasonal and annual fluctuations in forage quality (Chapter 2). Identifying and measuring changes in body composition and body condition within and between individuals may be crucial in identifying adaptive strategies for survival in the tropical savanna. This is the primary objective of this chapter.

4.1.2 Techniques for Measuring Body Composition and Body Condition

Body condition has traditionally been measured non-destructively using various ratios of body mass to body dimensions (Krebs and Singleton 1993; Norgan 1994; Arnould 1995) and visual estimates of condition (Gallivan *et al.* 1995). Other estimates of body condition require destructive sampling, and include the kidney fat index (Holand 1992; van Rooyen 1993), fat content of bone marrow (Ransom 1978) and depth of fat on the back (Brown *et al.* 1995), as does the direct measurement of body composition. Clearly, destructive techniques for the measurement of body condition and composition are inappropriate in many ecological studies.

Indirect studies of body composition which estimate body water pool size by *in vivo* isotope dilutions, are more common and rely on the fact that the size of the water pool as a percentage of a body is generally inversely proportional to the percent or mass of specific body fat in many species (Sheng and Huggins 1979). Water enriched in isotopes such as tritium (^3H_2O), deuterium (^2H_2O) and oxygen-18 ($H_2{}^{18}O$) offers reliable measurements of body water in many species, but may not apply to large herbivores where a large proportion of total body water is incorporated in the contents of the gastrointestinal (GI) tract and thus is independent of body composition.

Development of fast and reliable measures of body composition have received an increasing amount of attention with the use of bioelectrical impedance analysis (BIA) and total body electrical conductivity (TOBEC). BIA, originally developed for the study of human body composition, has recently been applied successfully to a number of domestic animals such as lambs (Berg and Marchello 1994), pigs (Swantek *et al.* 1992) and cattle (Marchello and Slanger 1994), and has also been used in ecological and management studies on seals (Gales *et al.* 1994; Arnould 1995) and bears (Farley and Robbins 1994). BIA offers the promise of a rapid, portable, and relatively inexpensive means of predicting body composition. It operates under the premise that the conduction of an electrical current is dependent upon body fluids and electrolytes with the conductivity being greater in fat free mass than body fat (Lukaski *et al.* 1986, Kushner 1992). BIA is therefore an estimate of total body water and must include the large proportion of body water associated with the gastrointestinal tract of herbivores.

The rapid, non-destructive means of measuring body composition has appeal to measure and quantify body composition in the NHN wombat. However, BIA has yet to be applied in an ecological study of a large free-ranging herbivore such as the NHN wombat. Therefore, the accuracy of BIA, along with other measures of body composition and body condition, need to be assessed before the methodologies can be successfully applied to the NHN wombat. The southern hairy-nosed (SHN) wombat, *Lasiorhinus latifrons*, is ecologically and morphologically similar to the NHN wombat and offered the ideal subject to calibrate body composition and body condition methodology. Once calibrated, BIA could then be applied to the NHN wombat.

4.2 A Calibration of Methods

4.2.1 Methods

4.2.1.1 Study Animals

Fifteen Southern Hairy-nosed (SHN) wombats (eight females, seven males) were captured by 'stunning' (Robertson and Gepp 1982). Two collections of wombats were undertaken, one in spring (September 1994; eight animals - four females and four males) and one in summer (January 1995; seven animals - four

females and three males) to maximise the range of body composition included in the study. A high velocity .22 calibre bullet was fired just above a wombat's head while the animal was held in the beam of a spot light. As the wombat could not identify the direction of approaching sounds, allowing a chaser with a net to capture it. Once captured, wombats were placed in sacks, weighed and anaesthetised (Zoletil™ 100, 5 $mg.kg^{-1}$ via intramuscular injection). Body length, head length, chest girth, pes length and tibia length were measured. The animals came from the Murraylands population of SHN wombats in South Australia over which a destruction permit had been issued by South Australian National Parks and Wildlife Service, for reasons not related to the research objectives.

Animals were held for measurement of isotope dilution space and bioelectrical impedance, and then killed by an injection of a sodium pentobarbitone (Lethabarb™). Each animal was shaved, the gastrointestinal (GI) tract (including contents) removed, and the carcass was stored at -20°C. Mesentery associated with the GI tract was removed and kept with the carcass. Differences in body fat and body mass between the sexes and collection time were examined by ANOVA.

4.2.1.2 Measurement of Total Body Water of Living Animals

Total body water pool was determined by isotope dilution, using water labelled with deuterium (^2H_2O) and oxygen-18 ($H_2{}^{18}O$). Blood samples (1 ml) were collected from the cephalic vein of anaesthetised wombats prior to isotope injection to provide background levels of ^2H_2O and $H_2{}^{18}O$. Isotopes (0.5 ml) were injected intra-peritoneally and allowed to equilibrate for four hours. Previous studies have shown that equilibration of the isotope with the body water pool occurs within this time using 0.5 ml of isotope (Murray Evans, pers. com.). After equilibration a further 1 ml of blood was collected for determination of isotope dilution space.

To determine whether the isotopes equilibrated evenly throughout the body, 1 ml samples of gut water were obtained at the time of dissection by syringe from seven individuals and compared to the blood values by a paired t-test. Standard isotope ratio mass spectrometry methodology was used to analyse isotopes, after blood samples had been vacuum distilled to dryness following the

methods of Vaughan and Boling (1961). Least squares linear regression was used to determine the relationship between total body water measured by isotope dilution and chemical composition of the carcass.

4.2.1.3 Chemical Analysis

Chemical analysis of the carcasses was performed by the Victorian Institute of Animal Science, Werribee. The carcass, minus the gastrointestinal (GI) tract and its contents, was homogenised in a whole animal grinder. Subsamples (100 g wet weight) of the homogenate were freeze-dried to determine dry matter content and consequently the total water content of the carcass. Analyses were performed in triplicate. Subsequent chemical analysis (protein, fat and ash: g/100g), was performed on the freeze-dried homogenate. Carcass fat was determined via AOAC Official Methods (Helrich 1990) using petroleum ether extraction in a soxhlet apparatus. Nitrogen (N) was determined by a semi-micro Kjeldahl technique with a selenium catalyst, with crude protein calculated as N x 6.25. Recoveries of ammonium sulphate standards were in the range of 99.7% to 101.6%. Ash content was determined by combustion in a muffle furnace at 550°C.

The GI tract was analysed separately. Contents of the GI tract were removed, the empty GI tract was weighed and then homogenised in a commercial meat grinder. Sub-samples of the homogenate were freeze dried to determine the dry matter content. Further subsamples were ground to a fine powder in liquid nitrogen using a mortar and pestle. Freezing in liquid nitrogen reduced potential loss of fat from the subsample by creating a brittle substance allowing efficient grinding. Protein and ash were analysed using the techniques described above. Fat was determined by petroleum ether extraction in a soxhlet apparatus. Each soxhlet thimble was oven dried at 105°C for a minimum of 6 hours, allowed to reach room temperature in a vacuum desiccator and then weighed. Five grams of dried sample was then placed in the thimble, which were subsequently dried to a constant mass at 60°C for dry weight determination. Duplicate samples were then extracted in petroleum ether (boiling point 40 to 60°C) for 12 hours. Thimbles were dried at 60°C overnight, and after cooling in a vacuum desiccator were weighed. Fat content was calculated by gravimetric difference. Extraction time was determined

by performing a time series (9, 10, 12, and 16 hours extraction time) on subsamples of the ground carcass where fat content had previously been determined on carcass samples. Duplicate hair samples were analysed for protein, fat (using extraction times determined for GI tract) and ash in the same manner as the GI tract. Carcass, GI tract and hair analyses were combined to provide measures of total body composition.

4.2.1.4 Bioelectrical Impedance Analysis

Bioelectrical impedance of all fifteen SHN wombats was recorded with a four terminal bioelectrical impedance plethysmograph (model BIA-101A, RJL Systems, Detroit, Michigan). This instrument delivers an 800 μA current at 50 kHz and measures resistance and reactance of the current. Anaesthetised wombats were placed in a sternally recumbent position. Vacutainer® needles were inserted subcutaneously laterally along the midline of the wombat in a similar configuration to that used by Swantek *et al.* (1992) and Gales *et al.* (1994). The first needle was inserted four centimetres from the base of the cranium towards the tail, the second needle four centimetres from the first along the midline. The fourth needle was inserted seven centimetres from the tail and the third needle four centimetres along the midline from the fourth needle. The four terminals of the plethysmograph were attached to the needles, allowing the current to pass through the animal. The precision of these procedures was determined by repeating the measurements five times on seven of the wombats: after each measurement, electrodes were removed, the wombat re-orientated 180° and four new electrodes appropriately positioned. Resistance (R) and Reactance (Xc) of the current were recorded. These values can be used to calculate Impedance (Z) where:

$$Z = (R^2 + Xc^2)^{1/2} \qquad \text{(equation 4.1)}$$

Further, from Kushner's (1992) review of the principles of BIA on humans, an equation for the 'electrical volume' (V) can be derived incorporating the length (L), cross-sectional area of the conductor (i.e. the wombat's body), and the applied signal frequency (ρ: the specific resistivity, ohm-cm):

$$V = \rho L^2/Z \quad \text{(equation 4.2)}$$

The relationship between percent body fat or percent body water and BIA measurements such as resistance, impedance and electrical volume were assessed by least squares linear regression. Further analysis, using multiple linear regression, generated a predictive model for both total body water and total body fat using resistance, impedance and total body mass as predictive variables.

4.2.1.5 Body Condition Indices and Body Condition Score

The accuracy of three standard body condition indices to estimate body fat and body protein were assessed by least squares linear regression. The first index of condition assessed was that described by Krebs and Singleton (1993). The second condition index assessed was the body mass index (BMI) (Norgan 1994) and its derivatives. The third index, closely related to the previous two, uses the pes length as the skeletal measurement (Bakker and Main 1980).

The Krebs and Singleton (1993) index is the ratio of observed mass to that predicted from a standard relationship between skeletal size and mass. Two methods were used to calculate the predicted mass value. The first used the relationship between body mass and total body length of southern hairy-nosed wombats calculated from unpublished data (Matt Gaughwin, Joe Stelman and Rod Wells). This data represented 268 SHN wombats captured by trapping and stunning from 1964 to 1989 and will be referred to as the "population data". These were: 155 Females and 113 Males; 7 Juveniles, 37 sub-adults and 224 adults using Gaughwin's (1981) classification for the ages of SHN wombats. The second method used the body mass and length data of the experimental animals (N = 15).

The BMI is calculated by dividing body mass by length (Norgan 1994; Arnould 1995) (equation 4.3), or by length raised to the power of 2 (equation 4.4) or 3 (equation 4.5).

$$BMI = M/L \quad \text{(equation 4.3)}$$

$$BMI = M/L^2 \quad \text{(equation 4.4)}$$

$$BMI = M/L^3 \quad \text{(equation 4.5)}$$

where BMI is the body mass index; M is body mass; L is total body length.

Similarly, the Bakker and Main (1980) index is calculated by:

$$CI = M/PL \quad \text{(equation 4.6)}$$

$$\text{or, } CI = M/PL^3 \quad \text{(equation 4.7)}$$

where CI is the condition index; M is body mass; PL is the pes length. The cubed pes length in equation 4.7 allows individuals of differing body sizes, and hence pes lengths, to be compared evenly (Bakker and Main 1980).

A more subjective body condition score (BCS) was also evaluated as a predictor of body fat and body protein. A consensus between observers familiar with the range of body condition of wombats gave a condition score to the wombat when BIA measurements were taken. The BCS (Table 4.1) summarises a series of attributes which are assumed to indicate covering of the skeleton with muscle and fat, these being judged visually or by palpation. Condition scores of this kind are widely used amongst wildlife managers.

4.2.2 Results

4.2.2.1 Chemical Composition

Chemical analysis accounted for 100.4 ± 1.1% of the original wombat live body mass, with the largest component being water (Table 4.2). The gastrointestinal tract, including contents, accounted for between 10.3% and 19.6% of total body mass, whereas the actual contents of the gastrointestinal tract accounted for between 4.8% and 16.6% of total body mass. Percent body fat varied widely between individuals with a range of 2.6% to 19.3% of body mass. Body mass was not significantly different between the sexes ($F_{1,13} = 2.06$; $p = 0.18$) or between collections ($F_{1,13} = 0.62$; $p = 0.44$). Body fat was not significantly different between sexes ($F_{1,13} = 3.62$; $p = 0.08$) but did vary between collection times ($F_{1,13} = 7.45$; $p = 0.02$) with animals fatter in January than September. A

Table 4.1. Subjective body condition score (BCS) for wombats.

Score	Description	Condition
1.0	• Ribs visible	Emaciated
2.0	• Ribs covered, not sticking out but easily felt • Vertebrae sharp, obvious laterally, can feel sides • Easy to clasp around pelvis • Sunken rump	Poor
3.0	• Intermediate	Good or "normal"
4.0	• Pelvis well covered	Very Good
5.0	• Captive, well fed wombat	Fat

Developed by Deryn Alpers (University of New South Wales) and Alan Horsup (Queensland Department of Environment) for the NHN wombat (pers. comm.).

Table 4.2. Mean chemical composition of southern hairy-nosed wombats as a percentage of total body mass (n = 15).

		Protein (%)	Ash (%)	Fat (%)	Water (%)	Recovery[1] (%)
Carcass	Mean	21.5	2.7	9.2	66.2	99.6
	S.D.	1.1	0.4	6.0	5.0	0.05
GI Tract	Mean	12.8	1.2	9.7	74.5	98.2
	S.D.	1.9	0.4	6.6	6.6	1.7
Hair[2]	Mean	99.6	3.6	0.7	-	101.2
	S.D.	2.1	1.4	0.7	-	1.3
Whole Body[3]	Mean	18.9	2.3	8.1	66.5	100.4
	S.D.	1.0	0.3	5.5	4.6	1.1

[1]Mean recovery for each individual.

[2]Hair was assumed to have a negligible water content.

[3]Dry matter of gut contents account for 2.8 ± 0.9% of body mass; removed teeth account for 1.5 ± 0.7% of body mass.

significant linear relationship was found between percent body fat and percent body water as determined by direct chemical analysis (Figure 4.1). The percent hydration of lean body mass (LBM) in southern hairy-nosed wombats was 72.6 ± 1.6% using the assumption that body fat is completely water free. This value is not significantly different from Pace and Rathbun's (1945) mean value of 73.2% for a range of mammals including rats, guinea pigs, rabbits, cats, dogs and monkeys.

4.2.2.2 Isotope Dilutions

Both isotopes, ^2H_2O and $H_2{}^{18}O$, provided good predictions of total body water (Figure 4.2). Dilution of deuterated water was found to overestimate total body water by 5.0 ± 2.9% and dilution of $H_2{}^{18}O$ was found to underestimate total body water by 2.2 ± 2.7%. The regression coefficients of the two lines did not differ significantly ($t_{15} = 0.104$, $p > 0.05$) but the intercepts were significantly different ($t_{16} = 3.155$, $p < 0.05$). Body water pool estimated by isotope dilution using blood samples did not differ significantly from that estimated using gut water samples ($t_7 = 1.439$, $p > 0.05$) suggesting complete or near complete isotope equilibration with all body water. Isotope dilutions with ^2H_2O provided a precise measure of body fat with 83% of the variation in body fat explained by variation in isotope dilution space (Figure 4.3). The relationship between body fat and body water by $H_2{}^{18}O$ dilution was not significant ($p > 0.05$).

4.2.2.3 Bioelectrical Impedance Analysis

Of the bioelectrical impedance measurements collected, resistance was found to be the best predictor of both body fat ($y = 0.58x - 23.02$; $r^2 = 0.63$; $F_{1,12} = 20.03$; $p = 0.0008$) and body water ($y = 91.04 - 0.45x$; $r^2 = 0.53$; $F_{1,12} = 13.67$; $p = 0.003$). The precision of the resistance measurements ranged between 1.86% to 8.94% in coefficient of variation. There were significant relationships between impedance (Z) and total body fat ($y = 0.56x - 22.61$; $r^2 = 0.59$; S.E. = 3.67; N = 14; $F_{1,12} = 17.03$; $p = 0.0014$), and Z with total body water ($y = 90.88 - 0.44x$; $r^2 = 0.51$; S.E. = 3.41; N = 14; $F_{1,12} = 12.28$; $p = 0.004$). No significant relationship was found between total body fat and electrical volume (V) ($F_{1,12} = 1.40$; $p = 0.26$) or between total body water and electrical volume ($F_{1,12} = 0.97$; $p = 0.34$). Multiple linear regression produced predictions of good precision for total body fat (Table

Figure 4.1. The relationship between percent body fat and percent body water determined by desiccation of a sub-sample of the whole carcass of fifteen southern hairy-nosed wombats. The equation of the line is $y = 79.23 - 1.07x$; $r^2 = 0.97$; S.E. = 1.00; N = 15; $F_{(1,13)} = 410.98$; $p = 3.2 \times 10^{-10}$.

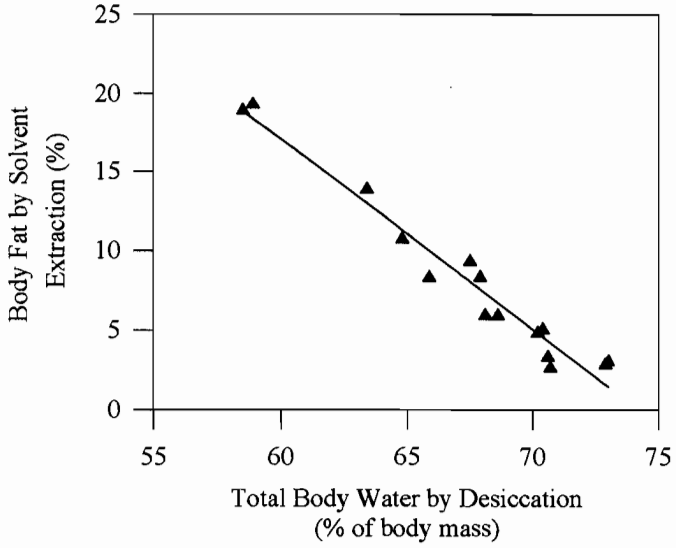


Figure 4.2. The relationship between total body water (litres) measured by desiccation and the isotope dilution space of ^2H_2O and $H_2{}^{18}O$ of fifteen southern hairy-nosed wombats. $H_2{}^{18}O$ samples were limited to six samples due to cost of the isotope. Part of the injection volume was lost in one case and these data for both isotopes have not been included here. The equation of the line for total body water and total body water predicted by dilutions of deuterium is $y = 1.20 + 0.85x$; $r^2 = 0.97$; S.E. = 0.43; N = 14; $F_{(1,12)} = 397.79$; $p = 1.44 \times 10^{-9}$. The equation of the line for total body water and $H_2{}^{18}O$ predicted total body water is $y = 3.42 + 0.79x$; $r^2 = 0.95$; S.E. = 0.44; N = 5; $F_{(1,3)} = 52.5$; $p = 0.005$.

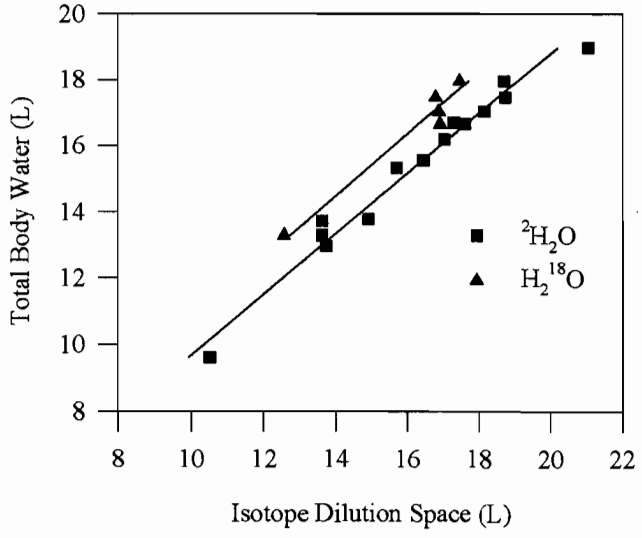
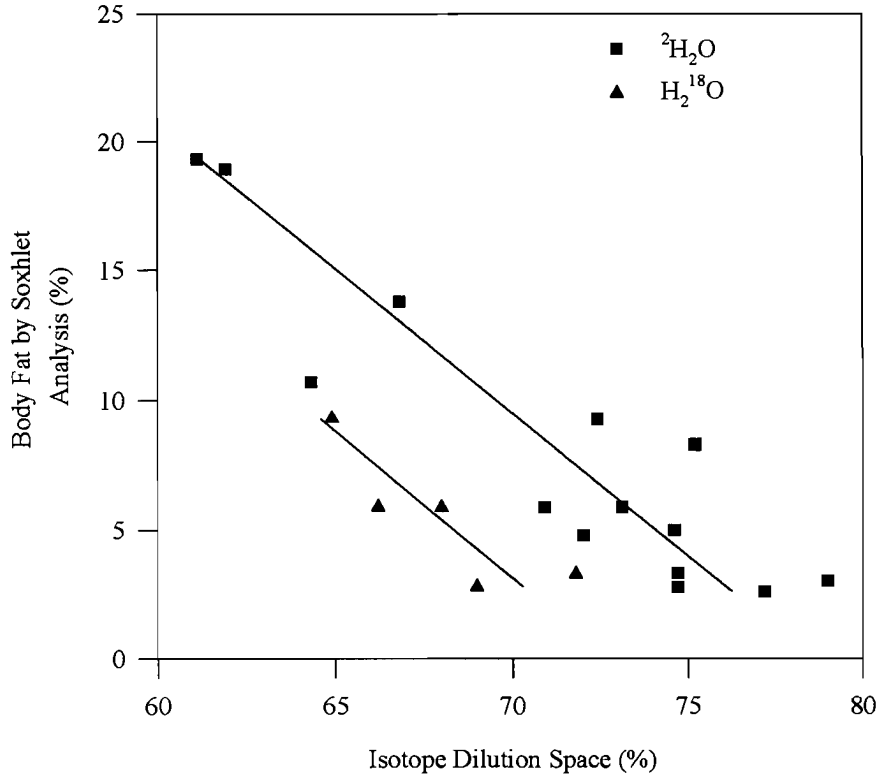


Figure 4.3. The relationship between percentage body fat measured by extraction of homogenised carcass samples with petroleum ether and percent total body water by isotope dilution space measured by $^{2}H_2O$ and $H_2{}^{18}O$ for fifteen southern hairy-nosed wombats. The equation of the line for body fat and body water by $^{2}H_2O$ dilution space is $y = 76.31 - 0.95x$; $r^2 = 0.83$; S.E. = 2.46; N = 14; $F_{(1,12)} = 58.60$; $p = 5.89 \times 10^{-5}$. The equation of the line for body fat and body water predicted by $H_2{}^{18}O$ dilution space is $y = 55.25 - 0.74x$; $r^2 = 0.70$; S.E. = 1.63; N = 5; $F_{(1,3)} = 7.15$; $p = 0.075$. While the relationship is not significant ($p > 0.05$) the illustrated line demonstrates the parallel relationship with $^{2}H_2O$.



4.3) using resistance, impedance and total body mass ($r^2 = 0.90$; $F_{3,10} = 29.22$; SE = 1.99; N = 14; p = 0.0000). Likewise, multiple linear regression produced predictions of good precision for total body water (Table 4.3) using resistance, impedance and total body mass ($r^2 = 0.90$; $F_{3,10} = 29.54$; SE = 1.64; N = 14; p = 0.0000).

Table 4.3. Multiple linear regression models for:
a) total body fat (%) of fifteen southern hairy-nosed wombats predicted by resistance (Ω) and impedance (Ω) (measured by BIA), and total body mass (kg) and;
b) total body water (%)of fifteen southern hairy-nosed wombats predicted by resistance (Ω) and impedance (Ω) (measured by BIA), and total body mass (kg). One sub-adult animal was eliminated from the analysis as it was hypothermic at the time of processing.

Dependent Variable	Variable	Estimate	S.E. of Estimate	*t*-value	Significance
a) Fat	Resistance	6.212	1.347	4.613	0.0010
	Total Body Mass	0.472	0.168	2.817	0.0183
	Impedance	-5.741	1.346	-4.267	0.0016
	Constant	22.293	4.834	-4.612	0.0010
b) Total Body Water	Resistance	-4.417	1.112	-3.973	0.0026
	Total Body Mass	-0.484	0.138	-3.496	0.0046
	Impedance	4.040	1.111	3.637	0.0046
	Constant	94.961	3.991	23.795	0.0000

4.2.2.4 Body Condition Indices and Body Condition Score

Southern hairy-nosed wombats differ in their morphological characteristics between individuals. Hence, in association with environmental factors over 25 years, the data set is variable but representative of southern hairy-nosed wombats.

The relationship between body mass and body length in the southern hairy-nosed population was significant ($F_{1,266} = 196.18$, $p = 0.00$), but only accounted for 42% of the variance in body mass being accounted for (the population) (Figure 4.4a). There was no significant difference in the relationship between males and females in body mass and body length (Figure 4.4b). The relationship between body mass and body length in sub-adults and juveniles was quite different from the adult population (Figure 4.4c). Although the body mass - body length relationship of sub-adults and juveniles was different to that of the adults, the sub-adult and juvenile captures represented only 16.4% of all capture records (N = 268). Therefore, the population data (Figure 4.4a) can be considered the most representative of the relationship between body mass and body length for all 268 SHN wombats, and will be used as the "population" data to calculate the Krebs and Singleton (1993) index of condition.

All 15 animals captured in the experimental group were classified as adults using Gaughwin's (1981) classification. The relationship between body mass and body length (Figure 4.4d), like the population data, was significant but only 53% of the variance in body mass was accounted for. There was no significant difference between either the population (Figure 4.4a) or the experimental animals (Figure 4.4d) in the relationship between body mass and body length. Therefore the animals in the experimental set can be considered "typical" SHN wombats on the basis of body mass and body length.

Condition indices were found to be less precise in predicting body fat than isotope dilutions and BIA. Although the relationships between condition index and percent body fat were linear for some indices (Table 4.4), only 4% to 38% of variance could be explained. Of the indices described, the body mass index was the best at predicting body fat (Figure 4.5). Other significant relationships were found for the pes length index (Figure 4.6), and the Krebs and Singleton (1993) index calculated using the 'population data' and the 'experimental data' (Figure 4.7). No relationship was found between condition score and percent body fat ($F_{1,11} = 0.95$; $p = 0.35$), or percent body protein ($F_{1,11} = 0.74$; $p = 0.41$).

Figure 4.4. The relationship between total body mass (kg) and total body length (cm) of southern hairy-nosed wombats for:
a) All capture records (population); $y = 0.38x - 9.75$; $r^2 = 0.42$; S.E. = 3.38; N = 268; $F_{1, 266} = 196.18$; $p = 0.00$.
b) Male and female southern hairy-nosed wombats in the population.
$y = 0.45x - 16.03$; $r^2 = 0.50$; S.E. = 3.30; N = 112; $F_{1,110} = 111.22$; $p = 0.00$ (Males) and
$y = 0.32x - 4.42$; $r^2 = 0.35$; S.E. = 3.36; N = 155; $F_{1, 153} = 83.41$; $p = 0.00$ (Females)
c) Age classes.
$y = 0.22x + 4.91$; $r^2 = 0.14$; S.E. = 3.10; N = 224; $F_{1,222} = 36.54$; $p = 0.00$ for adults;
$y = 0.79x - 42.8$; $r^2 = 0.25$; S.E. = 3.75; N = 37; $F_{1,35} = 11.61$; $p = 0.002$ for sub-adults;
$y = 0.65x - 30.21$; $r^2 = 0.76$; S.E. = 1.06; N = 7; $F_{1,5} = 15.52$; $p = 0.01$ for juvenilles.
d) Fifteen experimental animals in BIA calibration.
$y = 0.44x - 19.88$; $r^2 = 0.52$; S.E. = 3.14; N = 15; $F_{1,13} = 14.58$; $p = 0.002$.

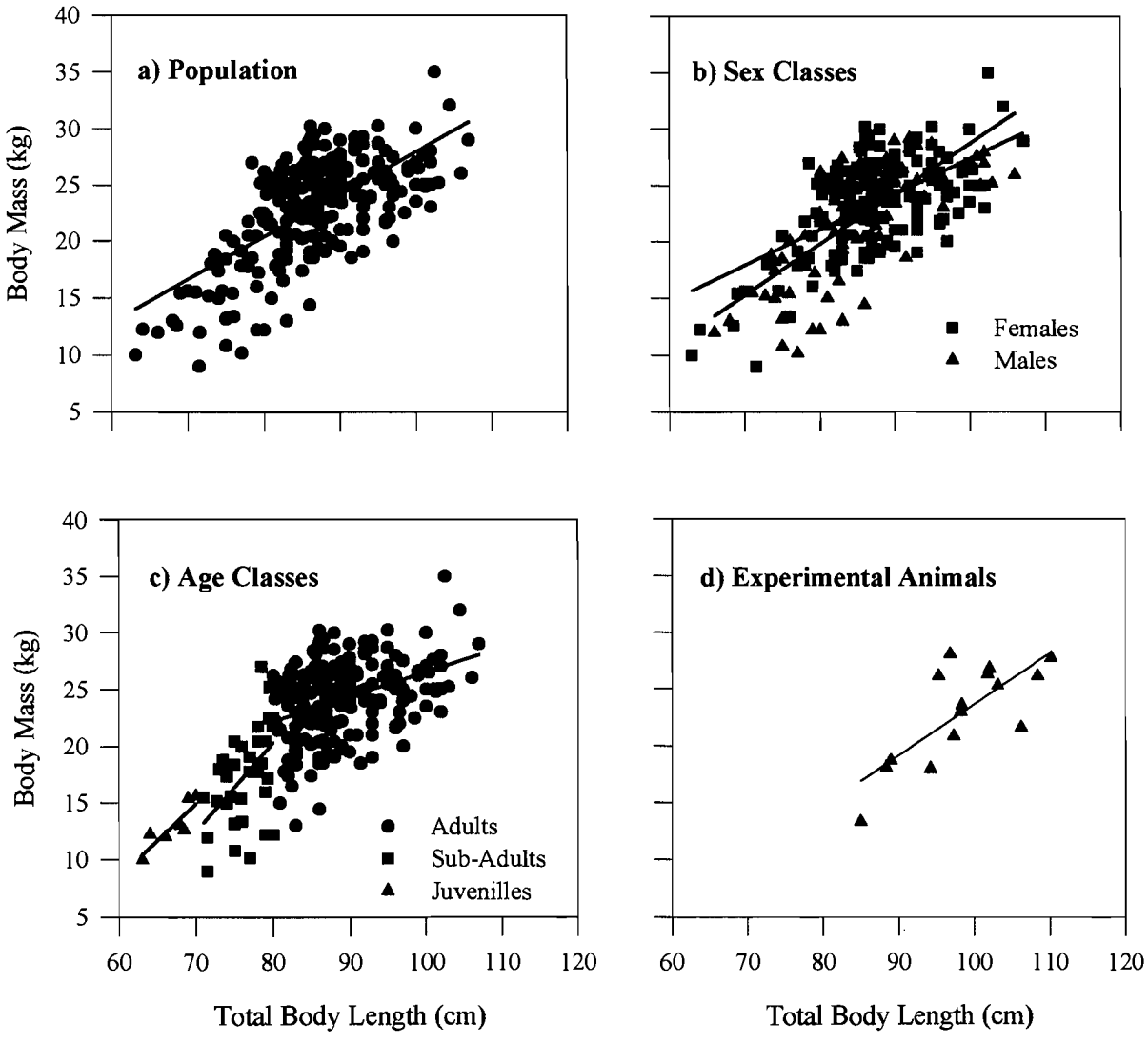


Figure 4.5. Relationship between percent body fat measured by extraction of homogenised carcass samples of southern hairy-nosed wombats (N = 15) with petroleum-ether and the body mass index (BMI).
The equation of the line is y = 94.05x - 14.01; r^2 = 0.38; S.E. = 4.47;
N = 15; $F_{1,13}$ = 8.08; p = 0.01.

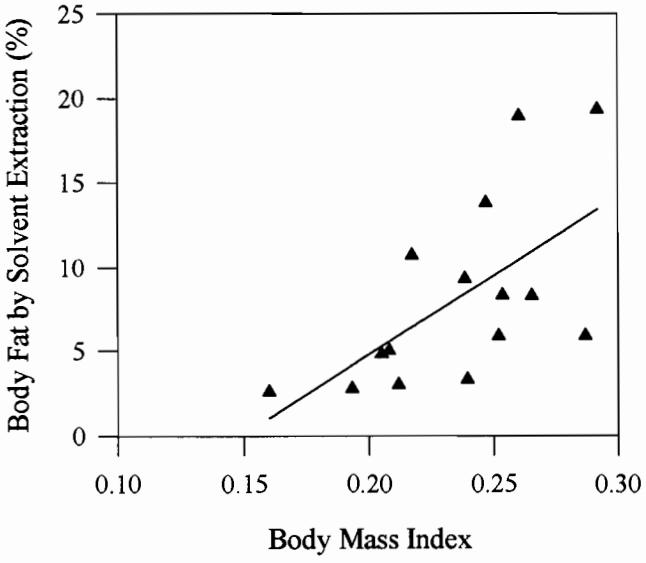


Figure 4.6. Relationship between percent body fat measured by extraction of homogenised carcass samples of southern hairy-nosed wombats (N = 15) with petroleum-ether and the pes length index.
The equation of the line is y = 7.96x - 10.40; r^2 = 0.30; S.E. = 4.75;
N = 15; $F_{1,13}$ = 5.65; p = 0.03.

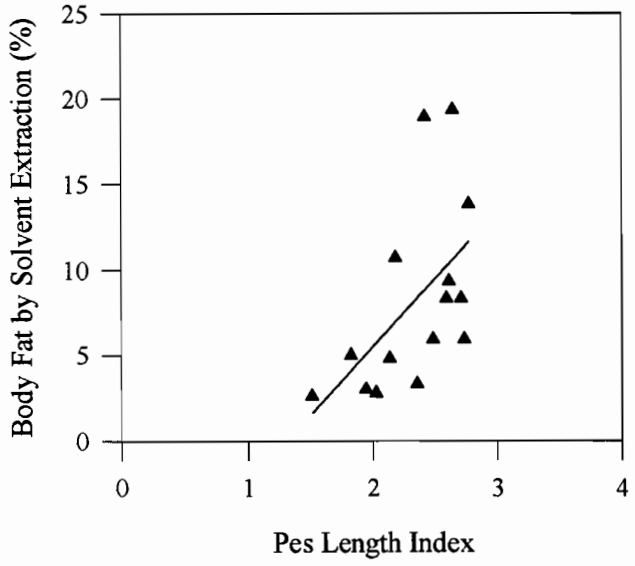
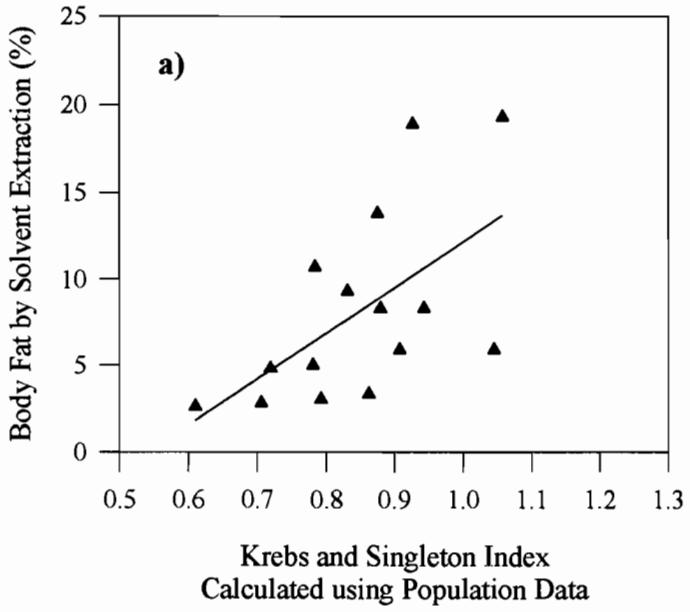


Figure 4.7. Relationship between percent body fat (measured by extraction of homogenised carcass samples with petroleum-ether) and the Krebs and Singleton (1993) index for:

a) All capture records (population data) of southern hairy-nosed wombats. The equation of the line is $y = 26.5x - 14.34$; $r^2 = 0.35$; S.E. = 4.59; N = 15; $F_{1,13} = 6.96$; $p = 0.02$.

b) The experimental animals (N = 15). The equation of the line is $y = 20.86x - 12.68$; $r^2 = 0.27$; S.E. = 4.87; N = 15; $F_{1,13} = 4.17$; $p = 0.05$.



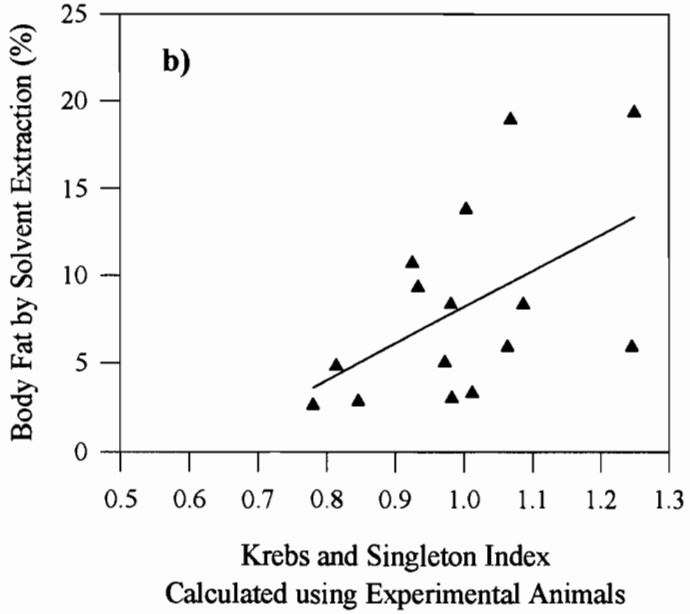


Table 4.4. Least squares linear regression analysis of indices of body condition with predict body fat and body protein for fifteen southern hairy-nosed wombats. Total body fat and total body protein were determined destructively by analysing the chemical composition of the fifteen southern hairy-nosed wombats (see text for details).

Index	Total Body Fat			Total Body Protein		
	r^2	F	Significance	r^2	F	Significance
BMI	0.38	8.08	0.01	0.05	0.67	N.S.
BMI (length component raised to the power of 2)	0.24	4.16	N.S.	0.03	0.42	N.S.
BMI (length component raised to the power of 3)	0.07	0.91	N.S.	0.01	0.11	N.S.
PI	0.30	5.65	0.03	0.01	0.19	N.S.
PI (length component raised to the power of 3)	0.04	0.59	N.S	0.02	0.25	N.S.
Krebs and Singleton (1993)	0.27	4.71	0.049	0.03	0.46	N.S.

Abbreviations: BMI - Body Mass Index; PI - Pes length index.

All F values have the degrees of freedom 1,13; and N = 15.

Significance value is $p < 0.05$

4.3 Application of Methodology to Northern Hairy-nosed Wombats

4.3.1 Methodology

Northern hairy-nosed wombats were trapped as part of a population study in 1993 (Dr Alan Horsup, Queensland Department of Environment) and to attach radio packages in 1995 and 1996 (described in Chapter 5). Wombats were trapped using methods described by Crossman (1988) using large, open ended, aluminium

traps. Fences were erected around the perimeter of a burrow complex, obliging the wombat to use their well-established runways. Traps were set on these runways. Once captured, the animal was anaesthetised by Zoletil™ anaesthetic delivered by intra-muscular injections (5 $mg.kg^{-1}$). Animals were measured (total body length, chest girth, body mass), blood samples taken, body condition scored and body composition measured by bioelectrical impedance analysis as described above.

Bioelectrical impedance and condition scores were recorded in the 1995/96 trapping program. A total of 25 measurements from 12 individuals were recorded during this period. In addition, the total body water pool of five individuals was measured using water labelled with deuterium (^2H_2O) and oxygen-18 ($H_2{}^{18}O$) as part of a water turn-over study (M. Evans, unpublished data). Total body water measurements obtained from Evans' study of wombat energetics were used to compare estimates of TBW measured by BIA.

The relationships between predicted total body fat (TBF) and total bimonthly rainfall, and predicted TBF and mean nitrogen concentration of the pasture were assessed using correlation analysis. Time effect was assessed by bimonthly time lags.

4.3.2 Results

4.3.2.1 Total Body Fat

Total body fat (TBF) of all NHN wombats exhibited little variation between individuals. The two lowest estimates of TBF predicted by the BIA multiple linear regression (MLR) model were 0.3% and 1.0% for two individuals. These two measurements are likely to be underestimates of the TBF for both individuals, as one individual was a juvenile and the integrity of both BIA measurements could not be guaranteed because of observer differences (positioning of electrodes by two independent observers may have differed). Ignoring these two results, the range of TBF (predicted by the BIA MLR model) encountered was 2.0% to 13.5%, with a mean TBF of $7.6 \pm 3.5\%$, comparing favourably to the whole body mean TBF for SHN wombats (Table 4.2).

Mean TBF in NHN wombats varied temporally, but did not reveal any clear trends (Figure 4.8). However, TBF was positively correlated ($p \leq 0.05$) with rainfall, two months after rain fell (Figure 4.9). A similar positive correlation of TBF with the mean total nitrogen concentration of available species of forage occurred with a lag of two months (Figure 4.9).

Each of the three methods (BIA, ^2H_2O and $H_2{}^{18}O$) produced different estimates of TBF (Table 4.5). Compared to ^2H_2O, both $H_2{}^{18}O$ and BIA tend to overestimate TBF (Table 4.5 & Figure 4.8) but most estimates of TBF fit within the relatively narrow range of actual TBF encountered in SHN wombats (range 2.6% to 19.3% TBF). This suggests that all three measurements are valid estimates of TBF for NHN wombats.

Table 4.5. Comparison of three methods for measuring total body fat in Northern Hairy-nosed wombats. The three methods are Bioelectrical Impedance Analysis using a multiple linear regression (MLR) predictive model, and isotopic dilution space measurement using the isotopes ^2H_2O and $H_2{}^{18}O$.

Animal ID	Collection Date	MLR Predicted Body Fat	^2H_2O Predicted Body Fat	$H_2{}^{18}O$ Predicted Body Fat
Male 32	21/09/95	5.95	6.7	11.84
Female 61	22/09/95	8.83	4.57	9.25
Male 25	19/06/96	13.49	7.47	9.22
Female 82	22/06/96	12.78	1.57	4.75
Female 152	21/06/96	9.67	9.42	9.22

There was limited temporal variation in body composition within individuals. Only three individuals were measured four or more times. For example, one male NHN wombat (Male #25) ranged from 4.7% to 13.5% TBF over a 14 month period. However, due to the narrow range of TBF for hairy-nosed wombats

Figure 4.8. Mean (± S.D.) bimonthly changes in body fat of northern hairy-nosed wombats predicted by BIA (N = 25), and isotopic dilutions of $^{2}H_2O$ and $H_2{}^{18}O$ (N = 5). Isotope data represents only two sampling periods.

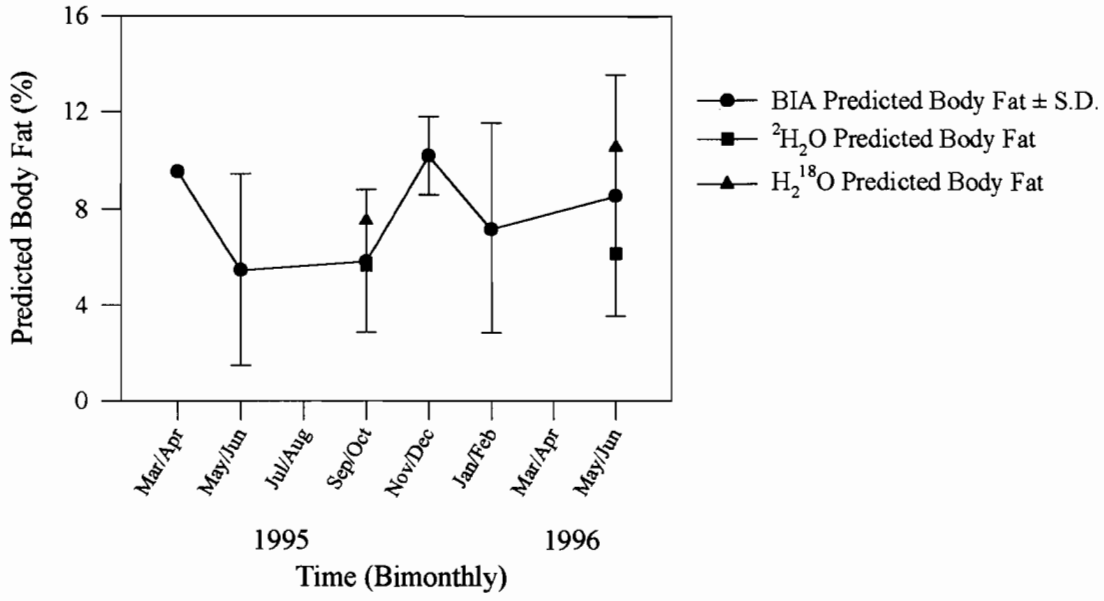
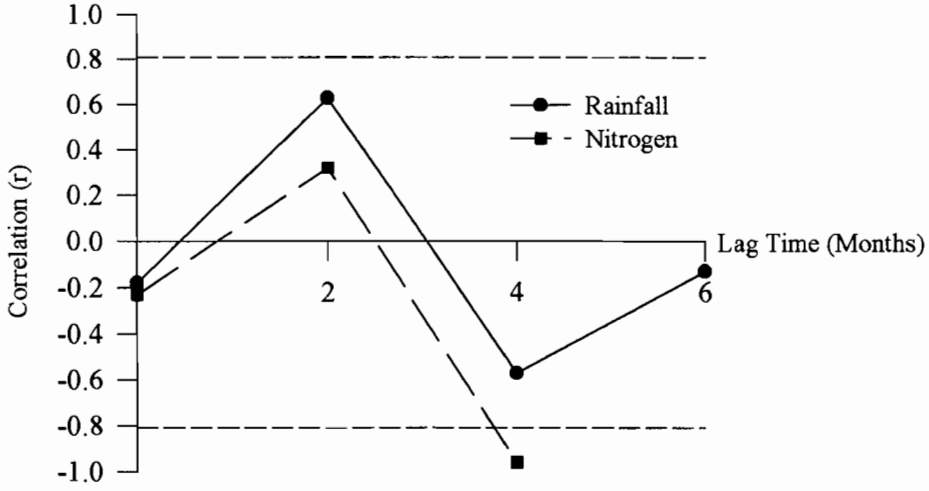


Figure 4.9. Correlation coefficients (r) of total body fat of the northern hairy-nosed wombat (estimated by BIA) over a lagged time-series of non-accumulative total rainfall and mean nitrogen concentration of forage species. [$r = 0.811$; $p < 0.05$; 4df]



in general and potential errors associated with gut water volume with BIA measurements (or even isotopes), defining temporal trends in TBF of individual hairy-nosed wombats is tenuous. This is best demonstrated by repeat measurements of body weight and BIA in a 15 day period during measurements of water turn-over (N = 2, one male (#32) and one female (#61)). Both individuals lost body weight during the study of water metabolism (1.7 kg for the male and 0.85 kg for the female) but increased in TBF (2.7% increase in TBF for the male and 6.72% increase in TBF for the female). The decrease in body weight may be attributed to variation in GI tract contents, rather than a stress response to trapping as suggested by Hoyle *et al.* (1995). The GI tract contents of free-ranging SHN wombats (N = 15) contributed between 4.8% and 16.6% of total body weight. Under the restriction of trapping which prevented foraging the two NHN wombats lost 5.75% (male) and 3.96% (female) of body weight - less than the equivalent difference between a full GI tract (16.6%) and an empty GI tract (0%). The empty GI tract would reduce the apparent hydration state of the animal, decreasing resistance during BIA plethsymography and produce elevated predictions of TBF.

4.3.2.2 Body Condition

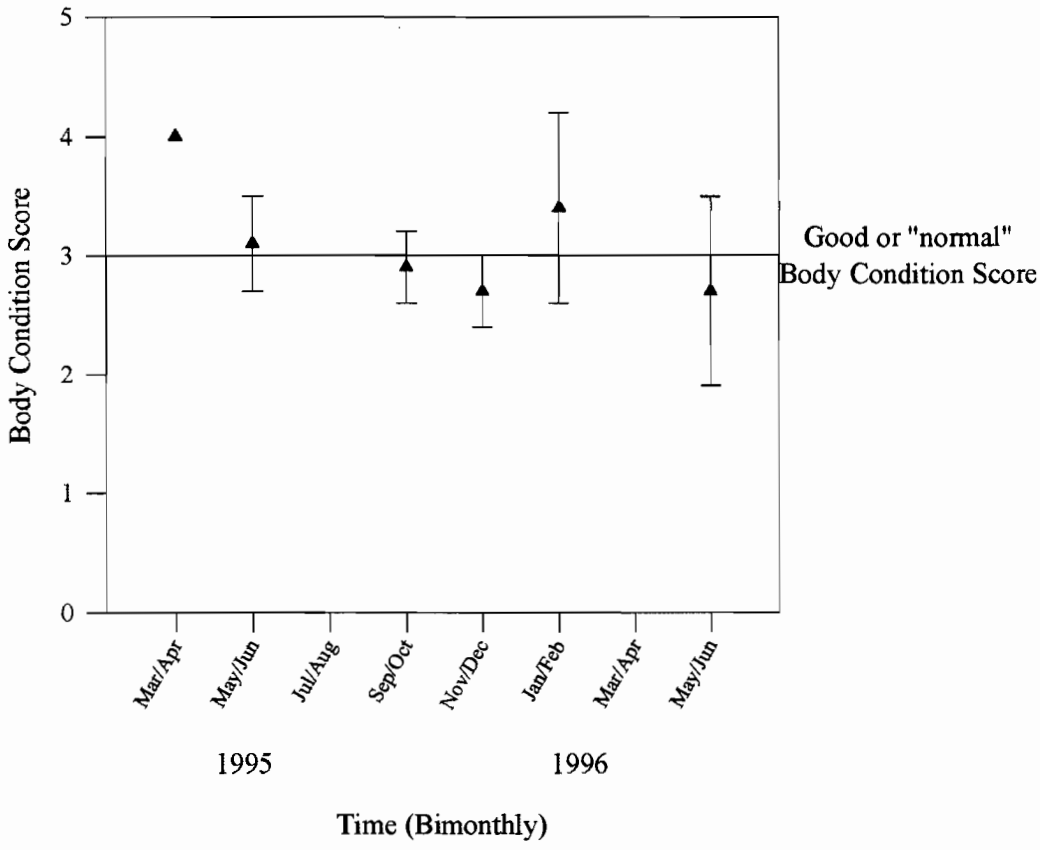
The qualitative BCS illustrates the general 'good' condition of the population (Figure 4.10). The BCS range from 2 to 4 for the NHN wombats examined in this study, following an assumed normal distribution (3 x '2'; 5 x '2.5'; 9 x '3'; 6 x '3.5'; 4 x '4'). Like body composition, there was slight temporal variation in the mean body condition score (Figure 4.10). However, the subjective nature of the score and relatively small sample size did not allow explicit seasonal comparison.

4.4 DISCUSSION

4.4.1 Body Composition and Body Condition of the Northern Hairy-nosed Wombat

Measurements of the body composition and body condition of the NHN wombat revealed that the population is in good condition relative to the fecund populations of SHN wombats that the measurements were calibrated against

Figure 4.10. Mean bimonthly (± S.D.) changes in body condition scores (BCS) for Northern Hairy-nosed wombats (N = 27).



(Figures 4.8 & 4.10). This is especially so considering that Epping Forest National Park was in a period of below average rainfall for the entire study. Johnson (1991b) investigated body condition of the NHN wombat by monitoring changes in body weight and blood parameters (blood haemoglobin (g/dl); packed cell volume (%); and red cell count ($x10^{12}$/l)) through the decline of forage quality throughout the year. He found that there was no evidence of a decline in any of the body condition measurements as the dry season progressed. However the value of using blood parameters to measure body condition is questionable. Blood parameters may only be useful when the herbivore is under nutritional restrictions (e.g. Del Giudice *et al.* 1992) or influenced by other factors such as those that cause deficiencies in energy and protein (e.g. parasite loads; Gallivan *et al.* 1995). Further, behavioural stress can also affect the value of blood parameters (e.g. Dickens 1976).

This study indicates that the NHN wombat may have the ability to maintain body composition, as well as body condition, throughout the sixteen month period when BIA measurements were taken encompassing wet and dry seasons. Further, trends in body composition of the NHN wombat are positively correlated with rainfall, and to a lesser degree the mean concentration of nitrogen of the pasture plants, with a two month time lag (Figure 4.9). This implies that the health status, inferred by body composition and condition, is intrinsically linked to rainfall. This is consistent with the findings of Crossman *et al.* (1994), that fecundity in females is positively correlated with rainfall. Therefore, it is highly likely that the number of breeding females should be correlated with body composition of all females.

Although the NHN wombat has the likely ability to maintain body composition and condition throughout the year, they maintain relatively low amounts of TBF compared to other mammals. Its low TBF, (mean 7.6% ± 3.5) suggests that its body composition may be negatively affected if forage quality, primarily total available nitrogen, falls below a critical level.

Observations on the SHN wombat suggest that when grasses become limiting in quality and quantity, the SHN wombat is capable of switching from

grasses to browse to maintain condition (Gaughwin 1981). Gaughwin *et al.* (1984) further suggest that short-term changes in the loss of body tissue are associated with seasonal changes in the food resource and these tissue losses may be overcome through storage of fat. The consistency of body composition and condition of the NHN wombat during this study suggests that they were not under nutritional duress. However, this issue requires further consideration, especially in the area of habitat management.

4.4.2 Reproductive Strategies of Northern Hairy-nosed Wombats: A Hypothesis

Reproduction, growth of a foetus and lactation exert considerable energetic demands on the mother (Oftedahl 1985). To meet the increased energy demands, the mother can either increase her food intake and/or mobilise energy reserves in the form of fat (Pond 1978, Oftedahl 1985), with the timing of the reproductive cycle in some environments intrinsically linked to the quality and quantity of food (Pond 1978, Angerbjörn 1986, Cameron *et al.* 1993, Atkinson and Ramsay 1995).

In large ungulates, body weight and body fat are greater in pregnant females than non-pregnant females (Cameron *et al.* 1993). This increase in body mass and body fat is necessary for both the survival of the off-spring and the survival of the mother (Oftedahl 1985, Cameron *et al.* 1993). There is also evidence that some species of marsupials rely on body fat to support the energy requirements for lactation (e.g. brush-tailed possums, *Trichosurus vulpecula*, Bell (1981)). It could therefore be hypothesised that the NHN wombat, foraging on grasses that vary significantly in quality and quantity, may also invest in body fat in order to successfully reproduce.

Supporting Evidence

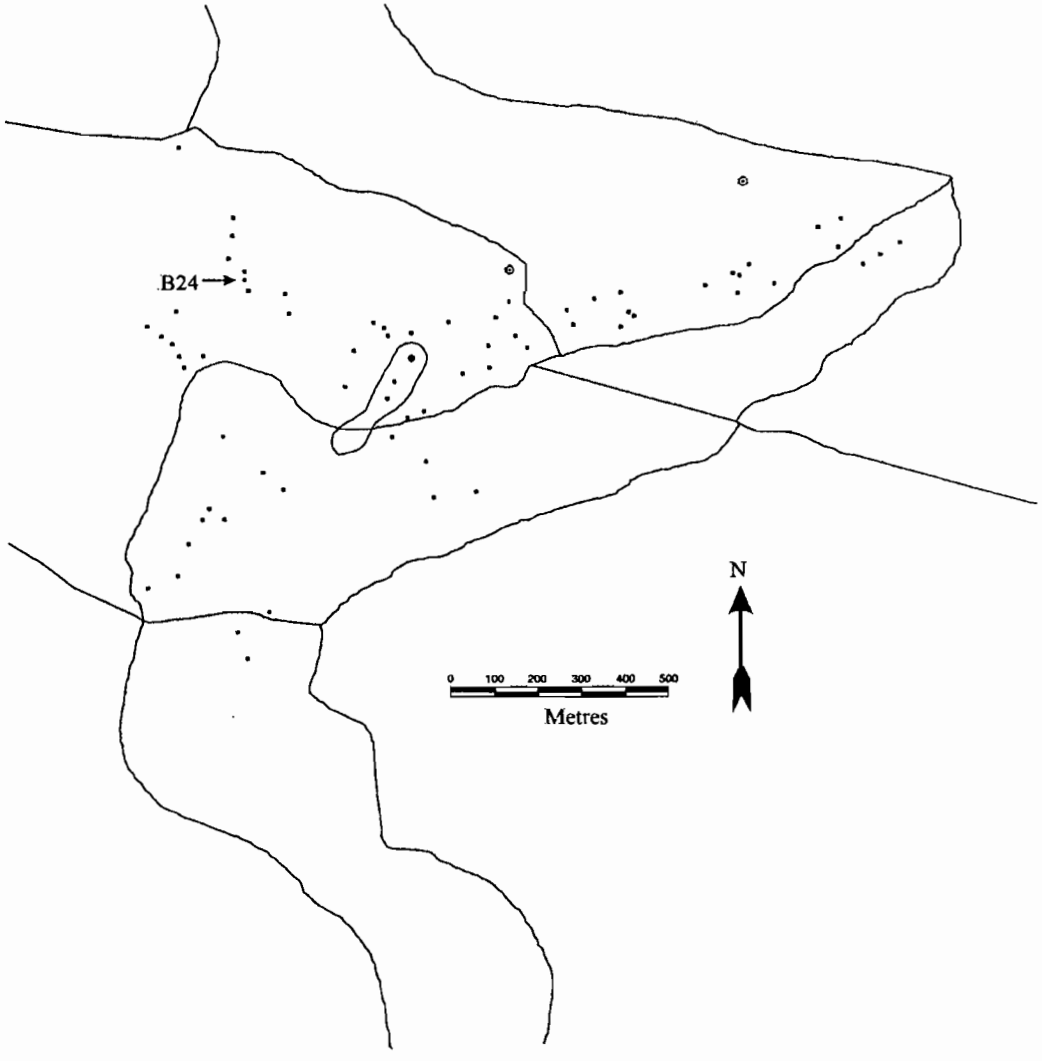
Evidence to support the hypothesis is limited by the population size and the dearth of knowledge regarding the life histories of any of the wombat species. However, information from one female NHN does provide preliminary support to this hypothesis.

In October 1993, female #82 had a capture body mass of 35.6 kg. In June 1995 she was recaptured with a body mass of 29.5 kg with signs that she had a young at foot (indicated by enlarged teat expressing milk and pouch condition). Although the identity of the off-spring has yet to be genetically confirmed, it seems more than likely that in the period between the two captures she successfully raised an off-spring. The loss of body mass between the two captures may be attributed to the energetic cost of reproduction.

Johnson and Crossman (1991) described the pattern of dispersal by the NHN wombat. Results of their trapping and radio tracking program from 1985 to 1989 show a strong tendency for adult females to disperse. They suggest the role of adult female dispersal is an important feature of wombat population dynamics allowing maximal range expansion in response to recruitment. Adult animals have the ability to establish new burrows and are less susceptible to stress than sub-adult and juvenile animals (Johnson and Crossman 1991). They further suggest that the dispersal of adult females is an indirect form of maternal investment, where the offspring inherits the natal burrow.

During the second capture of female #82 (June 1995), she was fitted with a radio collar and her home range measured (described in detail in Chapter 5). In order to measure the body composition of female #82 and check the condition of the radio collar, she was re-trapped in November 1995. In the period between August 1995 (following the radio tracking session) and November 1995 she had dispersed twice the diameter of her total home range from one burrow cluster to another (burrow cluster defined by Johnson and Crossman 1991) (Figure 4.11). Her body mass following dispersal had increased from 29.5 kg in June 1995 to 31.1 kg in November 1995, and total body fat measured by BIA from 4.9% to 7.7% of total body mass respectively. Although this dispersal event is relatively small in comparison to other dispersals described by Johnson and Crossman (1991) and the changes in body mass from June 1995 to November 1995 are negligible for a large herbivore, it does however support the hypothesis of female dispersal leaving the offspring to inherit the natal burrow.

Figure 4.11. Home range and dispersal of Female #82. The core home range (described in Chapter 5 and by Johnson 1991a) was estimated by the harmonic mean transformation in July/August 1995. The dispersal of Female #82 from her core home range (as indicated by the closed oblong shape in the centre of the map) to a neighbouring burrow group (B24) was recorded in November 1995. The map represents the northern range of the habitat of the NHN wombat (the full range is shown in Figure 2.5). Fire breaks are represented by continuous lines (———); wombat burrow are indicated by (•); and the position of the two telemetry towers are shown by (⊙).



4.4.3 Body Composition and Body Condition Methodology

The development of new methods such as BIA and TOBEC for measuring the body composition of animals has led to a number of recent studies aimed at validating the procedures used to measure body composition in animals and this in turn has re-focussed attention on the precision and accuracy of different approaches. Standard chemical analysis remains the reference method although the division of the carcass into water, protein, fat and ash has not changed for at least 100 years. Ecologists remain principally focussed on crude fat and its value as an energy reserve with the assumption that this partitioning reflects the ability of the animal to survive in times of nutritional stress and also reflects the capacity of the animal to direct energy to reproduction. These methodologies should be validated in a range of species that have different morphologies and lifestyles because it is clear that assumptions derived from domestic species (e.g. constant hydration of LBM) can lead to large errors in wild species (Arnould 1995).

In the present study, there was, as expected, a strong negative linear relationship between total body water (TBW) and fat although indirect estimates of TBW led to a small but significant error. Dilution space measured by deuterium overestimated TBW by 5% in wombats; this is at the upper end of the range of errors found in other species (Sheng and Huggins 1979). This error is usually attributed to exchange of the deuterium with non-aqueous H pools such as those in protein and fat (Nagy and Costa 1980) and most probably reflects isotope injection errors and/or analytical errors in determining $^{2}H_2O$ rather than any physiological variations in the amount of non-aqueous H pools in different species (Arnould 1995). In contrast, water labelled with $H_2{}^{18}O$ overestimated TBW by some 2.3%. This was not anticipated since several studies have shown a close correspondence between $H_2{}^{18}O$ and TBW (Nagy and Costa 1980). Of more concern, there was no relationship between body fat and $H_2{}^{18}O$ dilution space. This may reflect the limited number of observations made with this isotope and the narrower range of those observations compared with data derived using deuterated water. Since it is unlikely that $H_2{}^{18}O$ was incorporated into body tissue (Nagy 1980), the source of this error is uncertain.

Despite these errors, deuterium dilution space provided better estimates of TBW and body fat than those derived by BIA. Previous studies of BIA in wild species have shown a close correspondence between these two measures but all were concerned with carnivores (seals and bears). The major errors in the BIA procedure involve the position of the animal (Farley and Robbins 1994), injuries to the animal, placement and extent of contact of the electrodes. By carefully controlling these factors, this study obtained precise estimates of resistance and reactance. However, the range of resistance/reactance values was low compared to other studies (Table 4.6) - even in species with similar concentrations of body fat to wombats and this contributed to the relatively large SE of the regression. This may be due to the large amount of water contained within the gut, relative to body mass, associated with herbivorous habits (Barboza 1989). This pool would offer little resistance to the passage of current and so reduce the absolute magnitude of values measured.

This gut water volume is included in the estimate of TBW and thus must be recognised in the BIA measurement but there is some doubt about the extent to which this occurred. Other studies have demonstrated that the resistance value obtained with BIA is sensitive to the degree of hydration of the subject. More specifically, Cha *et al.* (1995) found that BIA was poor in detecting fluid in the trunk of rats following intraperitoneal injections of saline and concluded that this was because the trunk region had a large cross-sectional area compared to other body compartments. The theory of bioelectrical impedance analysis assumes that the current that is applied passes through all parts of the animal from one electrode to the other. Therefore, this study expected that incorporating a measure of electrical volume (which incorporates a measurement of length) into a calibration equation, would have provided the best estimate of total body water and total body fat. This did not happen in this study and there was no correlation between composition and electrical volume. This suggests that animals, particularly large herbivores such as wombats, are far from being perfect conductors.

Given the large proportion of total body mass found in the trunk of herbivores like wombats, multiple linear regression techniques incorporating a

Table 4.6. Ranges of bioelectrical impedance and body fat in studies of wild animals, domestic animals and people.

Species	Resistance (ohms)	Reactance (ohms)	Body Fat (%)	Total Body Water (%)	Body Mass (kg)	Body Length (cm)
Harp Seals[1]	102 - 180	-5 - 13	-	31 - 58	181 ± 59	-
Bears[2]	59 - 166	5 - 18	9 - 39	41 - 68	-	-
Antarctic Fur Seals[3]	300 - 500	-	-	-	25 - 50	112 - 138
Southern Hairy-nosed Wombats[4]	44 - 68	7 - 15	2.6 - 19.3	61 - 75	14 - 28	85 - 110
Pigs[5]	31 - 50	3 - 7	22 - 42	-	88 - 113	90 ± 5
Humans[6] (Male)	310 - 576	46 - 84	4 - 33	-	56 - 124	165 - 201
Humans[6] (Female)	458 - 691	47 - 92	13 - 40	-	43 - 104	148 - 192

Source: 1, Gales et al. (1994); 2, Farley and Robbins (1994), Robbins pers. com.; 3, Arnould (1995); 4, present study; 5, Swantek et al. (1992); 6, Lukaski et al. (1986).

body mass variable did increase the predictive ability of BIA to predict both total body fat and total body water. This suggests that the morphometrics of the conductor, as well as electrical current components, are important factors when deriving predictions for body fat and body water from BIA measurements.

No body condition indices correlated closely with body fat. This suggests that the indices examined do not have the ability to predict body fat or protein in wombats. Simple repeat measurement of an individual's body mass may be more appropriate than standard morphometric indices as an index of body condition in wombats. Similarly, the lack of correlation of the condition score with body fat may be due to fat deposit sites not being detected by palpation. Intra-abdominal deposits of fat will not be differentiated by the condition score from an animal with a full gastro-intestinal tract. Subtle variations in body fat will not be detected with a crude score if the range of body fat in the population is small (2.6% to 10.7% body fat in males, n = 7). However, the condition score may be most useful in describing animals of very poor or emaciated condition when physical characteristics may provide a better indication of wellbeing than body fat.

The choice of method to estimate body composition in the field must involve trade-offs between speed, accuracy and cost. BIA is fast, relatively cheap once the apparatus has been obtained and can offer good predictions of body fat and body water using multiple linear regression techniques. However, many questions still remain unresolved regarding the effects on the measurement of the subject, the nutritional status of the animal and a better appreciation of the factors affecting conduction of current through the animal. BIA has proved more successful in studies of wild species that contain substantially more fat than do wombats, such as bears (Farley and Robbins 1994) and seals (Gales *et al.* 1994; Arnould 1995). Provided that the time that the animal is under anaesthesia and the cost of stable isotope assays can be controlled it may be preferable to use isotopes over BIA in body composition studies. Where animal handling time is a concern, such as in studies on endangered species, BIA may be a more practical alternative.

This study has shown that body composition can be estimated accurately in the field by isotope dilution and to a similar degree of accuracy although

considerably more rapidly by BIA. The method of choice must depend on the circumstances of the particular study and the precise question being asked. However the quantification of crude fat should not be the end goal of studies of body composition. This is because the links between total body fat and animal wellbeing are poorly known. For example, we have little idea what proportion of the total body fat can be mobilised and under what conditions these lipid reserves are used. Pond (1978) has consistently pointed out that a simple knowledge of body fat may in fact be misleading if there are factors which prevent the animal making full use of the stores.

In order to address finer scale questions about the potential effect of TBF on differing levels of reproductive outputs in individual wild animals, there is a need for better a understanding of these links. Total body composition is an initial tool but more useful information may be available with relatively little effort whether it is using isotopes to study an individual's body composition or BIA to examine broad scale population trends.

CHAPTER 5

THE ACTIVITY AND BEHAVIOUR OF THE NORTHERN HAIRY-NOSED WOMBAT

5.1 INTRODUCTION

5.1.1 Herbivore Ranging Behaviour

The behavioural response of a herbivore to changes in its food resource is an important component of any study of its feeding ecology. Both optimal foraging theory and the mechanistic approach to foraging predict that the herbivore should forage so as to optimise its energy intake through minimal energy expenditure (Schoener 1971 & 1979, Pyke *et al.* 1977). However, these are not the only factors affecting the foraging behaviour of herbivores. Changes in the abundance, distribution and quality of plants invoke a behavioural response in herbivores to their forage (Spalinger and Hobbs 1992, Shipley and Spalinger 1992). This functional response affects specific aspects of herbivore behaviour including eating rate (Chacon and Stobbs 1976, Trudell and White 1981, Demment and Greenwood 1988, Spalinger and Hobbs 1992) and foraging and ranging behaviour (Trudell and White 1981, McNaughton and Banyikwa 1992, Murray 1992).

Norberg (1977) suggests that a herbivore should respond to variability in its forage environment by adjusting its rate of intake and time spent foraging. This response is, in part, intrinsically linked to the digestive physiology of the herbivore. As the digestibility of the forage decreases, colonic fermenters should increase their dry matter intake compared to foregut fermenters to extract the required amount of nutrients, if the necessary quantity of forage required to support increased rates of intake are available (Hume 1978). If the quantity of forage is abundant, colonic fermenters can maintain comparatively higher rates of intake and passage than foregut fermenters. Therefore, the ability of a colonic fermenter to extract nutrients

may be greater than that of foregut fermenters when forage is abundant and of low quality (Bunnell and Gillingham 1985, Duncan *et al.* 1990, Illius and Gordon 1993). However this must come at a cost. Time devoted to foraging by a colonic fermenter should be linked to the nutritional quality of the forage but only if the availability of forage is continuously high. Seasonal trends in forage quality should therefore translate into seasonal variations in the time devoted to foraging to meet intake requirements. Long foraging bouts should therefore equate to the time when the quality of the forage is lowest.

Chapter 2 has clearly demonstrated that the forage available to the NHN wombat is variable in quality and quantity. The quantity of forage changes seasonally but the standing biomass of plants is sufficiently high throughout the year that it may be considered abundant (e.g. compared to Evans 1992). However, the quality of the forage through the seasons varies significantly, particularly in digestibility and concentration of total N. It follows that the foraging activity of the NHN wombat may be seasonal in order to meet its forage intake requirements. Therefore, it is the prime objective of this chapter to identify responses by the NHN wombat to temporal fluctuations in the condition of available forage.

5.1.2 Indirect Methods of Examining Behaviour and Activity

Directly assessing the activity and behaviour of wombats is very difficult because of their wariness and nocturnal habits. For the NHN wombat the height of grass in its habitat increases the difficulty of behavioural observations. Rather than use direct techniques for observing the behaviour of the NHN wombat, indirect methods are preferred (e.g. Johnson 1991a).

Radio-tracking is commonly used to indirectly obtain information on the behaviour and ecology of free-ranging vertebrates (Banks *et al.* 1975, Hines and Zwickel 1985, White and Garrott 1990). Using the signal from a radio transmitter, locational data can be obtained through triangulation. The locational data can then be used to describe the home-range of the animal (Dixon and Chapman 1980).

Johnson (1991a) used radio telemetry to examine the activity patterns, ranging behaviour, habitat requirements, social organisation and dispersal of the

NHN wombat. He found that the ranging behaviour and activity of the NHN wombat was very conservative. This conservatism is best reflected in the hours spent above-ground per day. Johnson (1991a) found that individual NHN wombats spent between 2 and 6 hours above-ground per day. This is very conservative compared to the Eastern Grey kangaroo, *Macropus giganteus*, a herbivore of similar body size, that spends between 10.5 and 18.5 hours per day feeding (Clarke *et al.* 1989, Johnson 1991a). However, the time that the NHN wombats spent above-ground does not necessarily represent time spent feeding. Gaughwin (1981) describes the emergence pattern of southern hairy-nosed (SHN) wombats, suggesting that considerable time is spent around the burrow complex before they move off to graze. Observations of SHN wombats reveal that some time above-ground is spent inactive sitting on the edge of burrow complexes (*pers. obs.*). Common wombats spend between 3 and 8.2 hours above-ground per day (McIlroy 1973) but like SHN wombats they can spend a significant proportion of time (over an hour) inactive on the edge of the burrow before moving off to feed (Triggs 1988). The feeding activity of the common wombat varies greatly between individuals, particularly in preferred feeding areas, grazing intensity and ranging areas amongst individuals (McIlroy 1973). Despite this work, the proportion of time spent above-ground devoted to ranging and feeding remains unknown for the NHN wombat.

Recently, advances in telemetry studies on free-ranging animals have seen the development of sophisticated data collection and storage devices to indirectly measure animal behaviour and activity (e.g. Langbein *et al.* 1996). These devices have the capacity to record and store large quantities of information in a relatively small package. Coupled with radio telemetry, these devices can be used to remotely measure activity and behaviour of a large free-ranging herbivore, including its foraging behaviour. This chapter therefore attempts to interpret and describe the activity of the NHN wombat and estimate the time devoted to ranging and feeding. To comprehensively examine the activity and behaviour of the NHN wombat, methods of Johnson (1991a, b) are followed and the results compared.

5.1.3 The Short-term Effect of Radio Packages

Data collected from radio telemetry and activity loggers is valid only if the package does not itself modify the behaviour of subjects. In most studies this possibility is not investigated, but there are reports of radio-tracked animals behaving abnormally and suffering increased mortality as a direct result of the attachment of the radio package (e.g. Greenwood and Sargeant 1973, Perry 1981). Other studies report that individuals fitted with a radio package exhibit no differences in behaviour or survival compared to unmarked control individuals (e.g. Erikstad 1979, Johnson and Berner 1980, Garrott *et al.* 1985, Johannesen *et al.* 1997).

The attachment of a radio package involves capture and handling of the study animal. The process of capture may itself cause stress to the animal (Hoyle *et al.* 1995). With the attachment of the radio package, there may be an additional period of stress associated with the adjustment of the animal to the radio package (Greenwood and Sargeant 1973, White and Garrott 1990). If the package is too heavy, inhibits movement, or causes physical trauma or pain, this stress may persist for long periods with serious consequences for the health of the subject (and the objectives of the study). As part of this chapter it is important to describe the impact of radio-tagging on levels of activity in the NHN wombat by using activity loggers. In this way the existence and duration of a period of abnormal behaviour associated with recovery from capture or adjustment to the radio package can be examined.

5.2 METHODS

5.2.1 Study Animals

Northern hairy-nosed wombats were captured from the north-eastern distribution of burrows (Figure 2.5) as described in Chapter 4. Eight animals (four adult females and four adult males) were fitted with radio packages between June 1995 and July 1996. The radio package (designed by Murray Evans, University of New England, Armidale and built by Titley Electronics, Ballina, NSW) consisted of a collar constructed of plastic-coated cloth webbing, with an aluminium cylinder housing an activity data logger (described below) mounted on the collar, and a

radio transmitter attached with epoxy resin to the outside of the aluminium cylinder. The aerial of the radio transmitter was sewn in the cloth webbing, so that it exited the webbing 180° from the cylinder when fitted to the NHN wombat. The collar was scalloped around the neck to prevent injuries to the NHN wombat from the collar. The collar was closed with brass plates and bolts, and positioned so that the aluminium cylinder rested under the animal's chin, to minimise its impact on posture and locomotion. The health and wellbeing of the NHN wombats was assessed every three months by re-trapping, and the collar removed if there was any sign of discomfort or injury to the animal. Radio packages were attached to the NHN wombats for time periods ranging from 3 months to 15 months.

5.2.2 Radio Telemetry

Activity and movement of NHN wombats was recorded in two sessions of radio tracking (7 nights in duration per session), representing two extremes in rainfall and pasture condition. The July/August 1995 session was representative of the dry season with low ambient temperatures and rainfall (preceding 3 month total = 33 mm), while February 1996 represented the wet season with high ambient temperatures and comparatively higher rainfall (preceding 3 month total = 105 mm). Methods used in the radio telemetry study followed the procedures and protocols of White and Garrott (1990) and Johnson (1991b).

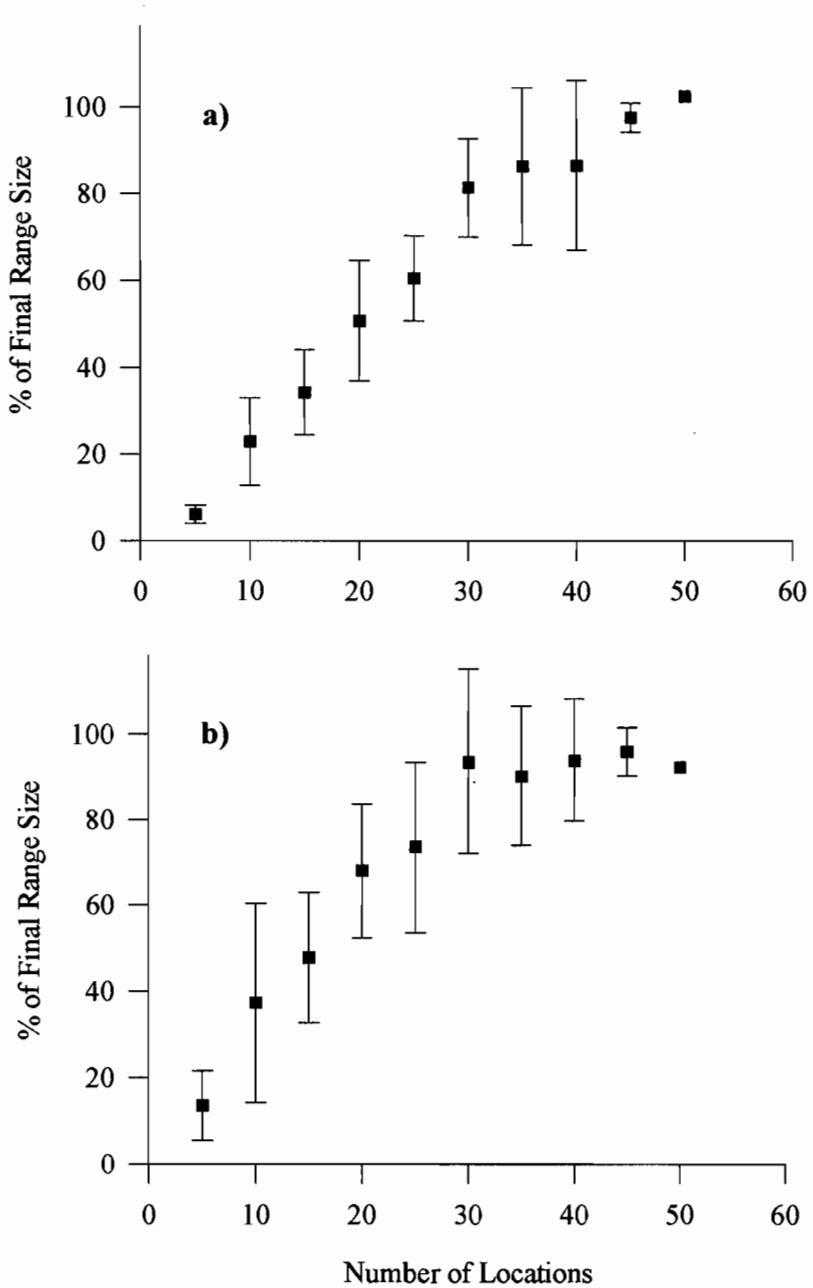
Movements of the NHN wombat were measured from two fixed receiving stations located on the periphery of NHN wombat habitat, in the north-eastern range of the burrows of the NHN wombat (Figs. 4.11 & 5.5 to 5.7). Each station consisted of a four element directional Yagi antenna mounted on a four metre aluminium tower over a compass rose. Towers were constructed and assembled to the specifications of the supplier (Sirtrack, New Zealand). The bearings of the NHN wombat from the two towers were determined using radio receivers with confirmation of direction using a Null-Peak system. A signal from a NHN wombat could only be detected by two towers if the NHN wombat was above-ground, or by one tower if the NHN wombat was in the vicinity of the burrow entrance or beyond the receiving range of the second tower. Locational bearings of the position of each NHN wombat were recorded every half an hour simultaneously

from each tower. Thus, in a nightly session of radio-tracking from 1800h to 0800h, the maximum number of independent position recordings of a wombat would be 29 per night. However, the actual number of position recordings for an individual in a single night ranged from 0 to 17 and a total of up to 65 position recordings per individual for a session of radio tracking.

Locational bearings were converted to grid points, referred to as locations, by triangulation using Wildtrak software (Version 1.11; I. Todd, University of Oxford, UK). During this procedure, potential outliers were identified and removed. Home-range and range-use contours were calculated using the harmonic mean (HM) transformation of Dixon and Chapman (1980) and the minimum convex polygon (MCP) estimate of Hartigan (1987). Home-range areas were calculated using Ranges IV software (R. Kenward, Institute of Terrestrial Ecology, Wareham, UK) and Wildtrak for the HM transformation and MCP estimate, respectively. Analysis followed the assumption of Johnson (1991b) that 30 minutes was a reasonable separation between locational fixes and that each NHN wombat location was independent of every other location. Although this assumption cannot be tested without direct observation of the NHN wombat, direct observations of both the SHN wombat and common wombat indicate that this assumption is generally valid (McIlroy 1973, Gaughwin 1981). One-way ANOVA was used to examine any differences between the HM and MCP methods for estimating home range areas for each radio-tagged wombat.

The estimate of home-range was dependent on the number of locations available for analysis. Johnson (1991b) found that the number of locations required to estimate the home-range (the 70% HM activity contour) of the NHN wombat within 10% of its true value was forty. In this study, forty-five locations provide the best estimate of the 100% HM calculation but only thirty are required for the 70% HM for the NHN wombat (Figure 5.1). For comparisons between sexes and seasons, home-range estimates were standardised to 30 locations selected at random from the total number of location recordings per individual for a session of radio tracking (Johnson 1991b). One-way ANOVA was used to examine

Figure 5.1. The effect of the number of locations on the estimated size of the home range of the NHN wombat for:
a) 100% harmonic mean activity contour (mean with one S.D.);
b) 70% harmonic mean activity contour (mean with one S.D.).
Home-range size at each number of locations expressed as a percentage of the home-range size calculated from 50 sample locations, with smaller number of locations calculated from random samples from the total number of locations following Johnson (1991b).



differences in the home range estimates between sexes and again to examine difference in the home range estimates between seasons.

Assessment of the above-ground activity of radio-collared NHN wombats followed the method of Johnson (1991b). Briefly, an index of activity (between 1.0 and zero) represented the proportion of radio-collared NHN wombats that were above-ground in a two hour period, determined by signals received by the two receiving stations. An index of one indicated that all radio-collared NHN wombats were above-ground, and zero that none were above-ground. The relationship between ambient temperature (measured at one of the radio tracking towers) and above-ground activity was examined using least squares linear regression.

Estimates of the home range of the NHN wombat were compared to home range estimates for the Macropodidae. Some adult female members of the Macropodidae are of similar body mass to adult NHN wombats and are also grazers (e.g. ranges of female body mass are: *Macropus fuliginosus* 4.5 to 27.5 kg; *M. giganteus* 3.5 to 32 kg; *M. robustus* 6.25 to 25 kg; and *M. rufus* 17 to 35 kg; Strahan 1995). The relationship between home range size and body mass for the Macropodidae was determined by least-squares linear regression using values from Norbury *et al.* (1989).

5.2.3 Data Loggers: Specification, Function, Calibration and Field Assessment

Specification and Function

Data loggers used were WB04 series Archival Tags, designed and manufactured by Zelcon Technic Pty. Ltd. (Glenorchy, Tasmania). The three channel data loggers were custom-built to record activity (stored potential energy), light (sensitivity scale of 254 counts) and temperature (range from -10°C to 35°C, with a resolution of 0.1°C), with an in-built real time clock. Data collection from the three channels occurred every five minutes. The storage capacity of the logger was 1 Mb allowing continuous data collection using a 5 minute, three channel schedule for three years. The lithium battery power source of the logger allowed data collection for six years, with the storage of data independent of the battery life. The whole logger package was assembled in heat-cured epoxy, weighing

approximately 25 g, measuring 55 x 24 x 12 mm, with a 5 cm external probe containing the light sensor and temperature sensor.

Data was collected in ASCII format, with a time stamp accompanying each five minute record. Transfer of data from the logger to a personal computer was via a purpose-built adaptor using Windows 3.1 Terminal program (Microsoft Corporation). Data was then deciphered from hexadecimal code into a decimal format by Murray Evans (University of New England) using custom-written software.

Calibration

Calibration of the temperature records was achieved by placing loggers in constant temperature rooms (3°C, 18 C, 25°C, and 28°C) for 24 hours. Hexadecimal measurements of temperature were then converted to a decimal format. Least squares linear regression was used to determine the relationship between actual temperature and the data logger record (Table 5.1). Since each channel of the data logger records data in an identical manner to the temperature measurements, the regression equation for the temperature probe was used to calibrate each of the three channels and decipher the logger data.

Table 5.1. Least-squares linear regression equations of data loggers calibrated via the temperature probe. All regression equations are statistically significant: $p < 0.001$.

Logger Identification	Regression Equation	r^2	S.E.
F82	$y = 0.34x - 31.53$	0.99	0.70
M41	$y = 0.33x - 30.81$	0.99	0.87
M25	$y = 0.34x - 31.41$	0.99	0.93
W501	$y = 0.36x - 36.17$	0.99	0.40
W502	$y = 0.36x - 35.74$	0.99	0.46
Mean Equation	$y = 0.33x - 31.26$	0.99	0.83

Field Assessment

The field sensitivity of the data loggers was examined using three data loggers under controlled conditions. Two data loggers were placed in a Stevenson Screen located in the north-eastern range of NHN wombat habitat. To differentiate between changes in temperature and light as a result of aspect, one logger faced north-east and the other south-west. The third logger was fixed in position 1.5 metres inside the entrance of the tunnel of a NHN wombat burrow. Light and temperature records were collected for 3 months, but high light intensity and high daytime temperatures damaged the probes, and consequently the sensitivity of the two loggers in the Stevenson Screen.

5.2.4 Data Loggers - Measurements of the NHN Wombat

Of the eight NHN wombats fitted with collars only five data loggers functioned successfully between June 1995 and July 1996. Further, of the five data loggers that successfully functioned, only four operated continuously for more than 60 days. However, these five data loggers were used to generate all of the data reported in this chapter. Further, the large quantity of data produced by the data loggers required sub-sampling for practical analysis of the data. Details of the sub-sampling of the data set are provided with each analyses.

5.2.4.1 Short-term Effect of the Radio Transmitter/Data Logger Package

Activity was recorded by a sensor positioned under the wombat's chin. This sensor recorded movement of the wombat by detecting and counting the movement of a small ball within a cage. The cumulative count of movements recorded by the activity sensor over a 5-minute period provided a measure of the presence and level of activity. Activity data were pooled for twenty-four hour periods: e.g. in one 24 hour period, 288 individual activity records are produced by the data logger; these were summed so there was one data point for each 24 hour period. The level of activity in each 24 hour period was expressed as a percentage of the level of activity recorded in the first 24 hour period following initial capture and fitting of the radio package (day one). Trends in the daily activity records with time demonstrate how the NHN wombat adjusts to the radio package.

5.2.4.2 Activity Trends of the NHN Wombat

The vast quantity of data collected by the data loggers allowed trends in the activity of the NHN wombat to be described as means of several individuals (up to five) or at the level of the individual NHN wombat. Data was collected from July 1995 to July 1996, although data from October 1995 to January 1996 were incomplete.

Mean Trends of Activity

The measure of mean trends in activity illustrated temporal variation in the daily activity of the NHN wombat. Each month of data was treated as a single group of data. From within each month, a continuous ten-day block of data was taken as a sub-sample (ten individual twenty-four hour periods). The representative sub-sample was further divided, so that 288 records of activity per 24 hour period were reduced to 24 records of activity (i.e. one value of activity per hour). Data were then averaged to generate single periods of 24 hours of activity of a mean NHN wombat that were representative of each of the ten-day periods of each month sampled.

Individual Trends of Activity

Of the five individual NHN wombats, 579 twenty-four hour periods of data were collected (Female #61: 94 days; Female #152: 31 days; Male #25: 175 days; Male #32: 106 days; Male #41: 173 days). Each of these 24h periods has 288 records of activity (i.e. a total of 166, 752 records of activity). Because of the large data set, representative twenty-four hour periods were selected at random for each individual (with the exception of Female #152 because of logger failure). Again, each month was considered a separate data set. A twenty-four hour period was selected at random and this day was selected for each of the months where data were available (e.g. the 17th day of each month was selected as the representative twenty-four hour period from February to July 1996 for Male # 25). Rather than pool data into hour intervals for each twenty-four hour sampling period (i.e. 24 records of activity per twenty-four hour sampling period), data were pooled into fifteen minute intervals (i.e. 96 records of activity per twenty-four hour sampling period). Data were then graphically displayed for each month sampled,

demonstrating temporal variation in activity at a finer resolution than the mean trends.

Bouts of Activity by the NHN Wombat

Bouts of activity were defined as any recording of activity by the data logger. The length and duration of each bout of activity was examined for four of the radio-tagged NHN wombats. As for above, each month was treated as an individual data set. Within each month a continuous block of five days was selected at random. Each record of activity (representing a sampling period of five minutes in duration) was either a positive whole number (active) or a zero (inactive). The duration of the bout of activity was determined by the number of continuous active records separated by zeros. The interval between bouts of activity was determined by the number of continuous zeros (records of inactivity) between each bout of activity. For example an activity record such as 0,0,1,20,6,0,1,0 represents two bouts of activity, the first being of 15 minutes duration and the second being of five minutes duration separated by an interval of five minutes. The number of bouts, the length of each bout and the interval between each bout of activity were calculated for each 24 hour period in the block of five days. A mean and standard deviation were calculated for each block of five days (or sampling month) and the range of values recorded for the length of the bout and the interval between bouts of activity.

5.2.4.3 Bouts of Activity of the NHN Wombat Occurring Above the Ground

Bouts of activity by the NHN wombat occur both above-ground and within the burrow. While underground activity is important in understanding the behaviour of the NHN wombat (section 5.4.2.6), bouts of activity that occur above-ground are specifically relevant to its feeding ecology. Radio telemetry and measurements from the data loggers were used to distinguish between activity occurring above-ground and that occurring within the burrow.

Above-Ground Activity From Radio Telemetry

Northern Hairy-nosed wombats were considered above-ground when radio telemetry signals were received at both receiving towers as described above. The real time clock in the data loggers allowed comparison of known above-ground

records and activity measured by the data logger. To compensate for potential time discrepancies between the time the telemetry signal was recorded and the data logger clock, a window of fifteen minutes (3 activity records) was assumed to represent an above-ground active event. Above-ground activity measured by radio telemetry was restricted to the sampling period of radio telemetry (July/August 1995 and February 1996) and to individuals with functioning data loggers (N = 5).

Above-Ground Activity From Data Loggers

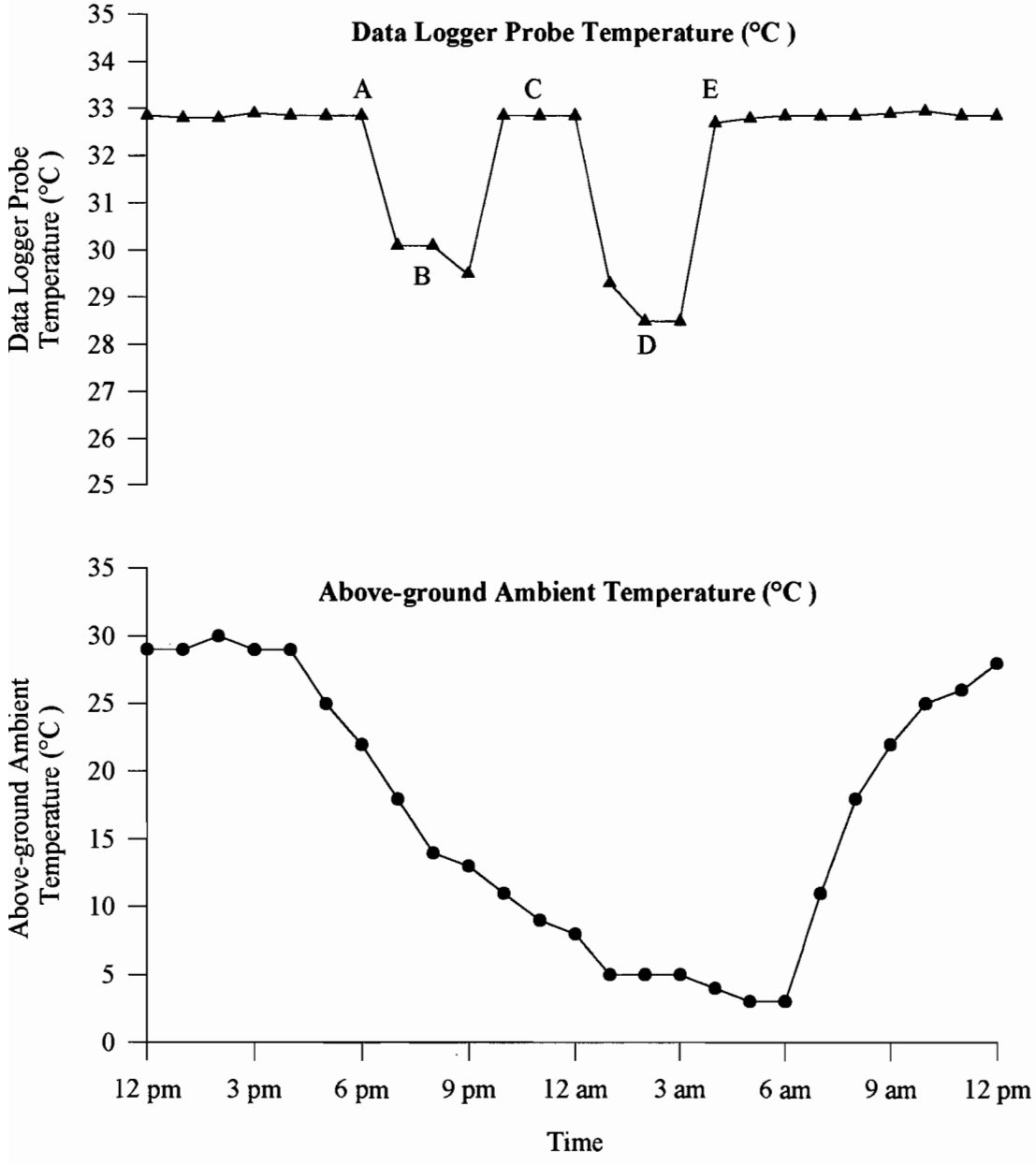
The temperature recordings from the thermocouple of the radio package were partly influenced by the body temperature of the NHN wombat because of the way the thermocouple was positioned. Consequently, the resolution of the temperature data was insufficient to differentiate between the relatively constant temperature of the burrow and above-ground ambient temperatures. However, during the months of the year when the ambient temperatures at night were low (May, June, July, August and September; Figure 2.1) the difference between the above-ground ambient temperature and the constant recordings of the temperature probe was sufficient to suggest that the NHN wombat was above-ground.

In the example shown in Figure 5.2, an above-ground activity profile can be suggested. For example, the NHN wombat may exit its burrow at about 7 pm, remaining above-ground until 10 pm before returning to its burrow. At 12 am, the NHN wombat exits its burrow and remains above-ground until 4 am, before returning to its burrow until the following night's activities. Although the above-ground activity of the NHN wombat was not confirmed by observation, the inference of above-ground activity by the temperature records seems reasonable.

The determination of activity by the temperature records were reduced to the winter months (May to September) and four wombats (Female #61, Male #25, Male #32 and Male #41). As with other analyses, the unit of a month was considered as a separate data set. A continuous period of nine days was then used as a representative sub-sample of each month. A NHN wombat was considered to be above-ground when the measurement of the temperature probe was a minimum of 2 °C below the temperature recorded by the probe at midday (when NHN wombats are definitely below ground). This allowed the mean number of hours

Figure 5.2. A graphical demonstration of variation in the temperature measurements of a radio-tagged NHN wombat coupled with variation in the above-ground ambient temperature for a hypothetical twenty-four hour period in the month of June.

Points on the temperature profile of the data logger represent:
A. Emergence of the NHN wombat;
B. Above-ground activities;
C. Retreat to a burrow;
D. Above-ground activities; and
E. Retreat to a burrow, staying in the burrow until A the following night.



spent above-ground to be calculated for each month. Activities undertaken by the NHN wombat while it was above-ground were then categorised as described below.

Quantification of Above-Ground Activity

The records of activity recorded by the data logger when an individual NHN wombat was above-ground varied in intensity (from 0 to 136 units of activity in a five minute period). Although the activity measurements recorded by the data loggers have not been validated with a behavioural study of captive common or SHN wombats, it can be assumed that variation in the value of the activity record is likely to be a true representation of differing types of activities of the NHN wombat. It is likely that low values of activity recorded by the data logger are representative of behaviours which conserve energy such as sitting and some grazing. Higher levels of activity are likely to be associated with more vigorous activities such as digging or scratching.

In the absence of a formal validation study, an arbitrary scale was devised to describe potential activity by the NHN wombat while it is above-ground. This scale utilised the variation in the value of the activity measurement in each five minute record.

Table 5.2. A subjective scale describing the potential categories of activity undertaken in a five minute period by a NHN wombat as recorded by the data loggers.

Score	Activity
0	Inactivity or Stationary Grazing
1-20	Foraging Activities
21-60	Other Activities (intensive foraging, scratching, social activities etc.)
>60	Intensive Activity (running, digging etc.)

5.2.4.4 Correlation of Activity with Environmental Variables

The influence of environmental parameters (rainfall, temperature, forage quality and quantity) on foraging activity is of key significance to the feeding ecology of the NHN wombat. However, at the time of writing, data on forage quality was not available to allow meaningful analysis of its effect on foraging activity by the NHN wombat. Instead, the analyses were restricted to correlation analysis of mean daily activity with rainfall and temperature, both minimum and maximum. The effect of time was examined by calculating the correlation coefficient for each of the environmental variables at time *t* and the rainfall at time *t-1* in a time-series.

5.2.4.5 Temperature Profiles from Measurements of the Data Loggers

Mean and individual trends in the temperature profiles, recorded by the temperature probe of the data logger attached to radio-tagged NHN wombats, were analysed in the same manner as activity (described in section 5.2.4.3).

5.2.4.6 Light Levels - Diurnal Activities of the NHN Wombat

The light sensor of the data logging device recorded light in an incremental scale from zero to 254. It was found that zero did not equate to absolute darkness, but a count of 22 corresponded to the absolute darkness of the burrow. Further, the light sensor was not sufficiently sensitive to detect variation between the absolute darkness of the burrow and starlight or moon-light, but was able to determine when the NHN wombat was in full daylight or within the region of the burrow where sunlight penetrated (approximately 3 metres). It was also able to determine when the wombat was above-ground immediately after sunset or immediately prior to sunrise when the environment was not in full light.

To determine the number of diurnal activities, data from four individuals were pooled (Male #32, Female #61, Male #25 and Male #41). Unlike the activity sensor, the light sensor was less likely to fail unless the probe was damaged by the NHN wombat. Therefore, the pooled data set comprised 15 continuous months.

Diurnal activities of the NHN wombat were categorised by the following scale:

- dawn activity: when light was recorded by the data logger before sunrise;
- dusk activity: when light was recorded by the data logger after sunset;
- daylight activity: when light levels recorded by the data logger exceeded a count of 185 (i.e. light levels 70% that of absolute darkness, where full sunlight is 100% above absolute darkness or a light count of 254);
- tunnel activity; when light levels recorded by the data logger had light counts ranging between 45 and 185 (i.e. light levels ranged between 10% and 70% above absolute darkness).

These categories were devised after the field assessment of the data loggers (section 5.2.3) and therefore provide a realistic estimate of diurnal activities.

Again each month was considered a separate data set. Monthly trends of diurnal activity were expressed as a percentage of the total of days within a month where diurnal activity occurred. The types of diurnal activities were then categorised using the scale described above. Monthly trends were assessed by one-way ANOVA.

Trends in the categories of 'daylight activity' and 'tunnel activity' were further examined to determine when diurnal activity occurred. All pooled data were divided into two-hour periods from 6 am to 6 pm, with the levels of diurnal activities expressed as a percentage ± one standard deviation of the number of days in the 15 month sampling period.

5.3 RESULTS

5.3.1 Radio Telemetry

5.3.1.1 Above-Ground Activity

There was no significant difference in the time spent above-ground between the sexes (one-way ANOVA $F_{1,2} = 2.63$, $p > 0.05$). However, there was a significant difference in the time spent above-ground between July/August and February (one-way ANOVA, $F_{1,7} = 11.3$, $p < 0.05$), with more time spent above-ground in February.

This study found that the relationship between ambient temperature and the proportion of NHN wombats that were above-ground was linear in February but not significant in July/August (Figure 5.3).

5.3.1.2 Home-ranges

Like Johnson (1991b), the home-range area of the northern hairy-nosed wombat grew steadily when 10% increments of the total set of locational records were added from the centre of the range to its periphery (Figure 5.4). Above 70% of the locational records, increases in home-range area are accelerated considerably. Consequently the 70% range area contour has been used to describe the core area of the NHN wombat.

The home-ranges of seven individuals were successfully measured by radio telemetry in July/August 1995 (Figure 5.5) and February 1996 (Figure 5.6). There was no difference between the size of the total home-range estimated by the HM transformation and that by MCP (one-way ANOVA, $F_{1,14} = 1.28$, $p > 0.05$), with the average size of the total home-range estimated to be 31.6 ha. Core home-ranges did not differ significantly between sexes (one-way ANOVA, $F_{1,6} = 0.24$, $p > 0.05$) but did differ between seasons (one-way ANOVA, $F_{1,6} = 6.20$, $p < 0.05$) with larger core home-ranges recorded in February 1996 (Table 5.3). The difference in core home-range is best demonstrated by the repeat measurements of Female #97 (Figure 5.7).

Comparison of the Home Range of NHN Wombat with the Macropodidae

The relationship between the home range size and body size using the macropod data of Norbury *et al.* (1989) is linear ($y = 27.3x - 115.6$; $r^2 = 0.74$; $F_{1,14} = 40.15$; S.E. = 0.15; N = 16; $p < 0.05$; Figure 5.8). Using this regression equation, the predicted size of the home-range for the mean body size of the NHN wombat is approximately 739 ha compared to actual values in Table 5.3.

5.3.2 Field Assessment of the Data Loggers

The temperature probe of the data loggers successfully recorded trends in daily temperatures and demonstrated their sensitivity in measuring ambient

Figure 5.3. Variation in the above-ground activity of the NHN wombat with nightly temperature during:
a) July/August 1995,
b) February 1996.
Index of activity represents the proportion of wombats above ground in a 2 hour period, following the method of Johnson (1991b).
No significant relationship (linear or quadratic) for July/August 1995.
Significant linear relationship ($p < 0.01$) in February 1996:
$y = 1.90 - 0.06x$; $r^2 = 0.73$; $F_{1,10} = 27.02$; S.E. = 0.15; N = 12.

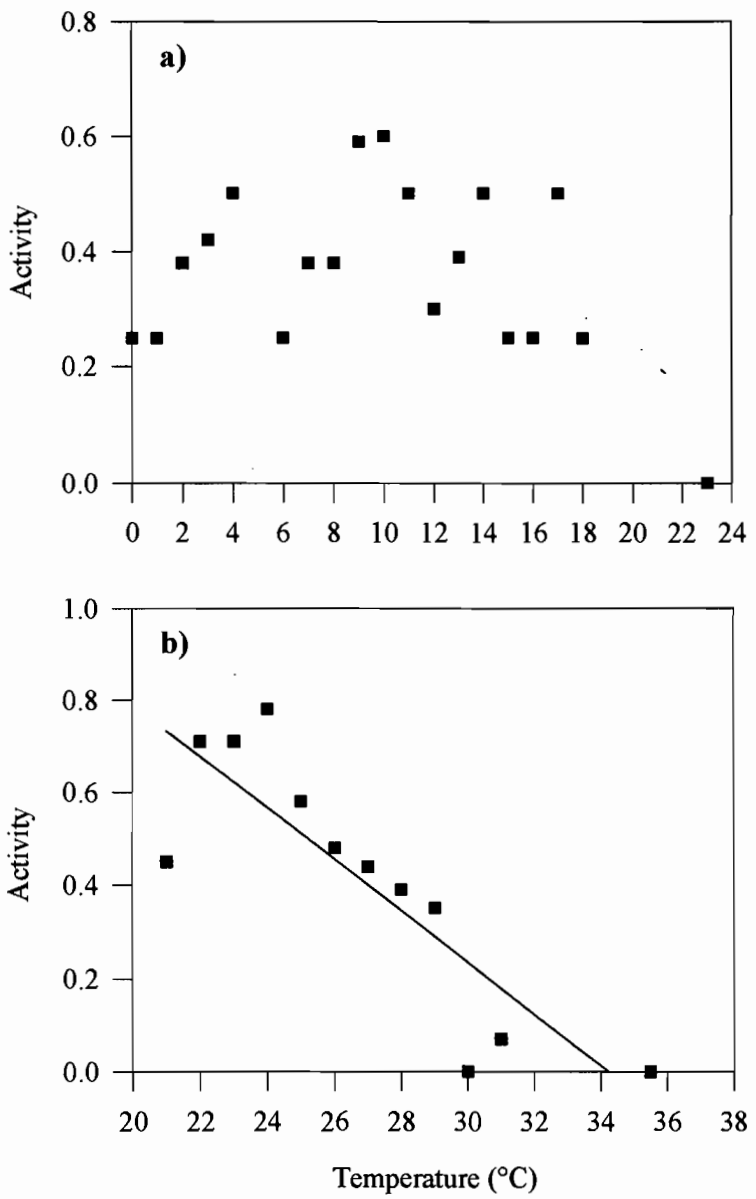


Figure 5.4. Changes in the area of the home-range of the NHN wombat, as successive increments, from the range centre to its periphery, of the total number of locations are added (Johnson 1991b). Home range measured by the harmonic mean transformation.

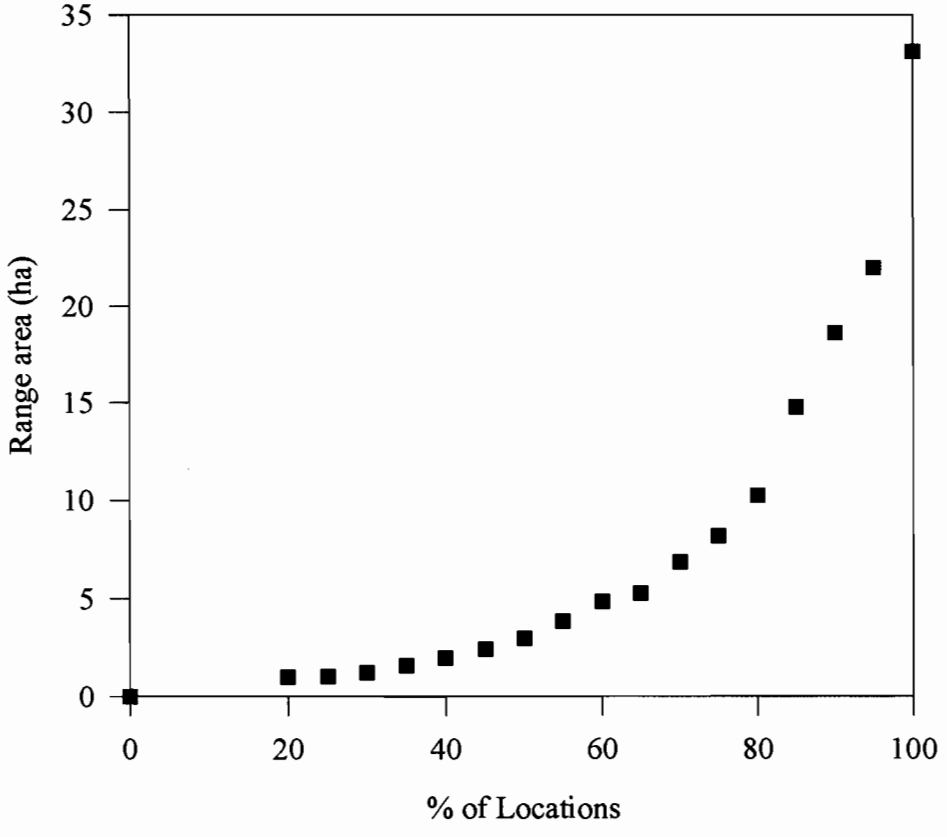


Figure 5.5. Distribution of the core home ranges (70 % of the harmonic mean transformation) of three northern hairy-nosed wombats in July/August 1995. Home range estimates are for Females #82 and #97 and Male #32. Home ranges are indicated by the closed "circles" in the centre of the map. The map represents the northern range of the habitat of the NHN wombat (the full range is shown in Figure 2.5). Fire breaks are represented by continuous lines (———); wombat burrow are indicated by (•); and the position of the two telemetry towers are shown by (⊙).

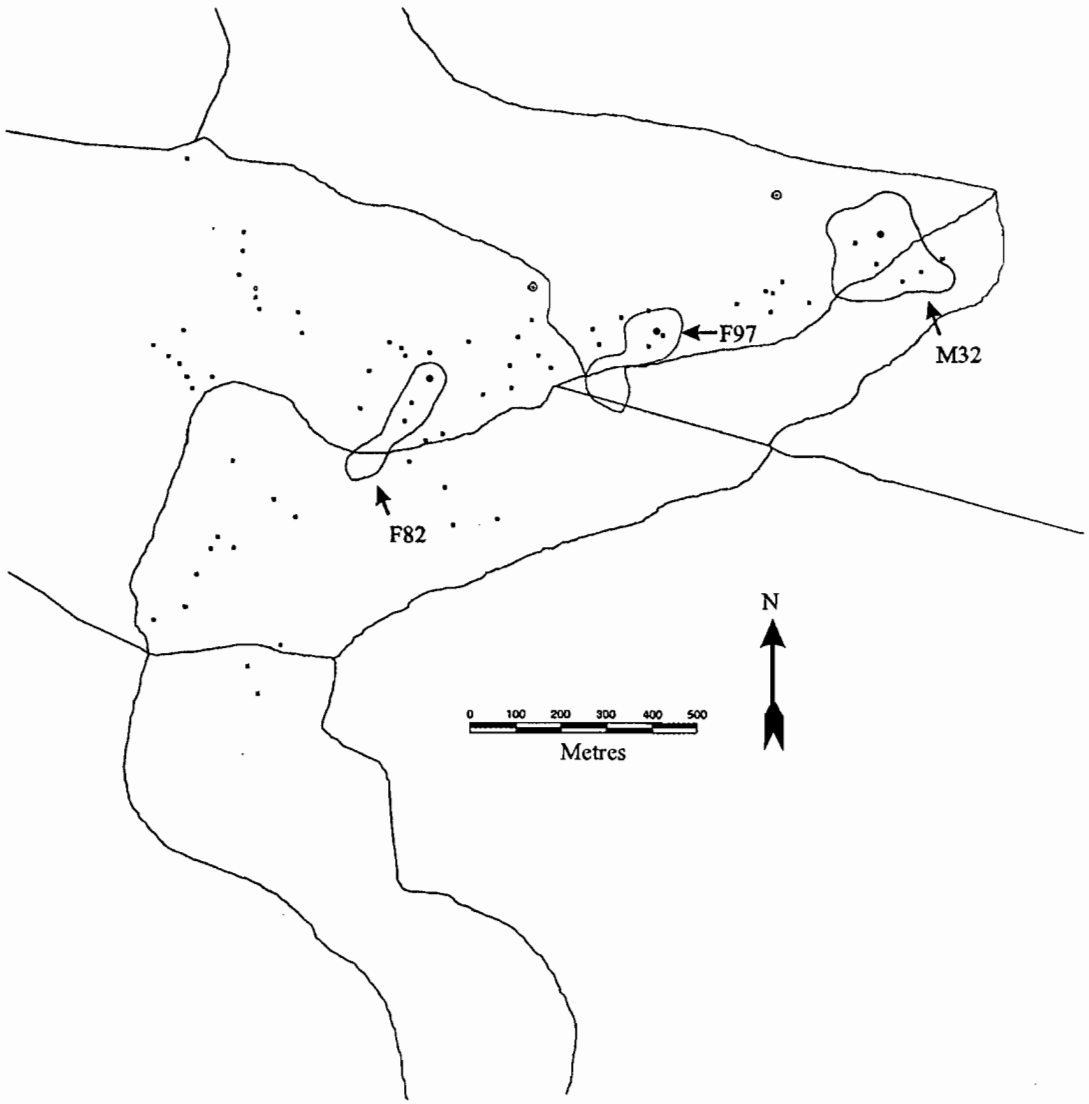


Figure 5.6. Distribution of the core home ranges (70 % of the harmonic mean transformation) of five northern hairy-nosed wombats in February 1996. Home range estimates are for Females #61, #152 and #97, and Males #25 and #41. Home ranges are indicated by the closed "circles" in the centre of the map. The map represents the northern range of the habitat of the NHN wombat (the full range is shown in Figure 2.5). Fire breaks are represented by continuous lines (———); wombat burrow are indicated by (•); and the position of the two telemetry towers are shown by (⊙).

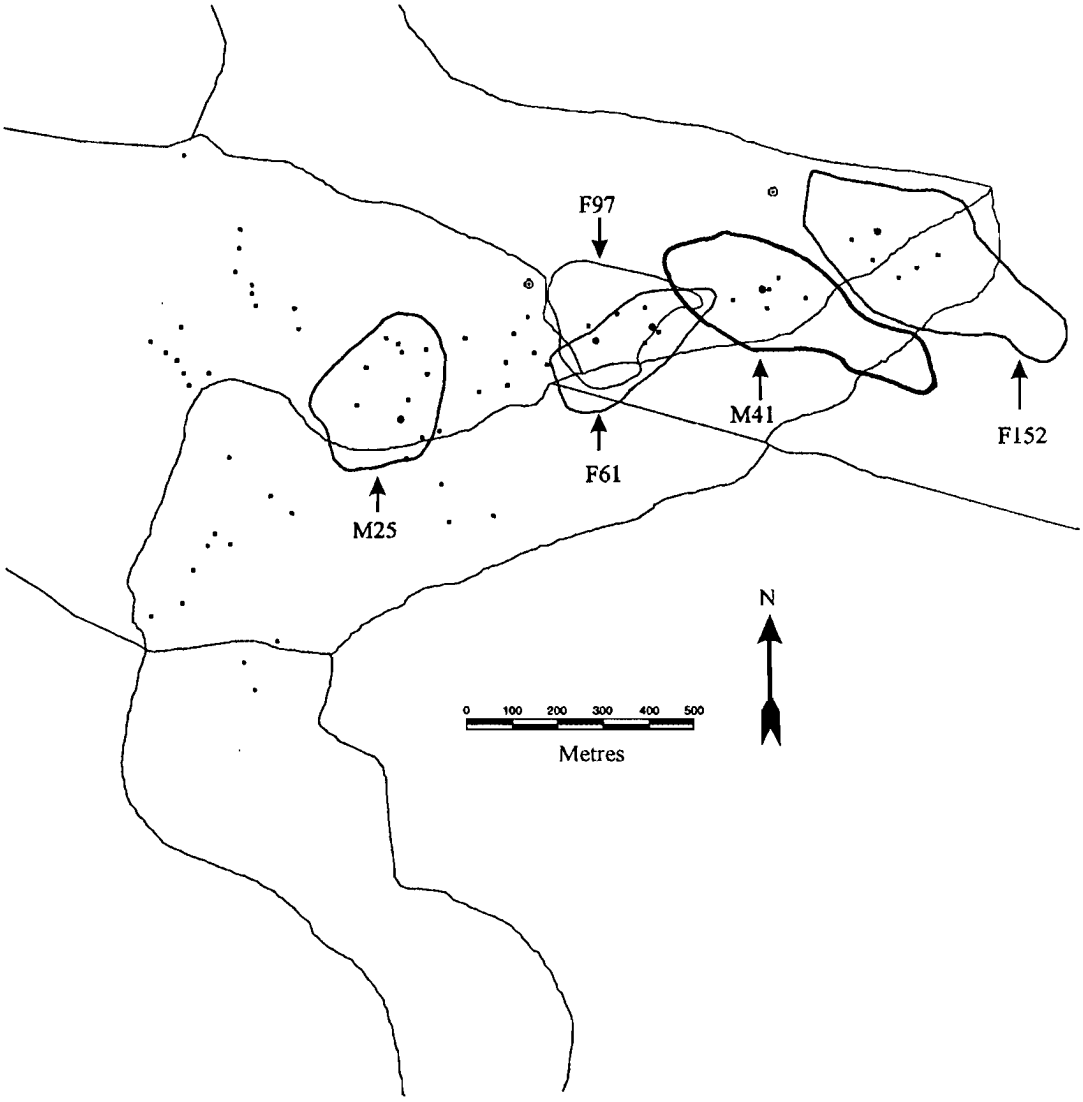


Figure 5.7. Seasonal variation in the core home range (70 % of the harmonic mean transformation) of Female #97, illustrating the difference in habitat utilisation between the dry season (July/August 1995) and the wet season (February 1996). Home ranges are indicated by the closed "circles" in the centre of the map. The map represents the northern range of the habitat of the NHN wombat (the full range is shown in Figure 2.5). Fire breaks are represented by continuous lines (———); wombat burrow are indicated by (•); and the position of the two telemetry towers are shown by (⊙).

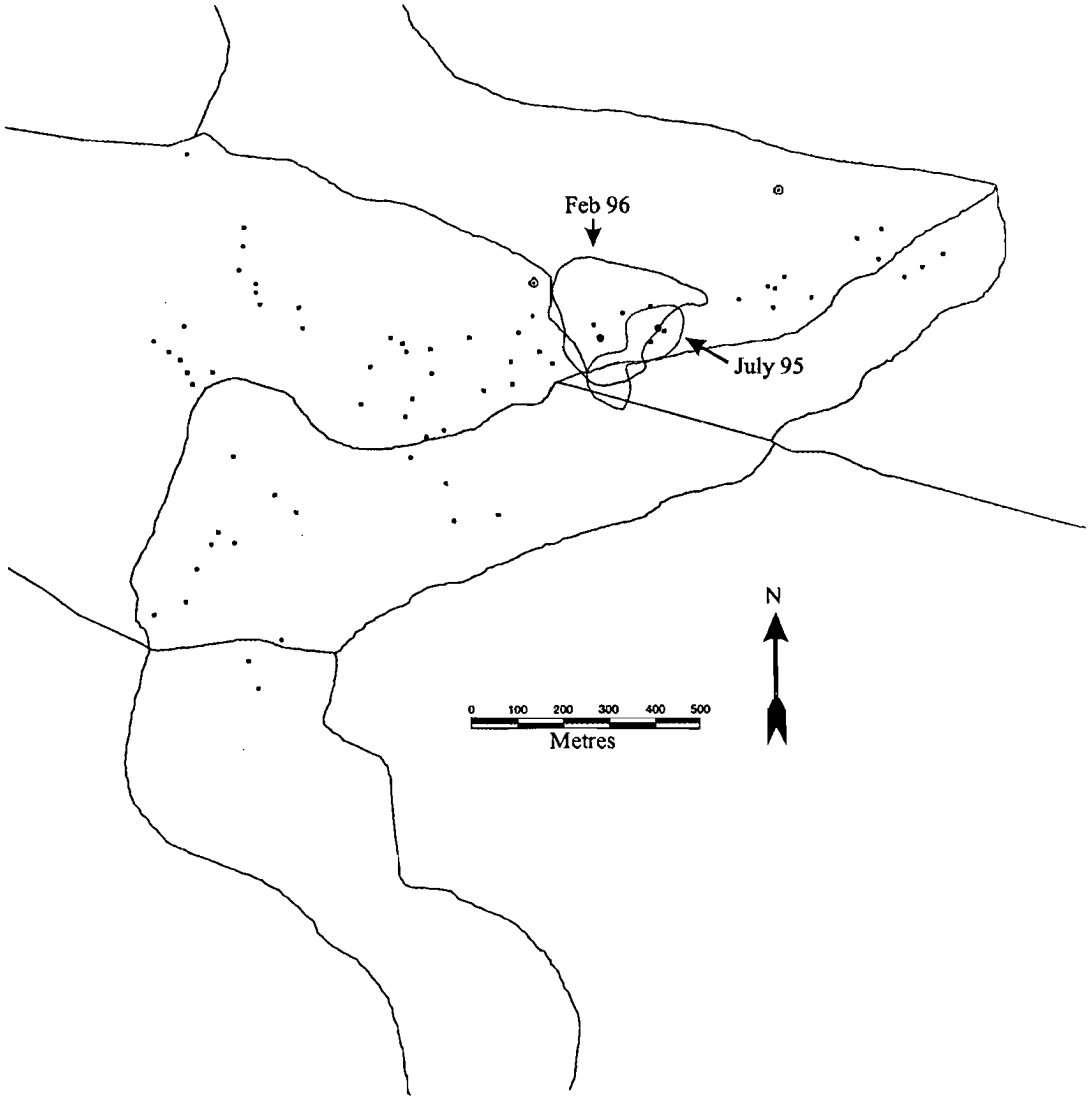


Figure 5.8. Estimates of the size of the home-range for some members of the Macropodidae and the NHN wombat in relation to body size (Macropodidae data from Norbury *et al.* 1989). The significant linear relationship for the size of the home range for the Macropodidae has the equation:
$y = 27.3x - 115.6$; $r^2 = 0.74$; $F_{1,14} = 40.15$; S.E. = 0.15; N = 16; $p < 0.05$
Species of macropod included in the analysis are: *Potorous tridactylus, P. longipes, Bettongia penicillata, B. gaimardi, Aepyprymnus rufescens, Lagorchestes conspicillatus, Thylogale billardierii, T. thetis, Macropus eugenii, M. parryi, M. rufogriseus, M. giganteus, M. fuliginousus, M. robustus, M. rufus, Setonix brachyurus, Wallabia bicolor.*

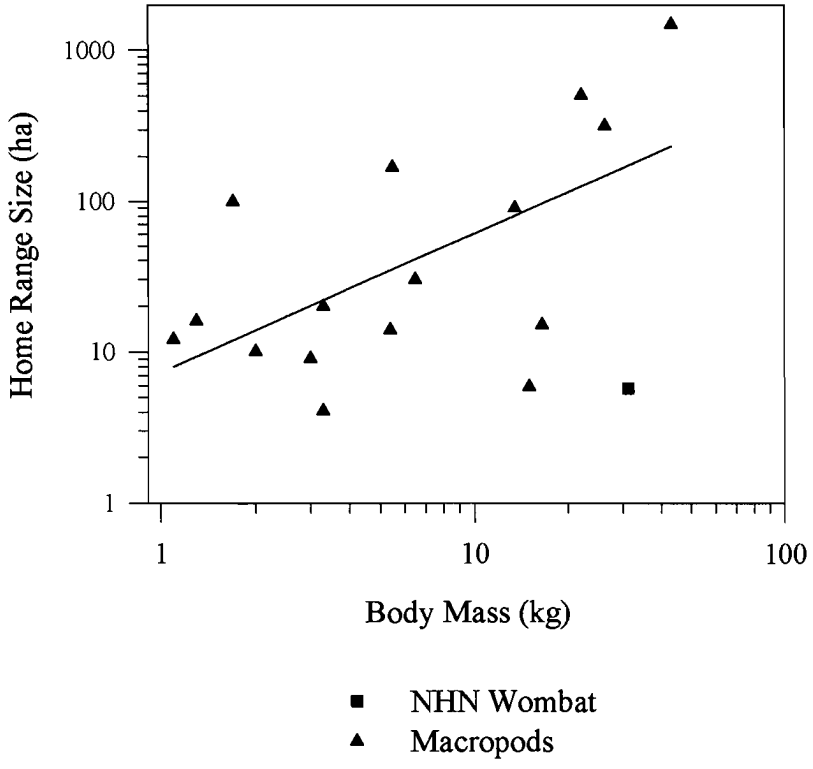


Table 5.3. Seasonal home-range estimates (hectares) of Northern Hairy-nosed Wombats using the harmonic mean (HM) transformation of Dixon and Chapman (1980) and minimum convex polygon (MCP) of Hartigan (1987). The number of NHN wombats sampled are in parentheses.

Home-Range Estimate	Sex	July/August 1995	February 1996
100% HM	Male	30.2 (n = 1)	37.4 (n = 2)
	Female	16.6 (n = 2)	42.2 (n = 3)
MCP	Male	23.5 (n = 1)	28.2 (n = 2)
	Female	8.9 (n = 2)	35.4 (n = 3)
Core Home Range (70% HM)	Male	5.9 (n = 1)	8.6 (n = 2)
	Female	2.4 (n = 2)	7.4 (n = 3)

temperature (Figure 5.9). The logger exposed to the most direct sunlight (north-east aspect) recorded higher daytime temperatures than the logger in shadier conditions (south-west aspect logger) and the logger located 1.5 metres in the tunnel of a NHN wombat burrow. Both loggers in the Stevenson Screen tracked identical patterns of temperature at night. Clear thermal benefits of a fossorial lifestyle are evident in the comparatively constant temperature levels recorded by the logger only a short distance within a NHN wombat burrow.

The light levels recorded by the two data loggers located in the Stevenson Screen quickly rise to their maximum value (a count of 254 or a value of 100%) following sunrise, while the light detected 1.5 metres in the burrow peaks at approximately 70% of the maximum value the sensor could record (Figure 5.10). Therefore, conclusions about daylight activities of NHN wombats using data measured by the light probe can confidently use the value of 70% as a reference for determining activity in daylight occurring above-ground.

Figure 5.9. Twenty-four hour trends in the temperature measured by the three calibration data loggers. Two of the calibration loggers were placed in a Stevenson Screen, one with a north-eastern aspect and the other with a south-western aspect. The third data logger was located 1.5 metres from the entrance of a NHN wombat burrow.

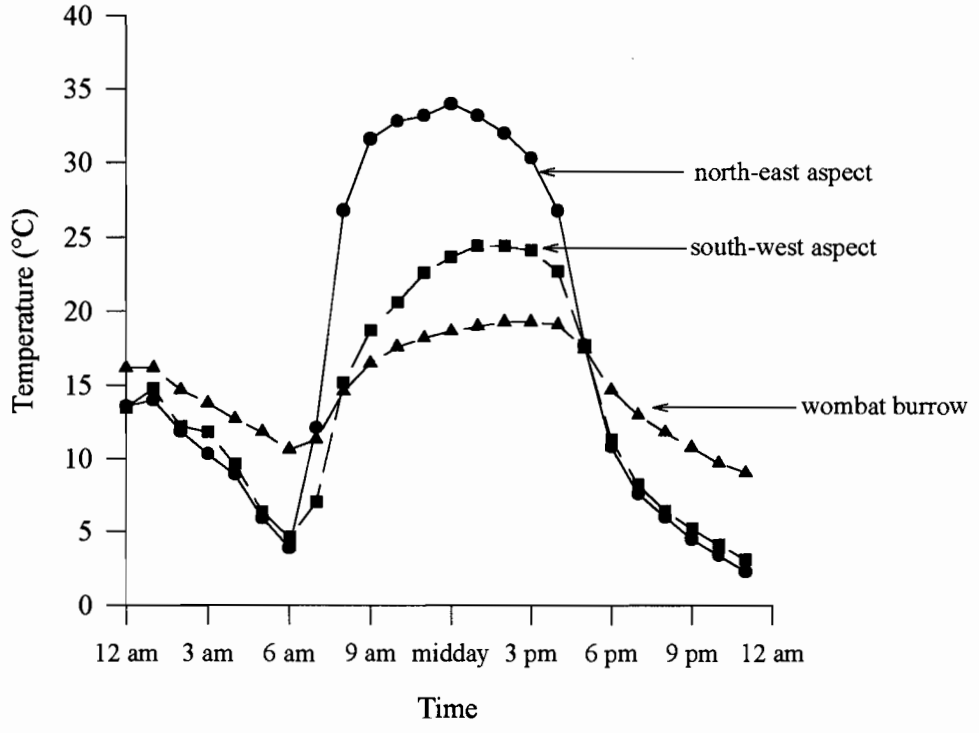
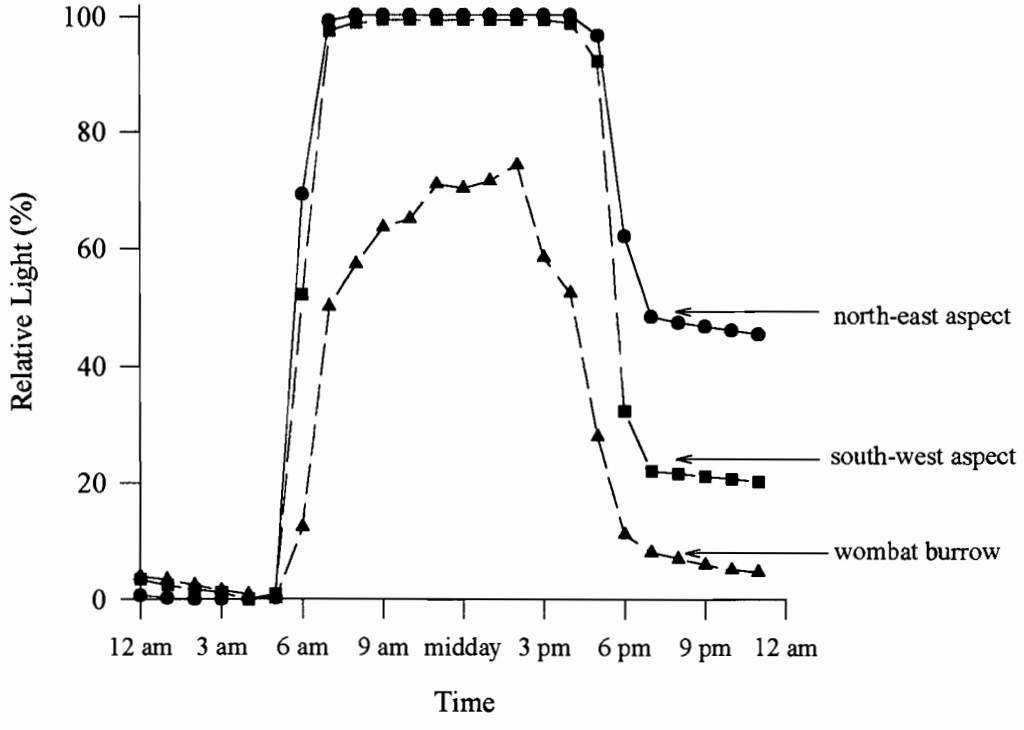


Figure 5.10. Twenty-four hour trends in the intensity of light measured by the three calibration data loggers. Two of the calibration loggers were placed in a Stevenson Screen, one with a north-eastern aspect and the other with a south-western aspect. The third data logger was located 1.5 metres from the entrance of a NHN wombat burrow.



5.3.3 Measurements of Activity

5.3.3.1 Short-term Effect of the Radio Package

All five NHN wombats exhibit a period of adjustment to the radio package before a stable level of activity was reached and maintained (Figure 5.11). This adjustment period was independent of seasonal conditions, with similar adjustment trends occurring in June (the dry season) and February (the wet season). Adjustment occurred between ten and eighteen days after capture and attachment. The stable activity range of a NHN wombat adjusted to the radio package is some 84 to 97% less than that of the initial activity level following collar attachment. The effect of trapping on NHN wombat activity was minimal (Figure 5.12). The greatest response in activity was exhibited by an individual fitted with a new collar of the same design in the second trapping session (Figure 5.12a). After an initial increase in activity, it rapidly resumed mean activity levels. No significant changes in daily activity levels were exhibited by an individual that was re-trapped but the same collar retained (Figure 5.12b), or by an individual that evaded capture (Figure 5.12c). The long term effect of the radio package on the NHN wombat was minimal, and all NHN wombats maintained body mass while wearing the collar (range: loss of 3.5% body mass to gain of 3.9% body mass, mean: gain of 0.3% body mass). When NHN wombats were re-trapped (Figure 5.12a&b), activity levels did not reach peak activity levels following initial capture and first attachment of the radio package (Figure 5.11).

5.3.3.2 Mean Trends in Activity

The mean trends in activity of the NHN wombat (Figure 5.13) differed significantly between months ($F_{10, 253} = 4.79$, $p < 0.05$). The apparently high levels of activity of the NHN wombats during February was likely to be an artefact of the adjustment period described above. However, the level of activity did vary significantly within a twenty-four hour period (one-way ANOVA, $F_{23, 240} = 6.18$, $p > 0.05$), corresponding with the NHN wombat's nocturnal lifestyle.

5.3.3.3 Individual Trends in Activity

The prime occurrence of bouts of activity by individual wombats were between 6 pm and 9 pm (Figure 5.14). Although activity occurred primarily at

Figure 5.11. Short-term effect of radio package on the activity of five individual wombats. Five wombats represent two females (a & b) and three males (c, d & e), captured in February (a, b, c & d) and June (e). Dashed line represents mean activity between 30 and 120 days following capture and attachment of radio package. The absence of a mean activity line for individual 'b' is the result of failure of the data logger.

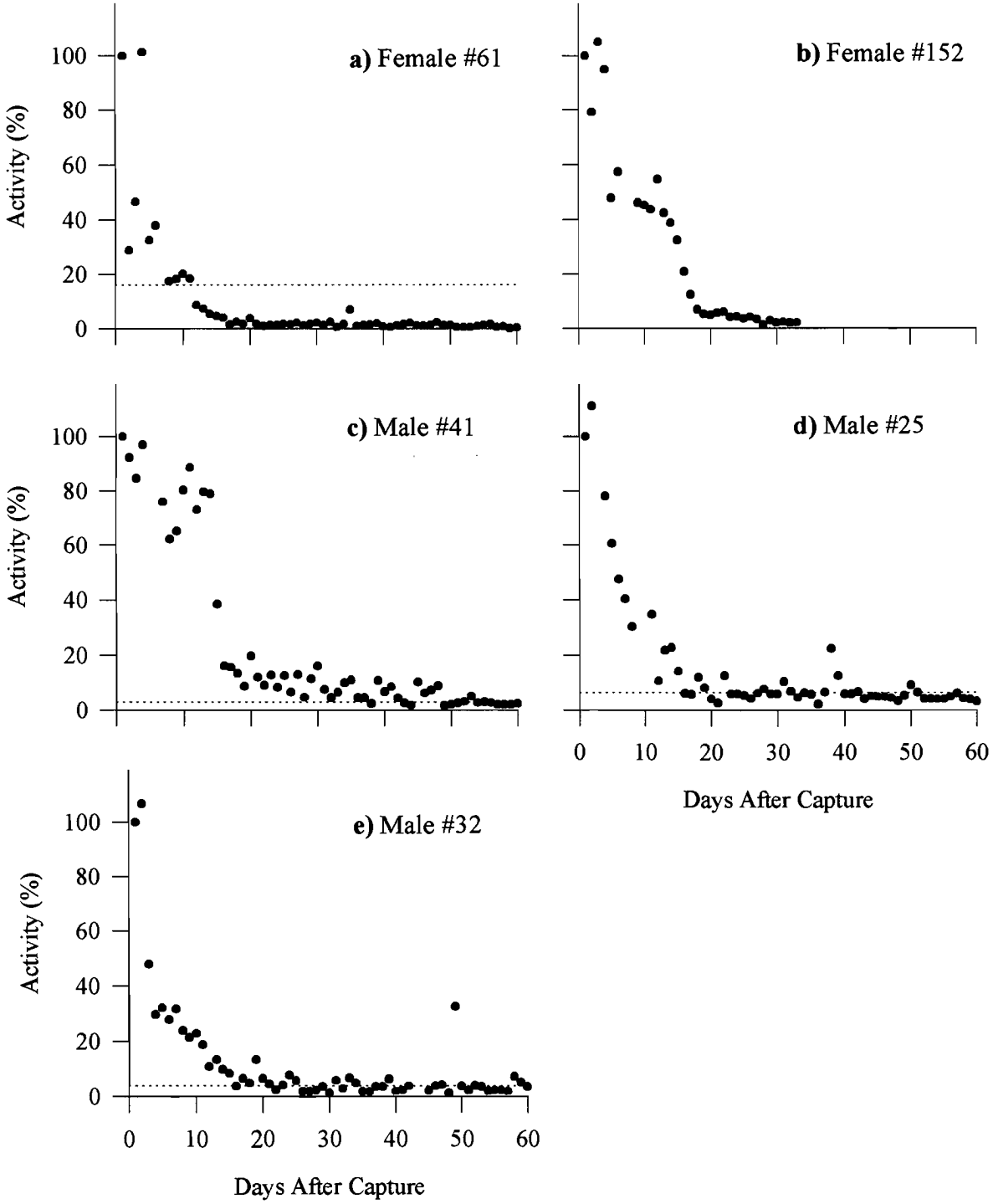


Figure 5.12. Effect of repeated trapping on the activity of three individual NHN wombats. A second trapping period occurred five months following initial capture. Individual 'a' was re-fitted with a new collar package, individual 'b' retained the same collar package and individual 'c' eluded capture. Time zero represents day of the second capture (a & b) or beginning of a 7 night trapping session (c). The time scale represents 30 days prior (negative) and post (positive) capture. Dashed line represents mean activity between 30 and 120 days following initial capture and attachment of radio package.

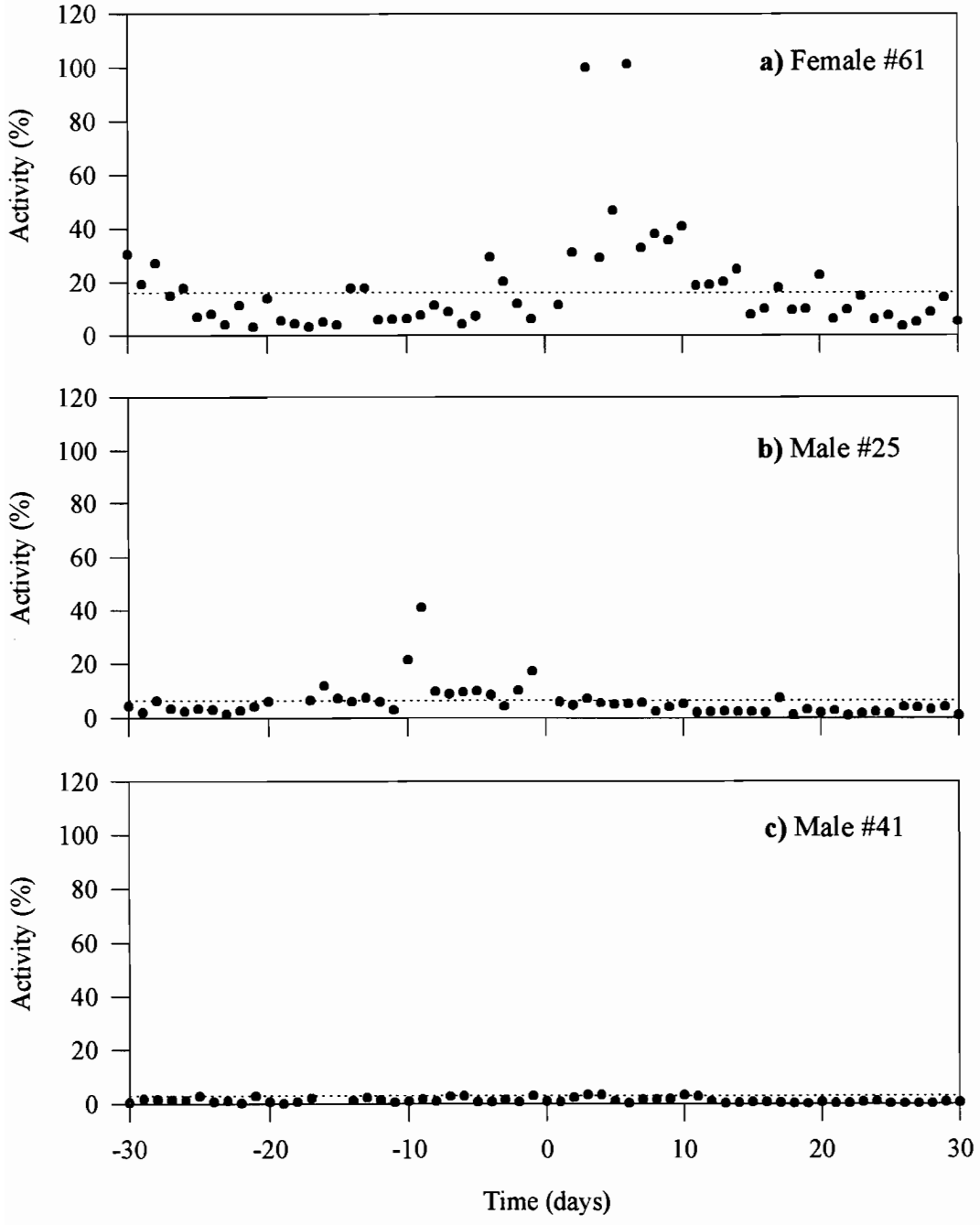


Figure 5.13. Mean hourly activity of NHN wombats expressed as units of 'activity' as recorded by data loggers. The illustrated twenty-four hour period is a mean of 10 continuous days selected at random within each month, with the number of individual NHN wombats included in brackets. Elevated levels of activity in February 1996 were a response to capture as shown in Figure 5.11.

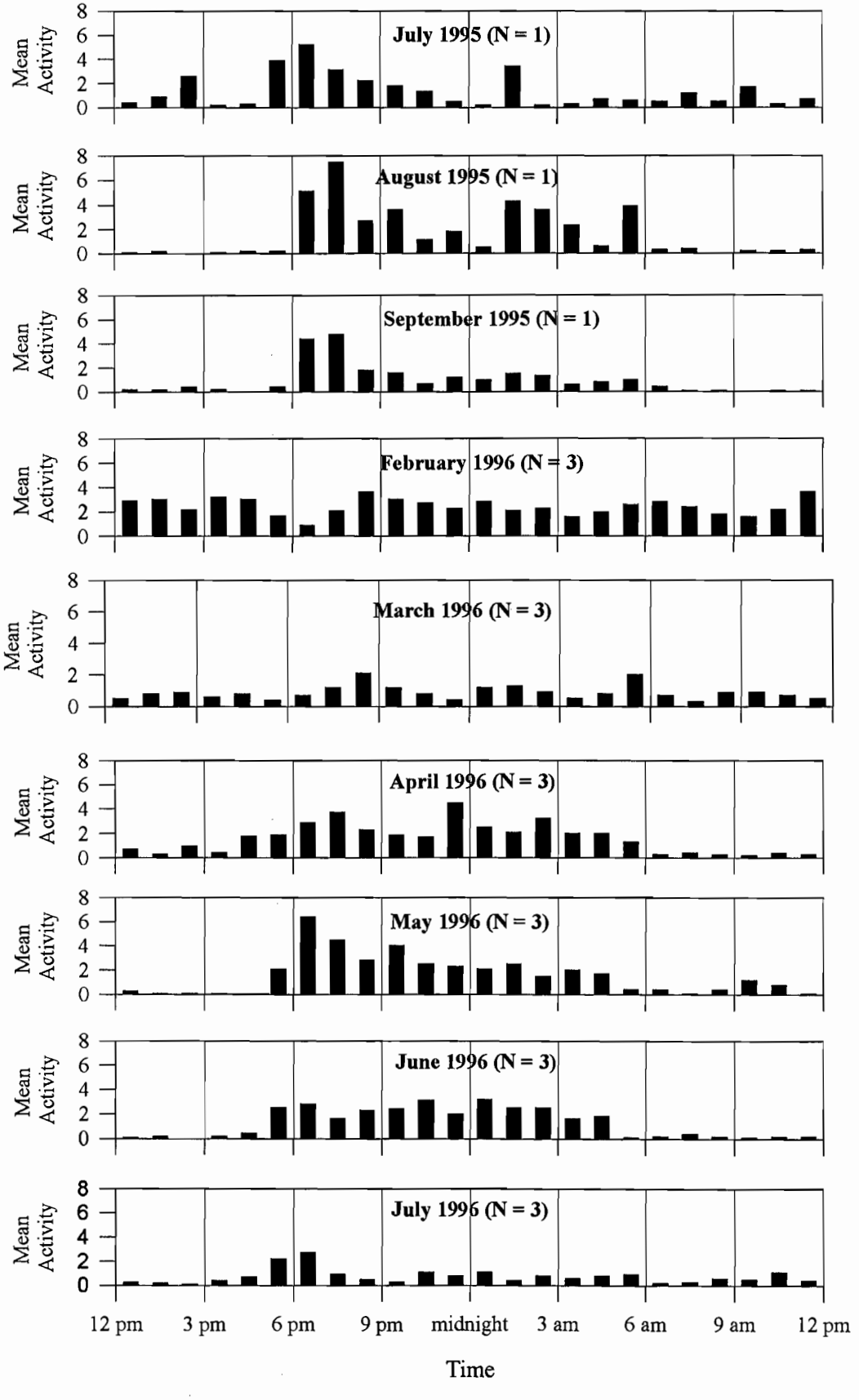
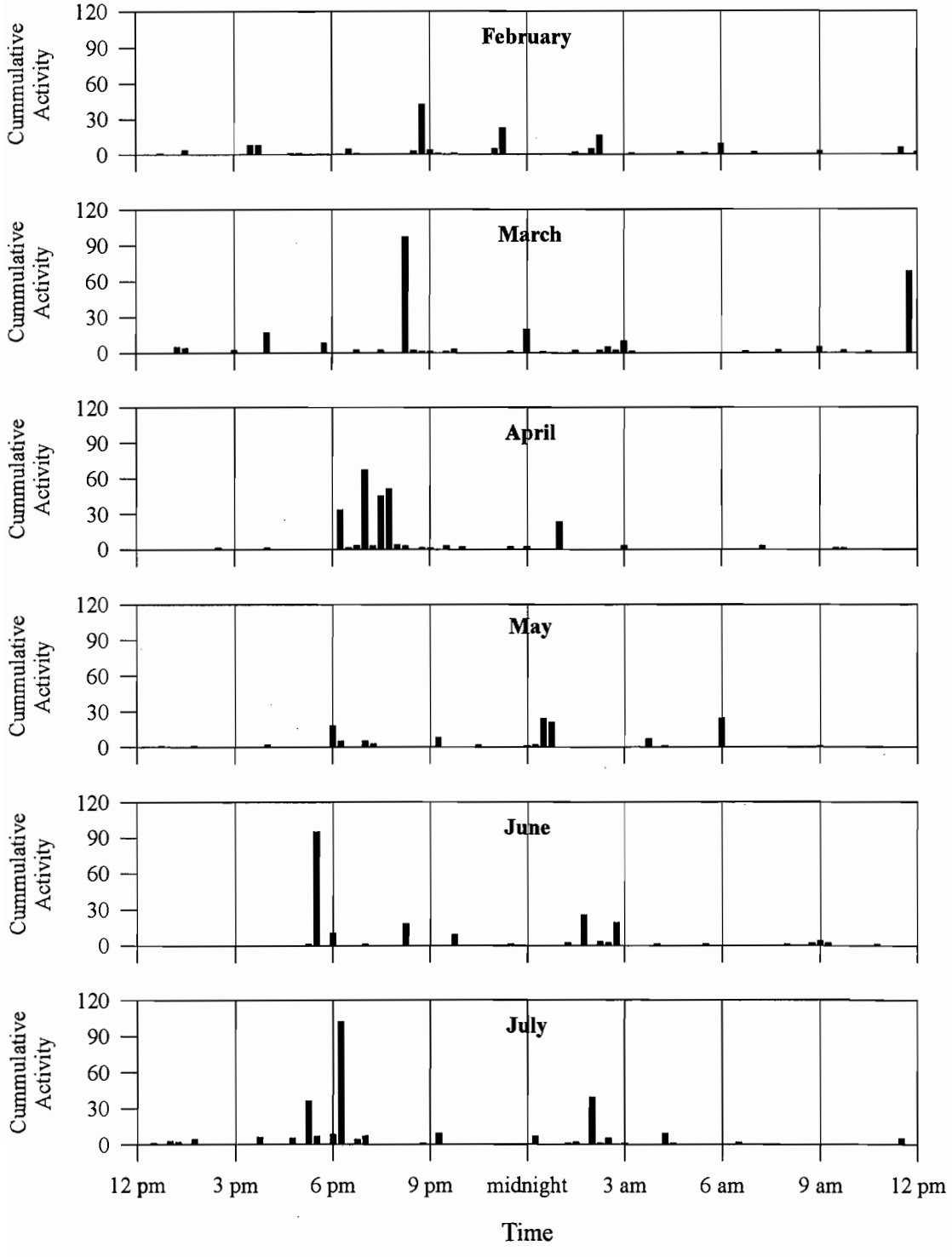


Figure 5.14. Daily trends of activity for Male #25. Activity represents the cummulative activity over a fifteen minute period recorded by the data logger, that is, each cummulative measure of activity represents three individual activity measurements. Twenty-four hour time period is from 12 pm on the 16th to 12 pm on the17th of each month in 1996.



night, bouts of activity occurred at random through any given twenty-four hour period. These findings are repeated for other individuals (Appendix 4).

5.3.3.4 Bouts of Activity

Bouts of activity by the NHN wombat occurred on average every 64.7 minutes (Table 5.4). The mean number of bouts of activity and the mean interval between bouts of activity varied between individuals and between months but this variation was irregular. Likewise the duration of each bout of activity varied between individuals and month, ranging from 5 minutes to 190 minutes in duration. Periods of inactivity ranged from 5 minutes to 675 minutes in duration. However, Figures 5.14 and 5.13 demonstrate that the majority of activity occurred during the night and the longest periods of inactivity occurred during the day.

5.3.4 Above-Ground Activity

5.3.4.1 Types and Duration of Above-Ground Activity

Quantification of above-ground activities of the NHN wombat varied depending on what method was used (Table 5.5). Results from radio-telemetry were conservative in comparison to the temperature differentiation method and are possibly an underestimate of actual levels of activity. However, the results of the temperature differentiation method are confined to the months of May to September. Therefore statistical comparison between the two methods was not possible.

Activity measurements recorded by the data logger were dominated by zeros (equating to inactivity) when the NHN wombats were determined to be above-ground (and also below ground). This suggested that the above-ground activity by the NHN wombat was conservative, with a large proportion of their time spent inactive or possibly foraging in a conservative manner (Table 5.5). Other activities requiring high levels of activity were minimal.

5.3.4.2 Correlation with Habitat Parameters

Mean daily activity showed a significant negative correlation to rainfall and temperature (both minimum and maximum) (Figure 5.15). This suggests that when rainfall was high, activity was low and *vice versa*, with similar trends for

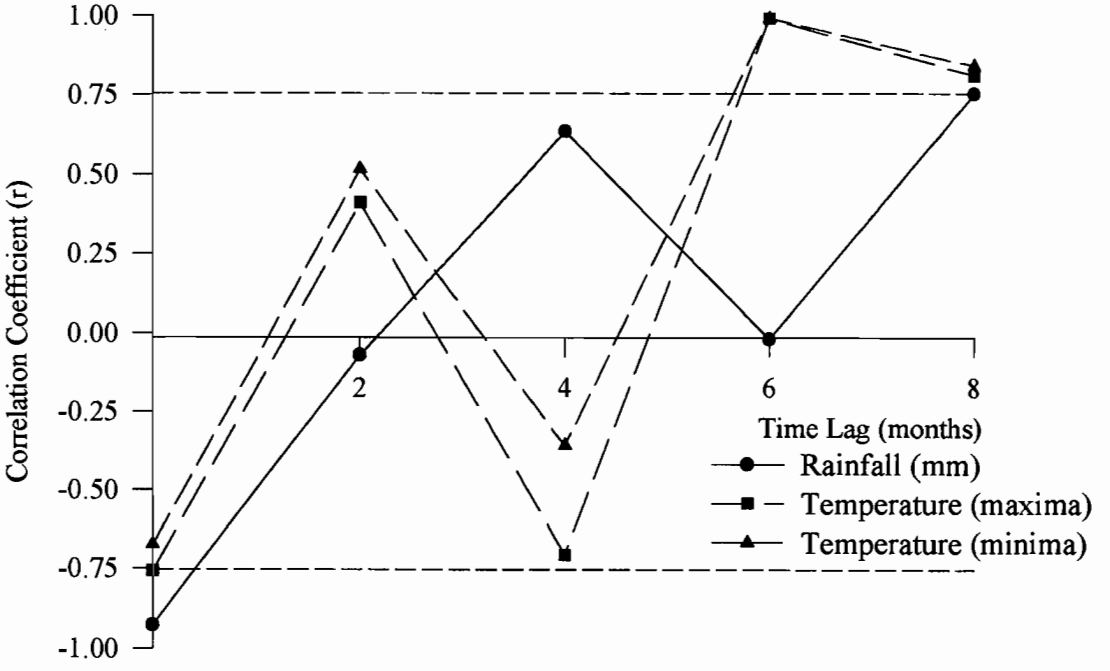
Table 5.4. Bouts of activity by the NHN wombat measured by data loggers. The sampling unit of activity is five minutes, therefore the minumum bout of activity is five minutes, although the actual duration of the bout may be less than five minutes.

NHN Wombat Identification	Month	Days Sampled	Mean Number of Bouts per 24 Hours	S.D.	Mean Length of Bouts (Minutes)	S.D.	Range of Bout (Minutes)	Mean Interval Between Bouts (Minutes)	S.D.	Range (Minutes)
Male #32	July 1995	5	29	9.2	7	0.9	5 to 25	46	16.2	5 to 220
	August 1995	5	27.6	4.4	8.9	2.1	5 to 60	44.5	8.5	5 to 280
	September 1995	5	26.2	7.2	8.12	1.5	5 to 70	51.2	22.6	5 to 510
Female #61	February 1996	5	18.6	2.2	6.1	0.3	5 to 20	70.2	9.5	5 to 290
	March 1996	5	10.8	0.8	6.2	1	5 to 20	124.7	17.4	5 to 675
	April 1996	5	36.4	9.7	8.6	2	5 to 50	33	10.8	5 to 260
	May 1996	5	27.6	5.3	14.9	2.8	5 to 150	38.1	8	5 to 405
	June 1996	5	23.4	8.2	8.2	2.2	5 to 45	60	29.9	5 to 315
	July 1996	5	30.8	5.4	7.8	0.9	5 to 40	40.2	8.1	5 to 270
Male #25	February 1996	5	28	4.1	6.6	0.5	5 to 30	45.6	8.4	5 to 190
	March 1996	5	31.6	5.1	6.3	0.7	5 to 25	40.2	7.5	5 to 190
	April 1996	5	15	2.5	5.8	0.8	5 to 20	91.9	12.9	5 to 395
	May 1996	5	14.8	5	7	0.9	5 to 20	103	59	5 to 485
	June 1996	5	26.6	8.8	8.9	3.4	5 to 65	50.3	22	5 to 385
	July 1996	5	17.8	6.2	5.9	0.5	5 to 20	79.4	34.7	5 to 505
Male #41	February 1996	5	55.6	5.9	12	2.5	5 to 190	14.3	1.6	5 to 120
	March 1996	5	43.2	3.1	7.3	0.6	5 to 40	25.7	2.4	5 to 150
	April 1996	5	25.8	5.5	6.2	0.4	5 to 30	51.2	11.3	5 to 325
	May 1996	5	13.2	3.7	6.8	0.8	5 to 20	114.5	40.1	5 to 590
	June 1996	5	17.8	5.2	6.3	0.6	5 to 15	81.6	32	5 to 390
	July 1996	5	9.2	1.6	5.7	0.6	5 to 15	153	25.1	5 to 490
Grand Means and Maximum Ranges			25.2	11.1	7.6	2.2	5 to 190	64.7	35.6	5 to 675

Table 5.5. Above-ground activity of the northern hairy-nosed wombat. The NHN wombats were determined to be above-ground by:
a) radio telemetry - simultaneous signals received at fixed receiving stations.
b) temperature records from the data loggers - NHN wombats were determined to be above-ground when the thermal difference between the midday temperature (assuming the NHN wombat was in its burrow) and a night-time record was at least 2 °C. See text for further clarification.
Categories of activity followed the subjective scale described in section 5.2.4.3.

	Time	Sex	Time Period (days)	N	Time (Hours)				
					Inactivity/ Stationary Grazing	Foraging	Other	Digging	Daily Total Time Above-Ground
a)	July/August 1995	Male	5	1	2.25	0.7	0	0	2.95
	February 1996	Female	5	2	4.61	1.5	0.05	0.03	6.19
	February 1996	Male	5	2	2.60	1.5	0.3	0.06	4.19
	Mean ± S.D.		5		3.2 ± 1.3	1.2 ± 0.5	0.1 ± 0.2	0.0 ± 0.0	4.4 ± 1.6
b)	July 1995	Male	9	1	3.66	0.89	0.19	0.04	4.78
	August 1995	Male	9	1	6.10	2.85	0.32	0.07	9.34
	September 1995	Male	9	1	4.84	1.85	0.17	0.02	6.88
	May 1996	Female	9	1	1.43	4.23	0.76	0.13	6.55
	June 1996	Female	9	1	3.11	1.27	0.09	0.02	4.49
	July 1996	Female	9	1	5.96	2.36	0.21	0.04	8.57
	May 1996	Male	9	2	7.08	0.58	0.10	0	7.76
	June 1996	Male	8	2	4.35	0.79	0.08	0.02	5.24
	July 1996	Male	9	2	6.67	0.41	0.06	0	7.14
	Mean ± S.D.		8.9		4.8 ± 1.9	1.7 ± 1.3	0.2 ± 0.2	0.0 ± 0.0	6.8 ± 1.7

Figure 5.15. Correlation coefficients (r) of the activity of the NHN wombat (recorded by data loggers) over a lagged time-series with non-accumulative rainfall and temperature (maximum and minimum). [$r = 0.754$; $p < 0.05$; 5 df]



temperature. With time lags, this relationship became positive for rainfall (after 4 months) and significantly positive for temperature (after 6 months).

5.3.5 Temperature Profiles of the NHN Wombat

Measurements of temperature by the data loggers were influenced by both body temperature of the NHN wombat and ambient temperature. Mean trends in the temperature measurements (Figure 5.16) demonstrate significant differences between months (one-way ANOVA, $F_{8, 207} = 56.49$, $p < 0.05$). These trends are repeated for individual NHN wombats (Figure 5.17, Appendix 4) with temperature records in a given twenty-four hour period varying significantly between months (one-way ANOVA, $F_{23, 192} = 1.67$, $p < 0.05$)

5.3.6 Diurnal Activities of the NHN Wombat

Diurnal activities of the NHN wombat varied significantly with time (one-way ANOVA $F_{10, 253} = 2.45$, $p < 0.05$; Figure 5.18). Of the diurnal activities, the majority occurred immediately prior to sunrise or post sunset, but more so post sunset (Figure 5.19). It is most likely that these pre-dawn or post-dusk episodes of activity are associated with retreats and emergences from the burrow respectively. However, evidence from the data loggers suggests that more than 60% of emergences and retreats occur in darkness.

Besides dawn and dusk activity, diurnal activities of the NHN wombat occurred in both full sunlight (Figure 5.19) and partial sunlight that was most likely diffused by the tunnel of a burrow (Figure 5.20). Full sunlight activities occurred primarily in the late afternoon (Figure 5.19) and most frequently in the winter months (Figure 5.18). This activity was most likely to be associated with basking, like the other wombat species (e.g. Wells 1973). Diurnal activities near the entrance of the NHN wombat burrow occurred in most months at low levels, but were most common in the warmer months of December, March and April (Figure 5.18). These visits from within the burrow to near the entrance of the burrow occurred most often between 1000 h and 1400 h (Figure 5.20).

Table 5.6 describes the diurnal activity of Male #25 during the month of March 1996. Details of this single month of data for one individual demonstrated

Figure 5.16. Trends in the temperature profile of NHN wombats for a representative period of twenty-four hours. Temperature measurements represent the hourly mean of a continuous sampling period of 10 days for each NHN wombat with the sample size of NHN wombats in brackets.

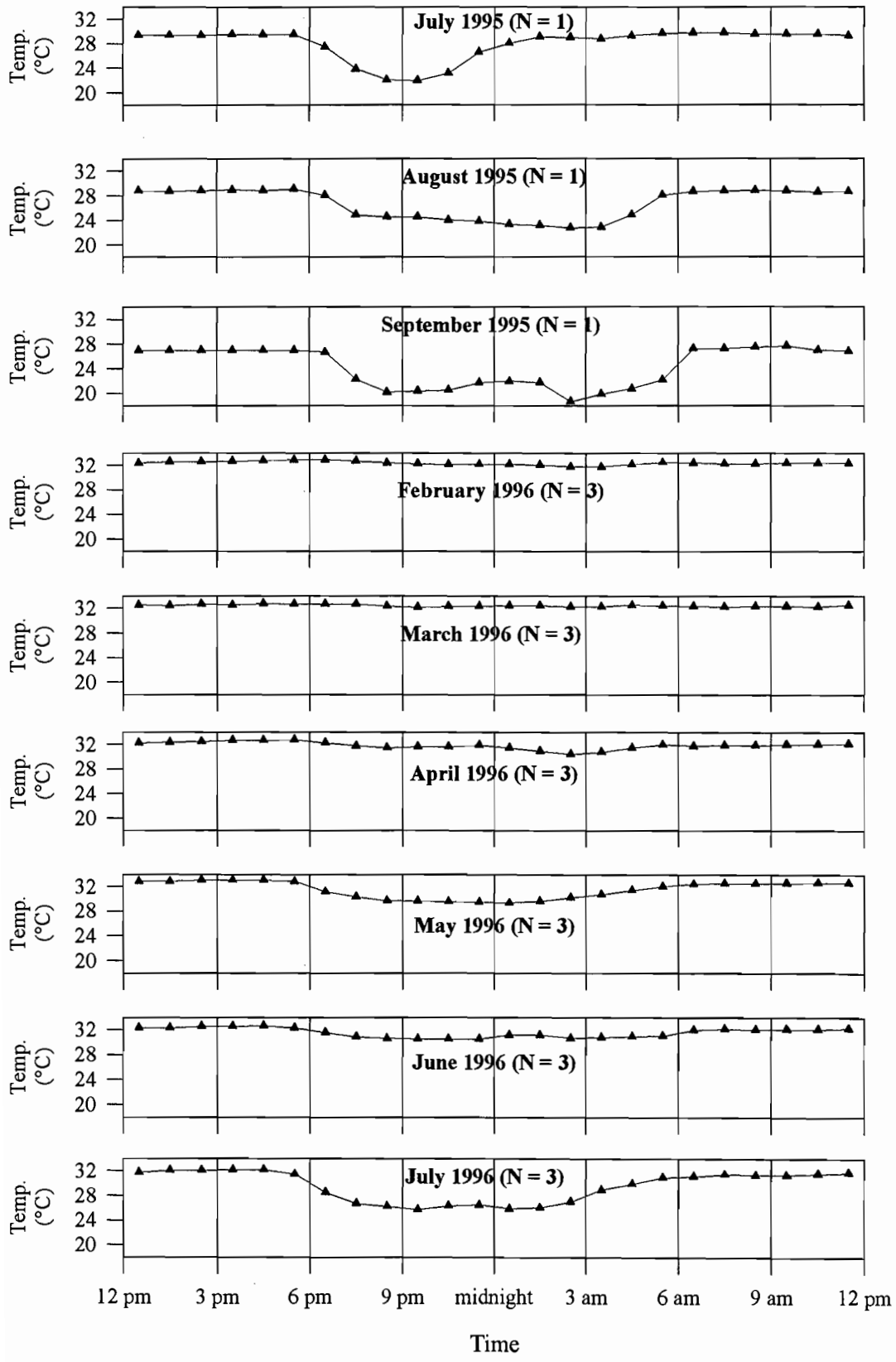


Figure 5.17. A temperature profile of a NHN wombat (Male #25), demonstrating monthly trends in temperature over periods of twenty-four hours. Temperature records are from 12 pm on the 16th to 12 pm on the 17th day of each month. Each data point was the mean value of a block of fifteen minutes (i.e. a mean of three temperature measurements).

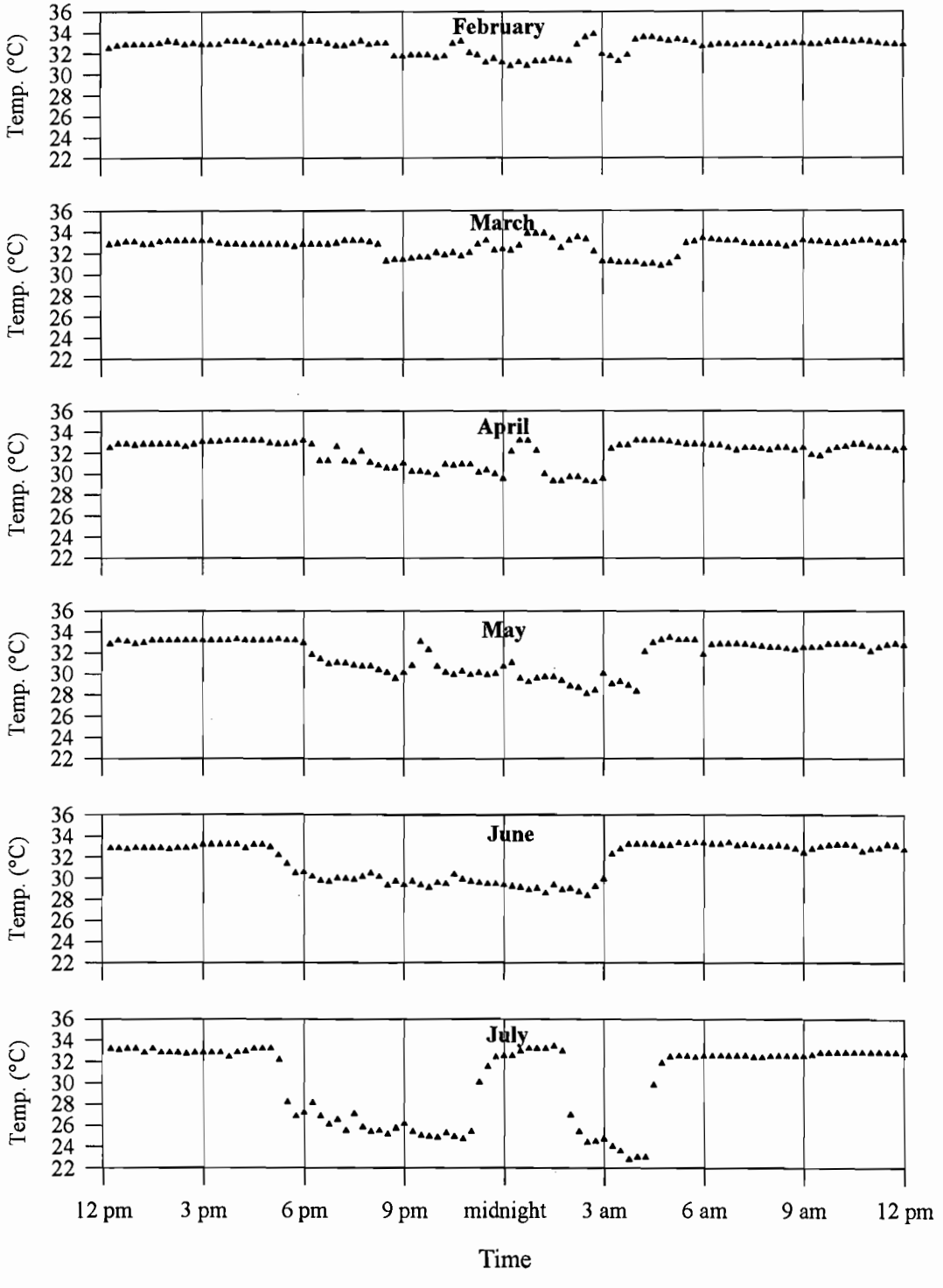


Figure 5.18. Diurnal activities of the NHN wombat. The NHN wombats were determined to be active during daylight when light levels were 10 % or greater above absolute darkness. The NHN wombats were assumed to be active at:
a) Dawn: when light levels were recorded immediately prior to sunrise.
b) Dusk: when light levels were recorded immediately post sunset.
c) Daylight: when light levels recorded by the data logger exceeded 70 % that of absolute darkness during daylight hours;
d) Tunnel: when light levels recorded by the data logger ranged between 10% and 70% above absolute darkness during daylight hours.
Percentage of daylight events represents the total number of days when diurnal activity occurred as a proportion of the total number of days for 15 continuous months of data.

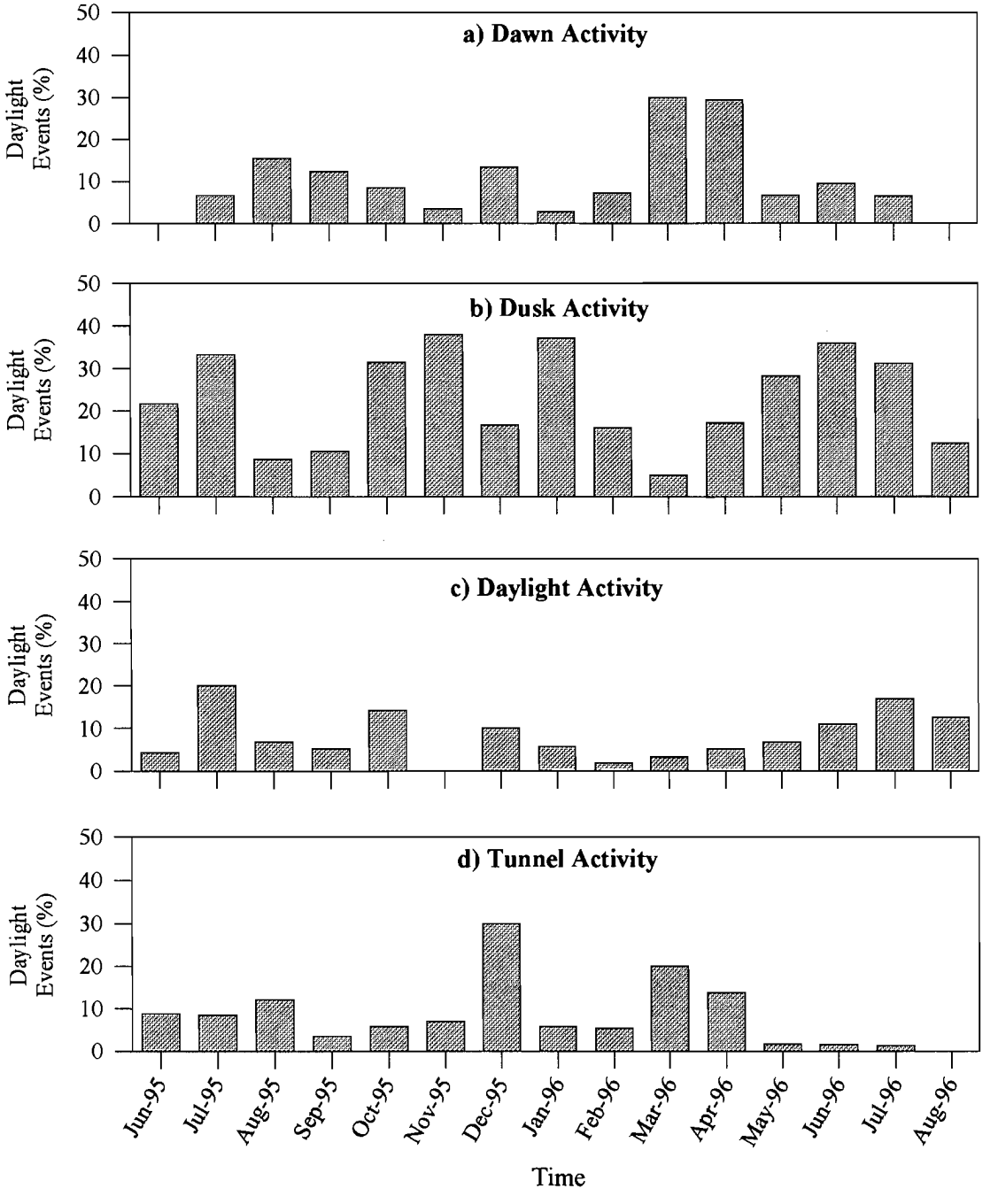


Figure 5.19. Diurnal activities occurring in full sunlight. The NHN wombats were determined to be in full sunlight when levels of light exceeded 70% that of absolute darkness. The percentage of daylight events is the mean number of days (± one S.D.) where daylight activity occured as a percentage of the total number of days examined (15 continuous months).

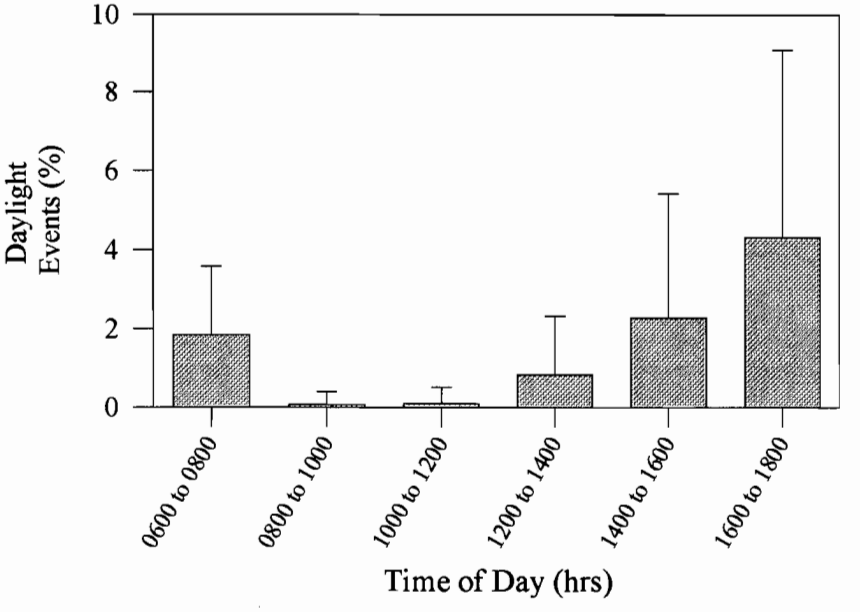


Figure 5.20. Diurnal activities of the NHN wombat occurring within three metres of the entrance of the burrow. Allocation of diurnal activities into this category occurred when light levels ranged between 10 % and 70 % above absolute darkness, during the hours of daylight. The percentage of daylight events is the mean number of days (± one S.D.) where daylight activity occured as a percentage of the total number of days examined (15 contniuous months).

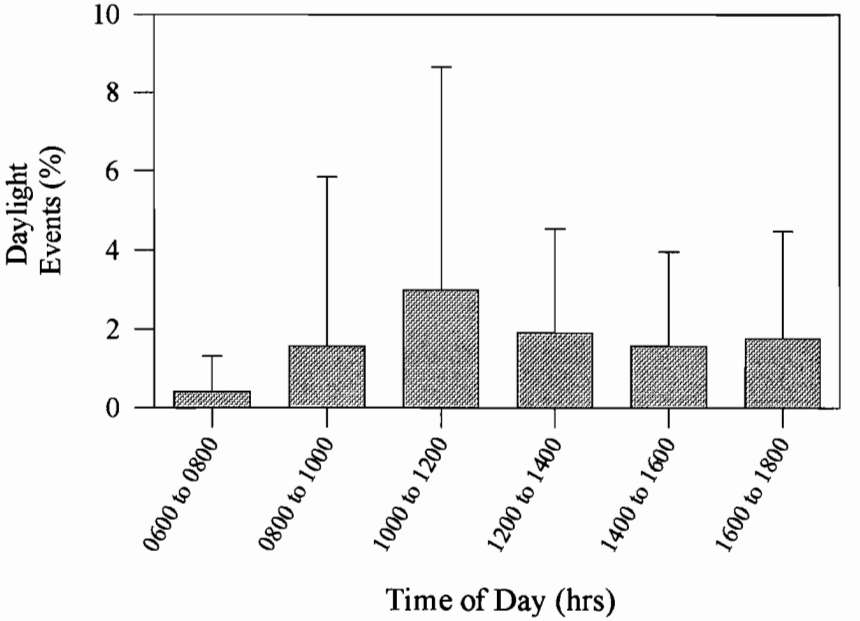


Table 5.6. Diurnal activities of Male 25 in March 1996. Light levels were detected by the data loggers, with a light level of 0% equating to absolute darkness and 100% equating to full sunlight. Category definitions and abbreviations include:
- Dawn - when light levels are above 10% prior to sunrise (SR);
- Dusk - when light levels are above 10% following sunset (SS);
- Daylight - when light levels are above 70% during daylight hours;
- Tunnel - when light levels are between 10% and 70% during daylight hours.

Date	Time	Relative Light Above Darkness (%)	Category	Time of Sunrise/Sunset
1/03/96	5:54 to 6:14	72.1	Daylight	SR: 6:09
3/03/96	5:39 to 6:04	14.9	Dawn	
6/03/96	5:44	12.3	Dawn	SR: 6:12
6/03/96	13:01 to 14:39	17.6	Tunnel	
6/03/96	18:44 to 19:19	43.5	Dusk	SS: 18:37
7/03/96	13:29 to 14:14	25.9	Tunnel	
7/03/96	14:59 to 15:54	19.8	Tunnel	
8/03/96	5:44 to 6:14	42.2	Dawn	SR: 6:12
9/03/96	6:29 to 6:59	76.1	Daylight	SR: 6:13
9/03/96	18:49 to 18:59	35.2	Dusk	SS: 18:35
10/03/96	18:24 to 18:59	64.6	Dusk	SS: 18:34
12/03/96	14:59 to 15:29	24.2	Tunnel	
15/03/96	9:39 to 10:04	15.8	Tunnel	
16/03/96	10:29	12.3	Tunnel	
18/03/96	5:49 to 5:59	15.4	Dawn	SR: 6:16
18/03/96	12:04 to 13:59	18.4	Tunnel	
19/03/96	10:54 to 11:14	11.4	Tunnel	
19/03/96	13:49 to 15:19	39.1	Tunnel	
20/03/96	11:54 to 12:19	17.6	Tunnel	
21/03/96	12:29 to 12:49	37.8	Tunnel	
23/03/96	6:04	37.4	Dawn	SR: 6:12
23/03/96	13:54 to 14:34	14	Tunnel	
24/03/96	12:29	10.1	Tunnel	
24/03/96	13:09	41.8	Tunnel	
25/03/96	5:49 to 6:04	42.2	Dawn	SR: 6:19
26/03/96	6:09	39.6	Dawn	SR: 619
30/03/96	11:29	15.4	Tunnel	

that diurnal activities are essentially random. However, there was a larger proportion of diurnal activities occurring in the first three metres of the burrow from the entrance than the other three categories. The behavioural significance of visits close to the entrance is unknown.

5.4 DISCUSSION

5.4.1 Home-range size of the NHN wombat

Estimates of the home-range size of the NHN wombat are close to those of Johnson (1991a, b), both in total home range and core home range. Therefore the fundamental conclusion by Johnson (1991a) that the ranging behaviour of the NHN wombat is extremely conservative is supported by this study.

5.4.1.1 Seasonal Trends: The Influence of Environmental Conditions

The forage of Epping Forest National Park fluctuates seasonally in quality and quantity (Chapter 2). As forage quantity and quality decrease, grazing animals usually compensate by increasing feeding times and range size (Owen-Smith 1982). Johnson (1991a) found similar trends for the NHN wombat. He found that there was distinct variability in the size of the home range of the NHN wombat with season; smallest when forage conditions were optimum (December to May) and largest when forage conditions were poor (June to November). Although these changes in home range were statistically significant, the changes represented only a modest change in actual area (Johnson 1991a). However, the seasonal trends observed by Johnson (1991a) were not repeated in this study.

The findings of Johnson (1991a, b) can not be easily disputed because his studies were very comprehensive. Johnson's (1991a) study encompassed six radio-tracking sessions over a twelve-month period, compared to only two sessions encompassing seven months in the current study. Further, Johnson's (1991a, b) data was gained from three radio-tracking towers as opposed to two, thereby having a higher precision (White and Garrott 1990).

Although these two factors highlight shortcomings in the current study, a further three differences between the two studies may offer explanations for

discrepancies. First, the recording schedule of each session of radio tracking differed between the two studies. Johnson (1991a) used an alternate six-hour shift system (dusk to midnight one night, midnight till dawn the next), whereas the current study relied on one continuous shift (dusk till dawn). Secondly, there were differences in the range of ambient temperatures encountered during each session of radio tracking. The minimum temperature in July/August of this study was 0°C (range of 0 to 23°C) compared to Johnson's (1991b) minimum of 4°C (range 4 to 18°C). Likewise, the minimum temperature in February in this study was 21°C (range 21 to 35.5°C) compared to Johnson's (1991b) minimum of 14.5°C (range 14 to 24°C). Very low and very high temperatures will reduce the activity of the wombat and will therefore impact on the measurements of home range size. The influence of temperature is discussed further section 5.4.2.2. Finally, there is no way of accurately assessing the condition of the forage between the two studies, particularly since there has been a substantial change in the composition of the vegetation between the two studies (Chapter 2).

5.4.1.2 Comparison with the Macropodidae

The total home range of the NHN wombat is considerably smaller than those of members of the Macropodidae of equivalent body size (Figure 5.8). Indeed the home range of the NHN wombat is comparable to the Tammar wallaby, *Macropus eugenii* (4 - 10 kg), rather than of species of equivalent body size such as the eastern grey kangaroo, *M. giganteus* (3.5 to 32 kg for females), or the western grey kangaroo, *M. fuliginosus* (4.5 to 27.5 kg for females). However, each of the three *Macropus* species share similar food preferences to the NHN wombat (Norbury *et al.* 1989). This raises the question of how the NHN wombat is able to maintain such a small home range compared to the sympatric *M. giganteus*?

Physiological constraints on the size of home ranges

McNab (1963) suggested that the home range of an animal is directly determined by its body size and metabolic rate. Harestad and Bunnell (1979) suggest that this relationship is further influenced by the type of foods consumed (trophic status) and its food resource (density and productivity). They further

suggest that the home range of herbivores, in particular, is directly affected by the quality and quantity of available forage.

The NHN wombat and the eastern grey kangaroo live sympatrically at Epping Forest National Park (EFNP). Both are of similar body size, their diets overlap significantly and the availability of forage resources to both herbivores is almost equal (Chapter 6), yet their home ranges are considerably different. Jaremovic and Croft (1987) estimated the home range of the eastern grey kangaroo to be about 300 ha at Sandy Creek Reserve, Bago State Forest, New South Wales. Despite the habitat of EFNP being distinctly different from that described by Jaremovic and Croft (1987) (Bago State Forest is cool temperate, has an annual rainfall of 1000 to 1250 mm and two seasons of pasture growth), the dynamics of the eastern grey kangaroo at EFNP (Chapter 6) suggest that their home ranges are considerably larger than the NHN wombat, although perhaps not to the extent of Jaremovic and Croft (1987). Therefore, using the reasoning of McNab (1963) and Harestad and Bunnell (1979), the lower metabolic rate of the NHN wombat, coupled with its physiological and behavioural dependence on its burrow, are the prime explanations why the home range of the NHN wombat is significantly smaller than the eastern grey kangaroo.

5.4.2 Activity of the NHN Wombat

5.4.2.1 Short-term Effect of the Radio Package

The attachment of the radio package to the NHN wombat caused an immediate behavioural response. The elevated levels of activity following the fitting of the collar package (Figure 5.11) were likely to be a direct response to the collar rather than a response to capture or impediment of regular feeding activities because there were limited effects on activity following subsequent captures (Figure 5.12). With a larger sample size, these trends could be clearly quantified. The type of unusual activity during the adjustment period is unknown, but was most likely attempts by the NHN wombat to remove the collar.

Adjustment to the collar occurred for a period of at least 10 days after capture. Therefore, in studies where a radio package is attached to an animal, an adjustment period incorporating the animal's response to the radio package and to

capture needs to be considered. Accounting for the adjustment period, the results of the activity of the NHN wombat described in this study are likely to be a true representation of normal NHN wombat behaviour.

5.4.2.2 Foraging Activity of the NHN Wombat

Like its ranging behaviour, the activity schedule of the NHN wombat was very conservative. On any given day, the average number of activity bouts was only 25.2 ± 11.1 with each bout lasting only 7.6 ± 2.2 minutes (Table 5.4), suggesting that the wombat is active for only 3.2 hours per day. Defining the types of activity measured by the data loggers was subjective and requires detailed calibration. Nonetheless, the arbitrary scale described in section 5.2.4.3 did allow useful categorisation of the activity of the NHN wombat.

The NHN wombat spends between 2.95 hr and 9.34 hr above-ground per night (Table 5.5). The majority of this time is spent being inactive or grazing in a conservative manner. Separation between inactivity and conservative grazing is impossible without direct observation. However, direct observations on SHN wombats (Gaughwin 1981) and Common wombats (Triggs 1988) indicate that wombats spend a significant proportion of time inactive. The function of this above-ground inactivity is unclear. Only limited time is devoted to other activities such as actively foraging, ranging, scratching or digging. Presumably these other activities are minimal because of their high energy costs.

Body size constraints on foraging

The foraging activity of large herbivores is constrained by body size (Demment and Van Soest 1985), forage availability (Belovsky and Slade 1986) and digestive strategy (Illius and Gordon 1983). In general, large herbivores tend to allocate more time to foraging than small herbivores (Owen-Smith 1992). Likewise hindgut fermenters adopting a 'bulk feeding' strategy should allocate more time to feeding than foregut fermenters (Demment and Van Soest 1985, Bunnell and Gillingham 1985). For example, horses spend more time feeding than equivalent-sized foregut fermenters such as buffalo or domestic cattle (Table 5.7). It follows that the NHN wombat should allocate at least as much time to foraging as

Table 5.7. A comparison of foraging times of free-ranging herbivores..

Species	Common Name	Adult Body Mass[1] (kg)	Mean Grazing and/or Browsing Time (hr)	Time Range	Source
Aepyceros melampus	Impala	40 to 65	9.6	8.0 to 10.7	Jarman and Jarman (1976)
Syncerus caffer	Buffalo	300 to 900	10.0	9.0 to 10.5	Sinclair (1977)
Rangifer tarandus	Caribou	81 to 110	12.7	9.2 to 20.2	Trudell and White (1981)
Rangifer tarandus	Reindeer	60 to 318	11.8	-	Trudell and White (1981)
Cervus elaphus	Elk or Red Deer	75 to 340	10.6	7.8 to 14.3	Craighead *et al.* (1973)
Macropus giganteus	Western Grey Kangaroo	3.5 to 66	-	10.5 to 18.5	Clarke *et al.* (1989)
Kobus ellipsiprymnus	Waterbuck	50 to 300	8.9	7.0 to 10.7	Spinage (1982)
Giraffa camelopardalis	Giraffe	550 to 1930	-	10.3 to 12.5	Pellew (1984)
Equus caballus	Domestic Horse	530	14.3	12.2 to 15.4	Duncan (1992)
Bos taurus	Domestic Cattle	800 to 1000	10.9	5.4 to 12.7	Arnold (1985)
Ovis aries	Domestic Sheep	32	9.4	8.4 to 10.6	Arnold (1985)
Marmota flaviventris	Yellow-bellied Marmot[2]	3.5 to 7	-	1.7 to 3.84	Armitage (1991)
Lasiorhinus krefftii[3]	NHN Wombat[2]	30.1 to 32.5	5.9	3 to 8.3	Current Study
Lasiorhinus krefftii[4]	NHN Wombat[2]	30.1 to 32.5	1.6	0.7 to 4.2	Current Study

[1]Body Mass data from Nowak (1991) and Strahn (1995).
[2]Burrowing herbivores.
[3]Foraging times of the NHN wombat estimated by the logger data where activity records from zero to twenty in a five minute period were assumed to be indicative of foraging when the NHN wombat was considered to be above-ground. This measure is likely to overestimate foraging.
[4]Again, logger data was used to estimate foraging times but only if activity records were between one and twenty in a five minute period and the NHN wombat was considered to be above-ground. This method is likely to underestimate foraging.

herbivores of equivalent body size such as the eastern grey kangaroo if forage or energy requirements are similar.

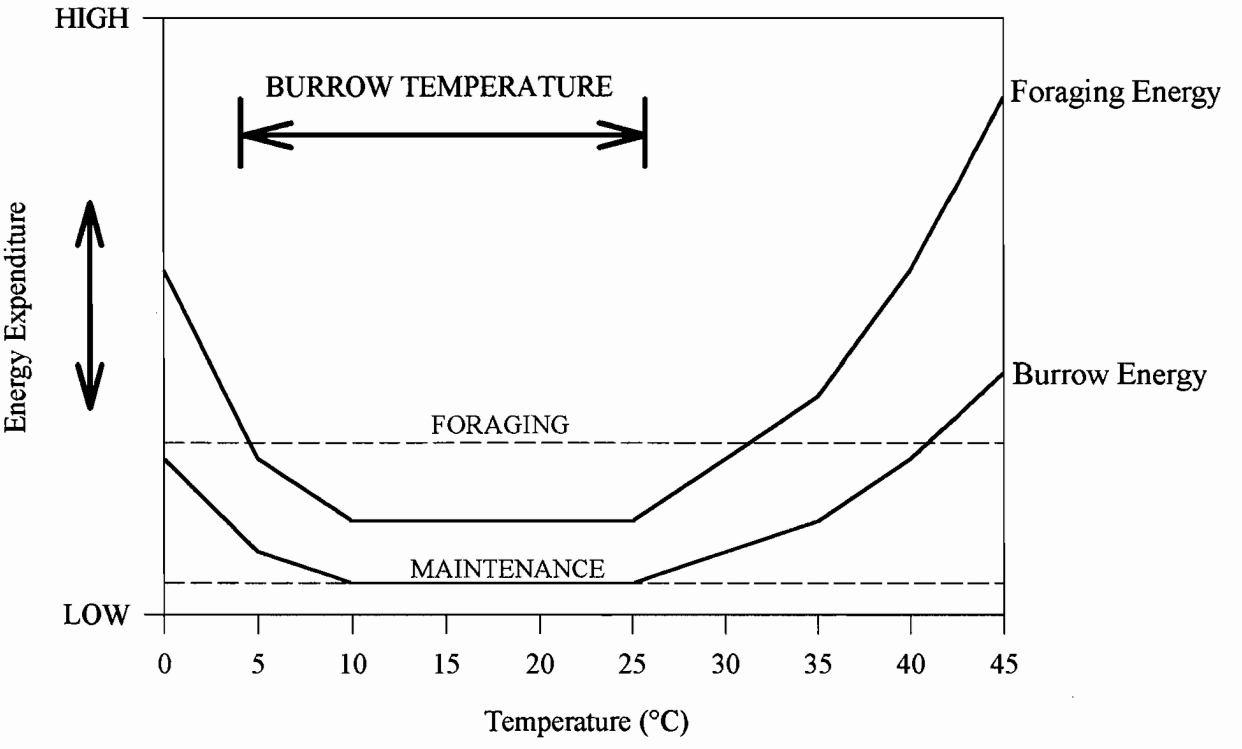
The NHN wombat devotes considerably less time to foraging than herbivores of similar body mass such as the eastern-grey kangaroo. Instead the NHN wombat allocates a similar amount of time to foraging as the smaller yellow-bellied marmot (*Marmota flaviventris*) (Table 5.7). Although, the yellow-bellied marmot shares the fossorial habits of the NHN wombat, although other aspects of its biology differ (Armitage 1992).

The low level of activity by the NHN wombat is reflected in its extremely low field metabolic rate (3802 kJ/day for the whole animal, Murray Evans pers. comm.). Further, wombats have low nitrogen requirements, a capacious colon and the ability to selectively retain fibre, resulting in similar digestive efficiency as larger animals (Barboza 1989). Low metabolic rate and high digestive efficiency allow a high energy gain per forage effort. Consequently, the forage activities of the NHN wombat can be described as conservative but efficient.

Thermal constraints on foraging

The activity of the NHN wombat is directly influenced by the thermal conditions of the environment, both extremes of heat and cold. Thermal conditions can affect the time allocated to foraging and the energetic cost of foraging (Melcher *et al.* 1990). For example, in the hypothetical model illustrated in Figure 5.21, energy expenditure by the NHN varies according to the thermal environment. Firstly, consider the range of temperatures encountered in the burrow. Wells (1973) found that the burrow of the SHN wombat ranged in temperature from 4°C to 28°C, 3 metres from the entrance. In this model, an energy cost above maintenance requirements occurs when the burrow temperature is less than 10°C or greater than 25°C. The use of several burrows (Johnson 1991a) may allow the NHN wombat to maintain burrow temperatures within this range. For example, returning to the same burrow for several consecutive days during the cooler months may allow the accumulation of body heat. Conversely, using multiple burrows in the warmer months allowing body heat to dissipate between visits.

Figure 5.21. A hypothetical model describing energy expendidature of a NHN wombat with variations in temperature. Values described in this figure are hypothetical. As a guide, burrow temperatures for SHN wombats range from 4°C to 28°C (Wells 1973). The lower threshold represents the minimum energy required for maintenance. The upper threshold is that of foraging and above-ground activity, above this threshold energy is expendid at a higher rate than it can be gained by foraging. Again, it should be emphasised that these thresholds are hypothetical estimates and require experimental validation.



However, this is only speculation and requires further investigation. Now consider the foraging activity of the NHN wombat. The optimum temperature range for foraging by the NHN wombat in this model is between 5°C and 30°C. If the ambient temperature is above 30°C or below 5°C, the energy gained through forage intake is less than the energy expended. Therefore, it is more energy efficient for the NHN wombat to return to the burrow when the temperature of the burrow is at an acceptable temperature (10°C and 25°C). By restricting its foraging activities to periods of optimum temperatures, the NHN wombat can minimise its total energy expenditure and maximise its energy intake.

Although this model is hypothetical, the effects of temperature have been briefly investigated for the SHN wombat and the common wombat. It is also possible that, rather than energy, water may be more limiting at high temperatures which then may cause a change in the behaviour of the wombat. Wells (1978b) found that the SHN wombat did not emerge from its burrow if the ambient temperature was above 26.7°C. Likewise, Brown (1984) found the common wombat only emerged from its burrow if the ambient temperature was below 25°C. However, neither study identified the effect of minimum temperatures. The relationship between temperature and activity is fundamental to the ecology and physiology of wombats, and therefore requires further investigation, particularly for the NHN wombat.

Environmental Constraints on Foraging

Rainfall is a key variable at EFNP. Habitat variables such as nitrogen concentration and primary productivity correlate positively with rainfall. Activity of the NHN wombat also correlates negatively with rainfall. This suggests that after rain, environmental conditions are most favourable and in order to meet energy requirements, activity can be reduced. However, peak rainfall also occurs with peak temperatures, and temperature may constrain foraging time rather than forage conditions.

5.4.2.3 Diurnal Activities

Above-ground Activities

The pattern of a dusk emergence and a dawn retreat of the NHN wombat is similar to those of common wombats (Triggs 1988) and SHN wombats (Wells 1973, Gaughwin 1981). Wells (1973) found that the emergence pattern of SHN wombats was seasonal, with early evening emergences in the cool winter months and late evening emergences in warmer summer months. While the data obtained from the light sensor was insufficient to conclude specific emergence and retreat times for NHN wombats, it is more than likely that trends in emergences are seasonal. By using remote logging systems described by Taylor (1977) or Steele *et al.* (1995) these trends could be examined.

Basking was evident in the cooler months, from May to September (Figure 5.18). The basking activities of SHN wombats are directly correlated with cold nights, with basking most often occurring in the cooler winter months (Wells 1973). Wells (1973) suggested that the basking activity by the SHN wombat was a means of behavioural thermoregulation. It seems likely that this behaviour is also used by the NHN wombat.

Overall, diurnal activities by the NHN wombat in full daylight are not very common and largely restricted to basking or late retreats/early emergences. Other diurnal activities that can be confirmed include drinking from temporary water following rain (A. Horsup and V. Steele pers. comm.), but such activities are rare.

Below-ground Activities

Activity by all three species of wombat within the burrow is not well understood. Brown and Young (1982), using radio telemetry, found that common wombats remain in the same location within the burrow with the occasional burst of activity from scratching, changing sleeping position or moving within the burrow. Triggs (1988) reported that common wombats move toward the entrance of the burrow in the late afternoon, possibly to assess the outside temperature. It would be easy to dismiss underground activity (as shown in Figure 5.11) as scratching, moving sleeping position or within burrow movements. It is likely that the majority of these activity events can be attributed to the aforementioned

behaviours. However, why would the wombat choose to move toward the entrance of the burrow, particularly in the hottest months of December, March and April (Figure 5.18) at the hottest times of the day (Figure 5.20)? Results clearly shows that this NHN wombat regularly moved to an area in the burrow where light can penetrate (approximately 3 metres from the entrance). Most of these activities in the first three metres of the burrow occurred in the middle of the day (from 10:29 am to 3:29 pm). Rather than a behavioural means to assess outside temperatures or a consequence of moving from one section of the burrow to another, it could be a means of promoting airflow within the burrow and increasing the thermoregulatory and gas exchange capacity of the burrow. The depth and length of a wombat burrow creates a microclimate stable in temperature and humidity (Wells 1973, Contreras and McNab 1990). However, the stability of the microclimate is influenced by several factors including the airflow caused by temperature and pressure gradients between the burrow and the environment (Maclean 1981, Vogel *et al.* 1973). This is most apparent in summer months when heat loss through conduction, convection and evaporation from the animal may be reduced. Then, burrow conditions may become uncomfortably warm, hypoxic or hypercapnic (Wilson and Kilgore Jr. 1978, Withers 1978, Maclean 1981, Contreras and McNab 1990). By coming within three metres of the entrance the NHN wombat may essentially act as a plunger, promoting gas flow within the burrow and allowing excess heat to dissipate. Movement within the daylight region of the burrow may then maximise the airflow potential. This hypothesis requires further testing, particularly on SHN wombats where added constraints imposed by the soil structure may impede flux of gas between the burrow and the soil spaces in close proximity to the wombat.

5.4.3 Concluding Remarks

The foraging behaviour of the NHN wombat is very conservative, both in range area and activity levels. Consequently, energy expenditure through activity is minimal. This allows the NHN wombat to be well-suited to a forage environment that is poor (Chapter 2) and enables it to maintain constant levels of body composition throughout the year (Chapter 4). Further, the semi-fossorial lifestyle of the NHN wombat results in additional energy conservation because energy costs

for thermoregulation are reduced. This is critical in a habitat where the ambient temperatures are extreme.

CHAPTER 6

THE FEEDING ECOLOGY OF THE EASTERN GREY KANGAROO: A COMPARISON

6.1 INTRODUCTION

The geographic distribution of the Northern Hairy-nosed (NHN) wombat is shared by three macropod species: the swamp wallaby, *Wallabia bicolor*; the rufous bettong, *Aepyprymnus rufescens*; and the eastern grey (EG) kangaroo, *Macropus giganteus*. The swamp wallaby is considerably smaller than the NHN wombat and ranges between 10.3 kg to 20.5 kg (Marchant 1995), preferring browse rather than grasses (Hollis *et al.* 1986, Merchant 1995). The rufous bettong is smaller, between 3 kg to 3.5 kg (Seebeck and Rose 1989). It also prefers browse, forbs and grasses but readily consumes tubers, roots, seeds, and fungi if available (McIllwee 1994, Dennis and Johnson 1995). The EG kangaroo is comparable to the NHN wombat in body mass and dietary preference, and therefore could potentially interact competitively with the NHN wombat. There have been several studies investigating the diet of free-ranging EG kangaroos (e.g. Kirkpatrick 1965, Griffiths and Barker 1966, Griffiths *et al.* 1974). Each study reached the general conclusion that EG kangaroos readily consume grasses and forbs in a non-selective manner (Norbury *et al.* 1989). Crossman (1988) briefly examined the diet of the EG kangaroo, concluding that the EG kangaroo and the NHN wombat share similar dietary preferences consuming almost 100% grass.

The similarities of body mass and dietary preference of the NHN wombat and the EG kangaroo present an ideal opportunity to compare the feeding strategies of two grazing herbivores with different digestive systems and ranging

behaviours. The response to temporal changes in the quality and quantity of the pasture by these two herbivores is the prime focus of this chapter. In addition, from a management perspective, it is important to quantify resource overlap between the NHN wombat and the EG kangaroo, particularly if resources become limiting and competition develops. This chapter, therefore, also describes the diet and population dynamics of the EG kangaroo at Epping Forest National Park (EFNP) and spatial interactions between the NHN wombat and the EG kangaroo.

6.2 METHODS

6.2.1 Diet Analysis

The diet of the EG kangaroo on EFNP was assessed using the same procedures described in Chapter 3 from EG kangaroo faecal pellets collected opportunistically from the seven BOTANAL transects (Figure 2.3). Faecal pellets were collected if they were characteristic of recent deposition, indicated by colouration and texture of the pellet. Collected pellets were placed dried at 60°C for 24 hours and stored at -5°C until preparation for analysis.

All dietary analyses of EG kangaroo faecal pellets were based on the following assumptions:

a) EG kangaroo defecation events consist of multiple pellets (Johnson et al. 1987); and

b) one defecation event was representative of one feeding bout.

Therefore the botanical composition of one pellet was assumed to be the same as another within the single defecation event using the same assumptions described in Chapter 3.

Measuring Dietary Overlap

Overlap between the EG kangaroo and NHN wombat was measured using Jarman's (1971) measure of overlap and an adaptation of the Feinsinger *et al.* (1981) proportional similarity (*PS*) index to describe niche breadth. The Jarman (1971) measure provides a descriptive account of overlap between herbivore species based purely on shared occurrence of plant species in the diet. In the example described by Jarman (1971), herbivore species 'P' consumes 10% of plant

species 'A', with herbivore species 'Q' consuming only 2% of plant species 'A'. The overlap between herbivores 'P' and 'Q' for plant species 'A' is the proportion that is common to both herbivore species: i.e. 2%. The sum of overlaps for all food types provides a measure of total dietary overlap.

The Feinsinger *et al.* (1981) *PS* index is calculated using the following equation:

$$PS \text{ Index} = 1 - 0.5(\Sigma \mid p_i - q_i \mid) \qquad \text{Equation 6.1}$$

where p_i is the proportion of resources in state *i* used by a population and q_i is the proportion of resources in state *i* available to the another population. Dawson *et al.* (1992) and Ellis *et al.* (1992) calculated the *PS* Index for two species of herbivore (the bridled nailtail wallaby, *Onychogalea fraenata*, and the black striped wallaby, *Macropus dorsalis*) and then compared the results for similarity, and hence dietary overlap, by using the Mantel test (Mantel 1967). However, since the *PS* index essentially acts as a comparison between two variables it can also serve as a direct measure of overlap, using the assumption that the proportion of resources (forage quality and quantity) are equally available to the NHN wombat and the EG kangaroo. Therefore if p_i is defined as the proportion of species *i* in the diet of the NHN wombat and q_i is defined as the proportion of species *i* in the diet of the EG kangaroo, an index between one and zero is generated describing overlap between the EG kangaroo and the NHN wombat rather than an index of niche breadth. A value of zero represents no overlap and a value of 1 occurs when the two herbivores have consumed identical proportions and composition of plant species.

Both the Jarman (1971) and *PS* index can only provide a descriptive measure of dietary overlap. How the two herbivores perceive and respond to the forage environment can not be quantified by either measure of overlap.

6.2.2 Population Estimates

6.2.2.1 Background

Monitoring the abundance of macropod populations has been the subject of many studies e.g. see Southwell (1989). Three techniques for measuring abundance

of macropods are commonly used. These include: mark-recapture, direct counts and indirect counts, with direct and indirect counts being more suitable for larger macropods such as the EG kangaroo.

Direct Estimates of Population Abundance

A variety of studies have used direct estimates of abundance to measure population densities of macropods (e.g. Newsome 1965, Bailey 1971, Southwell *et al.* 1995). Direct estimates of population abundance generally use 'Transect' methodology to estimate population density as defined by Burnham *et al.* (1980). Transects can be divided into two categories: line or strip transects (Southwell 1989) (Figure 6.1). This study used the line transect methodology described by Burnham *et al.* (1980) and used the same assumptions as Burnham *et al.* (1980) and Southwell (1989). Details of the technique are summarised in Figure 6.2.

Indirect Estimates of Population Abundance

Indirect estimates of population abundance do not rely on sighting the subject but record its presence through faecal pellets, tracks, dens, scent posts, etc. (Southwell 1989). Faecal pellets have been used to count macropods and record spatial distribution in a number of studies (e.g. Caughley 1964, Hill 1982). Density measurements by indirect counts measure the accumulation of faecal pellets per unit time (Southwell 1989) and assume that the rate of defecation and decay is known; and there is no loss of sample unit (faecal pellets or pellet groups) over the period between samples. These assumptions are not easily validated without extensive observations of the animal's behaviour (e.g. Johnson and Jarman 1987; Johnson *et al.* 1987) and careful monitoring of the fate of faecal material. Nevertheless the indirect estimates of population abundance can provide accurate measures of the densities of macropod populations (e.g. Perry and Braysher 1986, Southwell *et al.* 1995).The main advantage of the indirect estimates of population abundance is that it is independent of observing the subject, which is important for heterogenous habitats such as EFNP.

6.2.2.2 Field Application and Analysis

Direct Estimates of Population Abundance

Direct counts were conducted in the hour preceding sunset by an observer travelling in a vehicle. The vehicle followed 6.6 km of fire track passing through NHN wombat habitat. Two observers recorded the distance travelled, angle to the macropod ('A' in Figure 6.2) and distance to the macropod from the vehicle ('X' in Figure 6.2). Additional information was recorded including an estimate of cloud cover, commencement and finishing times of the survey, the side of the vehicle (passenger's or driver's side), macropod species (EG kangaroo or swamp wallaby), and an estimate of the age and sex of the macropod.

Data analysis followed the protocol of Burnham et al. (1980). However, because of the low numbers of EG kangaroos observed, an effective strip width was needed to estimate density. Using 185 observations of EG kangaroos, a frequency histogram found that an effective strip of 180 metres included 96% of all sightings. Therefore, the area effectively surveyed was 118.8ha.

The direct count methodology in this study unavoidably violated a number of assumptions stated by Burnham *et al.* (1980). These were:

- the transect was not randomly located - it followed a pre-determined route (fire track);
- female macropod behaviour is gregarious and several individuals may aggregate together (Southwell 1984, Jarman 1987);
- distances were estimates; and creation of an effective strip altered methods used to calculate density.

Despite this, the estimates of the population size of EG kangaroos at EFNP appeared reliable, with population estimates comparing favourably with those measured by the indirect method.

Indirect Estimates of Population Abundance

Indirect estimates of population abundances were undertaken along the seven BOTANAL transects (Figure 2.3). Each 400 metre BOTANAL transect was marked every 50 metres with a 1.5 metre wooden stake (eight stakes per transect). Transect seven (9 stakes) and transect three (7 stakes) were the exception. At each

Figure 6.1. Three types of Transects from Southwell (1989). Closed circles represent animals recorded, open circles animals not recorded (or seen in line transect).

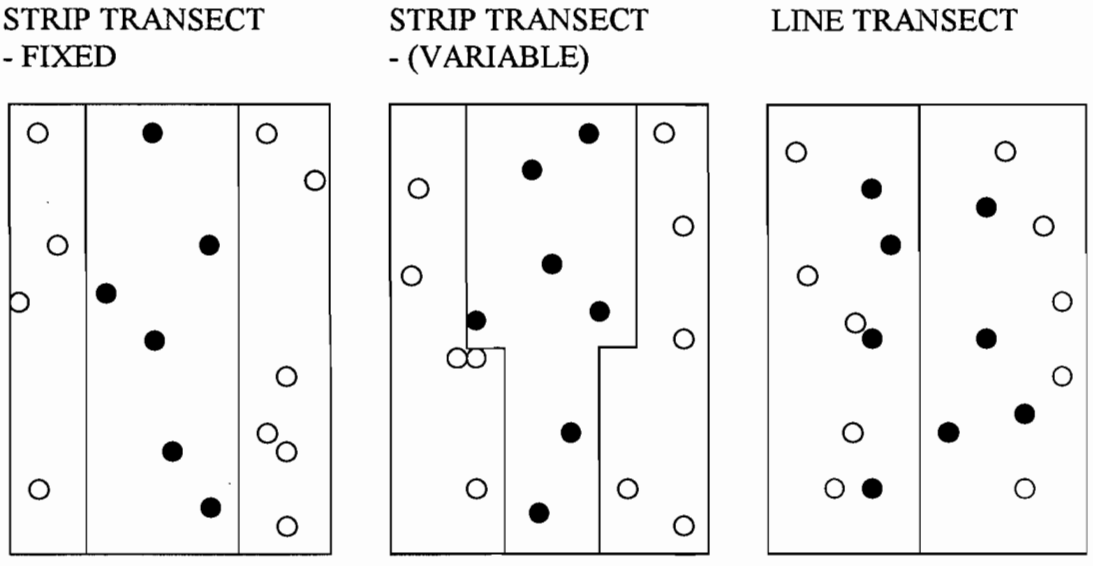
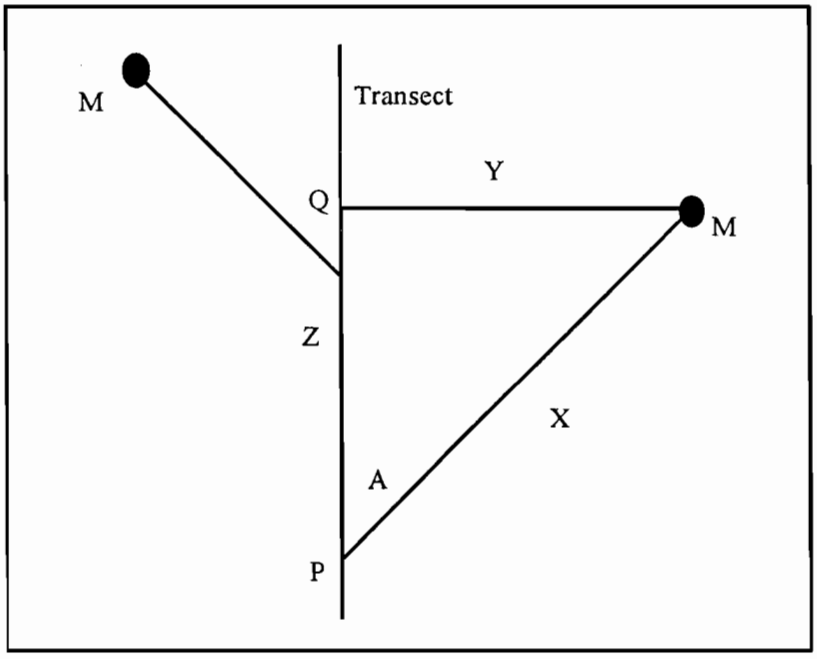


Figure 6.2. Diagram representing line transect measurements. The closed circles represent the detected object (M: Macropod); P is the position of the observer; X is the distance from the observer to the macropod; A is the angle of the macropod from the transect; Q is the point on the transect perpendicular to the macropod; Y is the distance from Q to the macropod; Z is the distance from P to Q. The observer is travelling from P to Q.



stake, a three metre piece of rope was looped over the stake allowing the observer to move in a circle with a radius of three metres. All pellets accumulated between collection periods were counted, collected for faecal analysis if recently deposited, or removed from the area inside the circle. Pellet group size was also recorded. The indirect survey technique intensely samples only 0.16 ha but is representative of a much larger effective area (Southwell 1989).

The indirect estimation of populations size relies on the faecal measurements of Johnson *et al.* (1987) to calculate the density of EG kangaroos (in units of animals/km^2/day). These measurements are the rate of defecation (89.5 defecation events/24 hours) and the number of pellets per defecation (5.51). By using these values the indirect count is violating the assumption that the rate of defecation is known. Johnson *et al.* (1987) measured EG kangaroo defecation patterns in an area of northern New South Wales, known as Wallaby Creek, described in detail by Jarman *et al.* (1987). It is likely that the EG kangaroo consumes a diet higher in fibre at EFNP than Wallaby Creek. This would therefore increase defecation rates and may lead to an overestimation of the EG population at EFNP.

The rate of decay and loss of faecal pellets were examined by trials. Decay rates are very low, and the time interval between sampling was less than the rate of decay. However, loss of pellets through trampling or movement by wind can occur which may result in an under-estimation of the population size of EG kangaroos at EFNP.

One-way ANOVA was used to examine any differences between the two techniques at estimating the population density of EG kangaroos. One-way ANOVA was also used to examine any difference in the densities of the EG kangaroo population with time. Correlation analysis also examined the relationships between the EG kangaroo population and rainfall, pasture biomass and primary productivity. The effect of time was examined by calculating the correlation coefficient for the population density at time t and the factor (rainfall, biomass or primary productivity) at time t-1 in a time-series.

6.2.3 Spatial Interactions

The habitat of the EFNP is characterised by a diversity of vegetation structure on differing soil types (described in Chapter 2). Northern hairy-nosed wombats are directly associated with the sandy soil of the ancient streambed, in which their burrows are constructed, and open grasslands (Steinbeck 1994). Eastern grey kangaroos do not depend on burrows for shelter and are comparatively more mobile than the sedentary NHN wombat so the potential habitat available to macropods is larger. It follows that EG kangaroo may actively 'seek out' the most suitable habitat with changing conditions in the forage environment.

Direct observation of interactions between the NHN wombat and the EG kangaroo was not possible because of the vegetation structure, the nocturnal behaviour of NHN wombats, the interference of an observer on the behaviour of both EG kangaroos and NHN wombats, and the logistics of such a time-consuming exercise. Instead, an indirect approach, using faecal pellets of both species, was incorporated into the design of the BOTANAL methodology (Chapter 2). The distribution of faecal pellets, measured by BOTANAL, for both herbivores allowed investigation of the spatial separation between the NHN wombat and the EG kangaroo, and correlations between NHN wombats and EG kangaroos with environmental variables.

The BOTANAL methodology recorded the presence or absence of NHN wombat and/or EG kangaroo faecal pellets in the interval between quadrats. This was used as an index of density for NHN wombats and/or EG kangaroos along transects, and subsequently spatial separation and habitat correlation. The index of density for the EG kangaroo and the NHN wombat was calculated as the number of intervals between quadrats where faecal pellets were present (maximum of 80 observations) divided by 80, to provide a measure of density of the EG kangaroo and the NHN wombat per transect per unit time. The relationships between the EG kangaroo and NHN wombat density indices and 'environmental factors' were examined using multiple-linear regression (MLR) techniques.

Attributes considered as 'environmental factors' included the composition, frequency and biomass of plant species and ground cover. Exploratory statistics (including principal component analysis and MLR) refined the list of 'environmental factors' to include: species composition and species frequency for *Aristida* spp.; *Enneapogon* spp; *Chrysopogon fallax*; *Fimbristylis dichotoma*; *Cenchrus ciliaris*; other grasses; other forbs; and ground cover.

The 'environmental factor' data was bimodal. All data were therefore transformed using the following transformation: $\sqrt{\text{variable}} + 0.5$. All transformed environmental factors were selected used in an exploratory MLR to identify the significant variables. The significant variables were then used in a second MLR, where any outlying data points identified in the first MLR eliminated.

6.3 RESULTS

6.3.1 The Diet of the Eastern Grey Kangaroo

6.3.1.1 Botanical Composition

Stable carbon isotope analysis revealed that the EG kangaroo diet was dominated by C_4 plants (Figure 3.1). This is supported by the Vanderploeg and Scavia (1979) Relativised Electivity Index (described in Chapter 3) measuring forage selection by the EG kangaroo (Figure 3.2b). The species composition of the EG kangaroo diet was dominated by *Aristida* spp., *Enneapogon* spp., *Cenchrus ciliaris*, *Chrysopogon fallax* and *Fimbristylis dichotoma* depending on whether conventional histological analysis (Table 6.1) or the three variations of the long-chain alkane analyses were used to measure diet (Table 6.2).

The diet of the EG kangaroo, determined by histological analysis (Table 6.1), does not exhibit any significant changes with time (ANOVA $F_{2,200} = 0.032$; $p = 1.00$). Again, the validity of the histological technique was assessed by comparing the botanical composition measured by two independent observers for the October 1993 sampling period. There was no difference between the two observers and their estimated diet for EG kangaroos (ANOVA $F_{1,50} = 0.13$; $p = 0.738$; Table 6.1).

Table 6.1. Temporal trends in diet composition of the eastern grey kangaroo over 12 months determined by conventional microscopic analysis with a comparison to the mean NHN wombat. Diets expressed as a percentage. October 1993 compares the findings of two independent observers.

Plant Species	May-93	Jul-93	Sep-93	Oct-93 Observer 1	Oct-93 Observer 2	Nov-93	Mar-94	Apr-94	May-94	Mean Kangaroo Diet	S.D.	Mean Wombat Diet	S.D.
Fimbristylis dichotoma	0.41	0	0.41	0	0	0	0	0	0.27	0.12	0.10	1.85	1.63
Aristida spp.	16.42	21.06	29.46	19.09	7.23	21.73	30.69	24.61	27.69	22.00	7.32	20.64	5.95
Bothriochloa ewartina	0	0	0	0	0	0	0	0	0	0.0	0.0	0.0	0.02
Brachiaria spp.	0	0	0	0	0	0	0	0	0	0.0	0.0	0.01	0.03
Cenchrus ciliaris	18.8	18.57	14.24	17.07	13.15	16.3	5.94	15.29	19.94	15.47	4.2	14.46	5.49
Chrysopogon fallax	0	0	0	0	0.	0	0	0	0	0.0	0.0	0.04	0.09
Cymbopogon refractus	0	0	0	0	0	0	3.46	0	0	0.38	1.1	0.0	0.0
Dactlyoctenium radulans	0	0	0	0	0	0	0.49	0.13	0	0.06	0.1	0.0	0.0
Enneapogon spp.	20.46	19.99	14.04	19.3	21.71	22.81	25.73	24.06	24.91	21.45	3.56	17.21	6.28
Enteropogon ramosus	0	0	0	0	0	0	0.49	0	0	0.05	0.1	0.12	0.22
Eragostris lacunaria	0	0	0	0	0	0	0	0	0	0.0	0.0	0.03	0.07
Eragostris schultzii	0	0	0	0	0	0	0	0	0	0.0	0.0	0.04	0.08
Heteropogon contortus	0	0	0	0	0	0	0	0	0	0.01	0.0	0.0	0.0
Perotis rara	0	0	0.13	0	0	0	0	0	0	0.0	0.0	0.02	0.06
Themeda triandra	0	1.07	0	0	0.65	0	3.46	1.62	0.27	0.78	1.1	0.01	0.02
Tragus australianus	0	0	0	0	0	0	0	0	0	0.0	0.0	0.02	0.08
Triraphis mollis	0	0	0	0	0	0	0	0	0	0.0	0.0	0.01	0.04
Carissa ovata	0	0	0.27	0	0	0	0	0.13	0	0.04	0.0	0.10	0.15
Helichrysum sp.	0	0	0	0	0	0	0	0	0	0.0	0.0	0.15	0.35
Cleome viscosa	0	0	0	0	0	0	0	0	0	0.0	0.0	0.06	0.09
Salsola kalii	0	0	0	0	0	0	0	0.13	0	0.01	0.0	0.29	0.58
Evolulus alsinoides	0	0	0	0	0	0	2.47	1.48	0	0.43	0.9	1.85	2.92
Abutilon sp.	0	0	0.13	0	0	0	0	0	0	0.01	0.0	0.12	0.35
Sida sp.	0	0	0	0	0.65	0	0	0	0	0.07	0.2	0.14	0.33
Waltheria indica	0.69	0.35	0.13	1.34	0	0.13	0	0.54	0	0.35	0.4	1.96	2.01
Tribulus terrestris	0	0	0	0	0	0	0.49	0	0	0.05	0.1	0.05	0.11
Unidentifiable Monocots	39.55	36.42	37.15	36.17	47.36	32.6	25.74	27.6	26.59	34.35	7.0	37.83	12.79
Unidentifiable Dicots	3.62	2.50	1.11	6.96	9.21	6.38	0.99	4.33	0.27	3.93	3.0	2.51	2.01

Table 6.2. Mean annual botanical composition of the diet of the eastern grey kangaroo by histological analysis and three variations of the least squares optimisation procedure for alkane analysis (see Chapter 3 for descriptions of the three estimation procedures). Dashes represent plant species not included in the analysis. Histological analysis incorporates twelve months of sampling with 8 collection periods. Alkane analysis incorporates thirty-three months with 15 collection periods.

Plant Species	Microscopic Analysis	Alkane		
		Estimation Procedure 1	Estimation Procedure 2	Estimation Procedure 3
Fimbristylis dichotoma	0.1	9.8	16.1	25.6
Aristida spp.	22.0	12.5	25.4	17.2
Bothriochloa ewartina	0.0	0.3	0.0	-
Brachiaria spp.	0.0	7.6	10.0	-
Cenchrus ciliaris	15.5	4.0	9.4	13.0
Chrysopogon fallax	0.0	13.7	9.8	23.5
Cymbopogon refractus	0.4	-	-	-
Dactlyoctenium radulans	0.1	3.9	-	-
Enneapogon spp.	21.5	19.1	17.9	15.7
Enteropogon ramosus	0.1	-	-	-
Eragostris lacunaria	0.0	9.5	2.9	5.0
Eragostris schultzii	0.0	-	-	-
Heteropogon contortus	0.0	2.3	0.8	-
Perotis rara	0.0	1.5	0.0	-
Themeda triandra	0.8	-	-	-
Tragus australianus	0.0	-	-	-
Triraphis mollis	0.0	2.2	-	-
Carissa ovata	0.0	0.0	0.0	-
Helichrysum sp.	0.0	2.5	2.1	-
Cleome viscosa	0.0	-	-	-
Salsola kalii	0.0	0.2	-	-
Evolulus alsinoides	0.4	-	-	-
Malvaceae	-	1.8	3.6	-
Abutilon sp.	0.0	4.4	0.3	-
Sida sp.	0.1	1.5	0.1	-
Waltheria indica	0.4	3.4	1.7	-
Tribulus terrestris	0.1	0.4	-	-
Unidentifiable Monocots	34.4			
Unidentifiable Dicots	3.9			

Table 6.3. Percentage contribution of plant parts of species consumed by the eastern grey kangaroo. The relative proportion of identifiable fragments represents the number of identified fragments for each species as a proportion of the total fragments identified (N = 2872).

	Leaf	Stem	Sheath	Unknown	Relative Proportion of Identifiable Fragments
Fimbristylis dichotoma	85.7	14.3	0	0	0.3
Aristida spp.	55.9	17.7	26.1	0.3	37.3
Bothriochloa ewartina	0	0	0	0	0
Brachiaria spp.	0	0	0	0	0
Cenchrus ciliaris	45.5	20.5	33.7	0.3	24.5
Chrysopogon fallax	0	0	0	0	0
Cymbopogon refractus	100	0	0	0	0.2
Dactyloctenium radulans	100	0	0	0	0.1
Enneapogon spp.	37.5	39.5	22.7	0.2	35.5
Enteropogon ramosus	0	0	0	0	0
Eragostris lacunaria	0	0	0	0	0
E. schultzii	0	0	0	0	0
Perotis rara	100	0	0	0	0
Themeda triandra	22.7	0	77.3	0	0.7
Tragus australianus	0	0	0	0	0
Triraphis mollis	0	0	0	0	0
Carissa ovata	100	0	0	0	0.1
Helichyrsum sp.	0	0	0	0	0
Cleome viscosa	0	0	0	0	0
Salsola kali	100	0	0	0	0
Evolvulus alsinoides	100	0	0	0	0.6
Abutilon sp.	0	0	0	0	0
Sida sp.	100	0	0	0	0.1
Waltheria indica	100	0	0	0	0.6
Tribulus terrestris	100	0	0	0	0
Mean Consumption	47.4	25.7	26.6	0.3	

Eastern grey kangaroos select leaf as the preferred part of the plant (Table 6.3). Personal observations of EG kangaroos feeding at EFNP confirm the selection for leaf, particularly for green leaves of *Cenchrus ciliaris*.

Although this account of the diet of the EG kangaroo at EFNP is the best possible estimated composition of the diet, the techniques used to measure the diet are influenced by the shortcomings described in Chapter 3.

6.3.1.2 Dietary Overlap of the Eastern Grey Kangaroo with the NHN Wombat

The botanical composition of the diet of the EG kangaroo and the NHN wombat were similar (Table 6.1). Both the *PS* Index and Jarman's overlap revealed that there is high total overlap of forage species consumed (Table 6.4). Mean overlap is higher than overlap during representative dry and wet season collections, suggesting that high overlap was consistent throughout the year without seasonal variability. Selection of plant parts of individual plant species by NHN wombats and EG kangaroos was almost identical. The *PS* Index and Jarman's overlap show agreement in the quantification of overlap between the EG kangaroo and the NHN wombat.

6.3.2 Population Estimates of the Eastern Grey Kangaroo

The population of EG kangaroos in the habitat of the NHN wombat was distinctively seasonal (Figure 6.3) with peak densities occurring prior to the irregular rainy season (September to December). Following wet season rains (January to March) the EG kangaroo population declined. This seasonal variation with time is statistically significant (one-way ANOVA; $F_{8,6} = 9.022$; $p = 0.008$).

The densities of the EG kangaroo population measured by the two survey methods were not statistically different (ANOVA $F_{7,7} = 3.046$, $p = 0.082$), with both methods identifying the strong seasonal variability (Figure 6.3a&b). Eastern grey kangaroo densities ranged from 0.4 ± 0.6 animals/km^2 (May 1994) to 16.9 ± 12.3 animals/km^2 (November 1995) measured by the direct survey method and 2.3 macropods/km^2/day (January 1994) to 13.6 macropods/km^2/day (October 1993) by the indirect method.

Figure 6.3. Bimonthly trends in eastern grey kangaroo density measured by two survey techniques (see text for explanations). Survey methods are:
a) direct counts and
b) indirect counts.
Surveys were not conducted in March/April 1996 using either method; Direct counts were not conducted in January/February 1994. Indirect counts began in Sepember/ October 1993 although no indirect counts were conducted in November/December 1993.

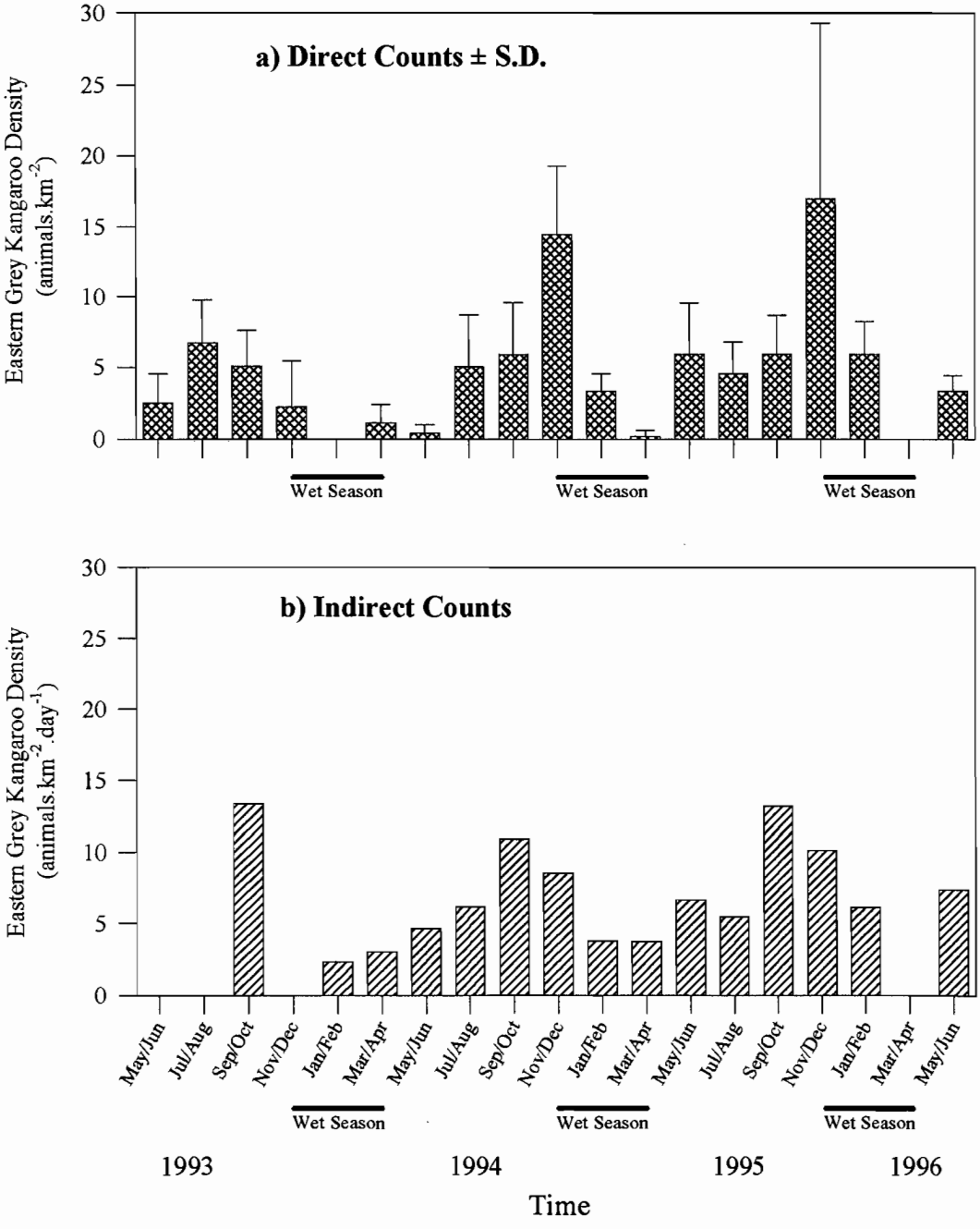


Table 6.4. Dietary overlap between the eastern grey kangaroo and the northern hairy-nosed wombat measured by the Proportional Similarity (*PS*) Index of Feinsinger *et al.* (1981) and Jarman's (1971) measure of overlap (see text for definitions). Indices of overlap can range from zero (no overlap) to one (complete overlap). Histological analysis is used as the measure of diet, with dry season data represented by September 1993; wet season by March 1994; mean by all months sampled ± one standard deviation. *N* represents the number of species/plant parts included in the measure of overlap.

Category	Time	*PS* Index	Jarman's Overlap	*N*
Plant Species	Dry Season	0.88	0.86	28
	Wet Season	0.85	0.85	28
	Mean	0.87 ± 0.04	0.85 ± 0.03	28
Plant Parts	Mean	0.99	0.95	4

The age structure of the EG kangaroo population in NHN wombat habitat was seasonal (Figure 6.4) due to the seasonal trends of the population (Figure 6.3). In general, the population consisted largely of females with young at foot, females with no pouch young, males or females that could not be distinguished, and males of medium body size. The presence of large males was seasonal, occurring prior to the wet season but only in low numbers, reflecting the hierarchy of the EG kangaroo social system. Overall, the EG kangaroo population was stable and appears to be breeding successfully.

Eastern grey kangaroo densities correlated with rainfall, total forage biomass and primary productivity (Figure 6.5). Maximum positive correlation of EG kangaroo density and rainfall occurs two months following rainfall. This trend is repeated for primary productivity, with Chapter 2 demonstrating the relationship between rainfall and primary productivity. Maximum correlation between total

Figure 6.4. Age structure of the Eastern Grey Kangaroo population at Epping Forest National Park. Contribution to the population represents the proportion of individuals in each age class for each direct survey and are expressed as a percentage. Direct counts of eastern grey kangaroos were not conducted in January/February 1994 or March/April 1996. 'N' is the total number of observations for each age class.

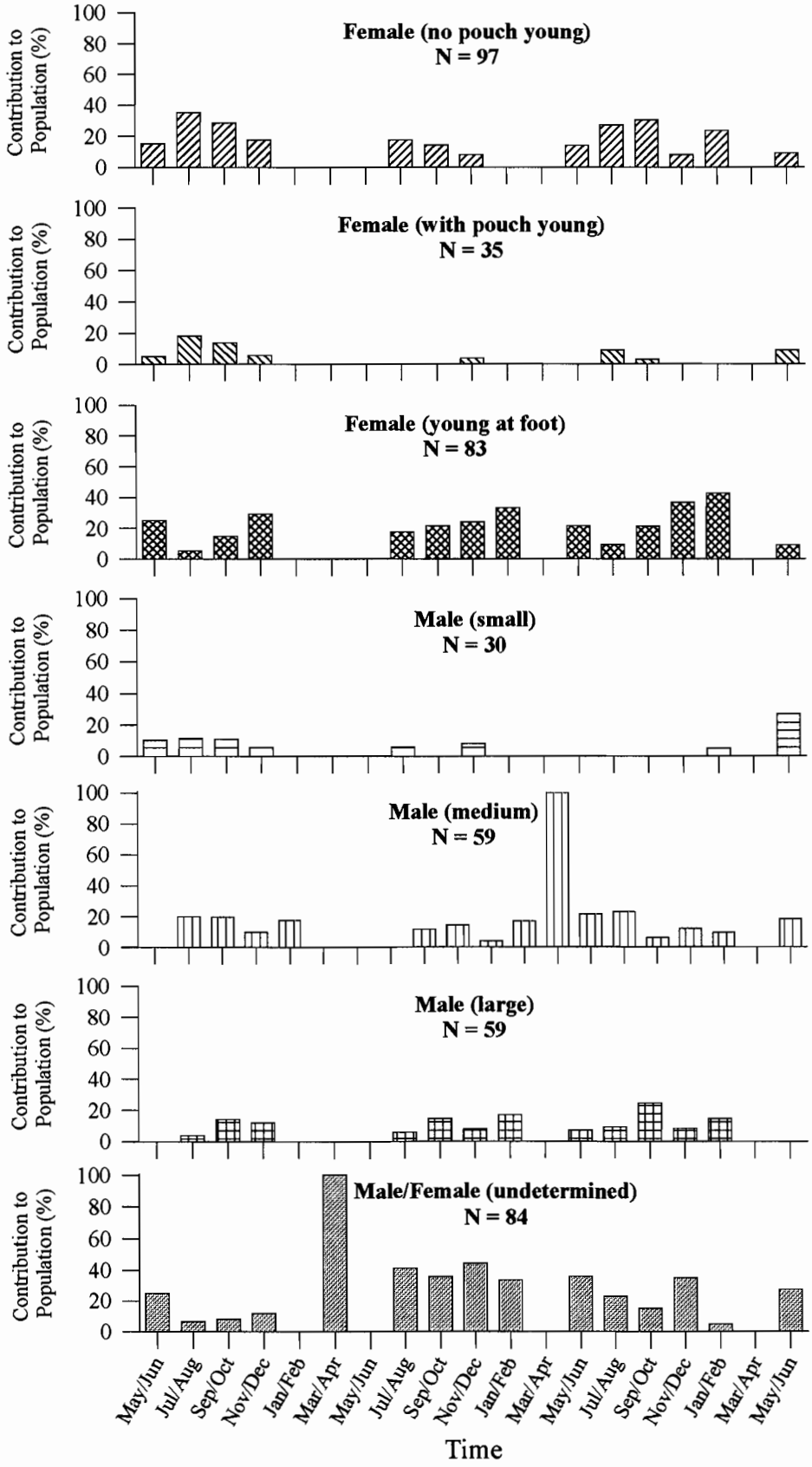
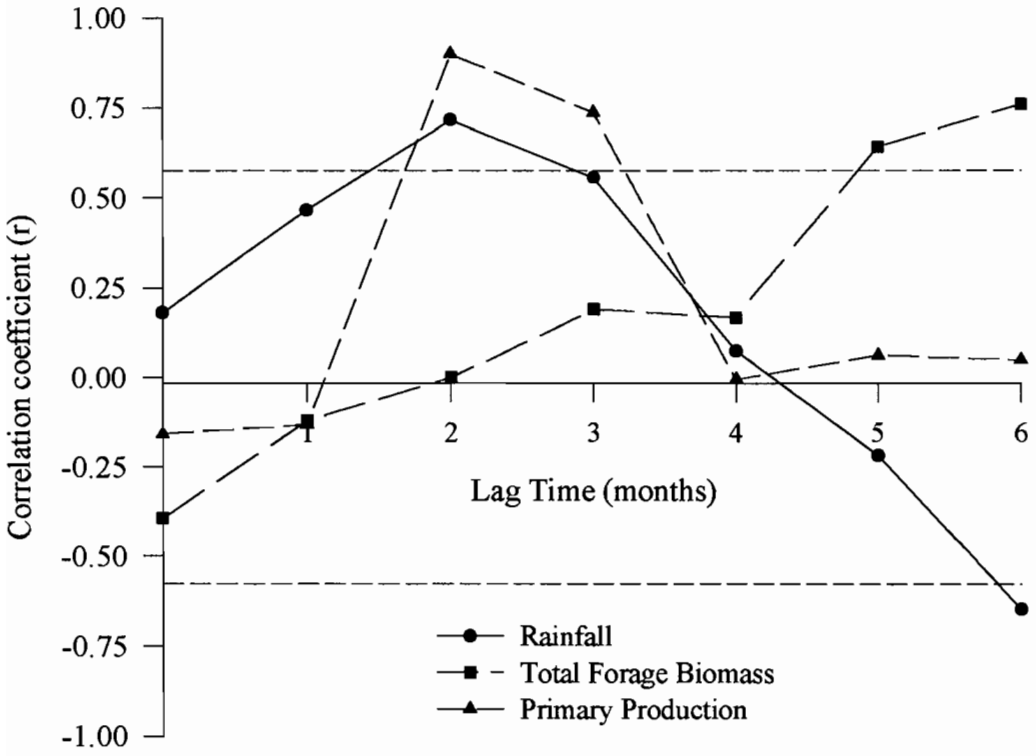


Figure 6.5. Correlation coefficients (r) of the densities of eastern grey kangaroos over a lagged time series of non-accumulative rainfall, total forage biomass and primary productivity. [r = 0.576; p < 0.05; 10 df]



forage biomass and EG kangaroo densities occurs six months following peak biomass levels. This is in agreement with seasonal trends in EG kangaroo densities in Figure 6.3.

6.3.3 Spatial Interactions Between Wombats and Kangaroos

The spatial separation of NHN wombats and EG kangaroos can only be implied indirectly by the presence of faecal pellets. Although Johnson *et al.* (1987) demonstrated that EG kangaroos defecate where they feed, the presence of NHN wombat faecal pellets may indicate a location of social importance rather than evidence of feeding activity because wombats use faecal deposits for scent-marking (Triggs 1988). Hence, the results pertaining to NHN wombats can only be regarded as speculative and illustrative of trends.

6.3.3.1 Spatial Separation of Wombats and Kangaroos

Figure 6.6 and Figure 6.7 show the relative density of faecal pellets along the seven BOTANAL transects for EG kangaroos and NHN wombats respectively. When comparing the two figures it becomes clear that when the relative density of NHN wombat faecal pellets is high (e.g. Transect 1, distance 400 metres), the relative density of EG kangaroo faecal pellets is low. The presence of NHN wombat faeces is usually associated with NHN wombat burrows or feeding trails. Conversely, the absence of EG kangaroo faecal pellets is indicative that levels of foraging by the EG kangaroo is low. It seems likely, therefore, that EG kangaroos do not feed in the vicinity of NHN wombat burrows, so at least some spatial separation between the two herbivores does occur. However, the home range of the NHN wombat extends beyond the vicinity of the burrow (Chapter 5) and overlap of forage resources with the EG kangaroo may occur.

6.3.3.2 Environmental Associations

Eastern Grey Kangaroos

Multiple linear regression indicated that the distribution of the EG kangaroo was influenced by ($F_{6,97} = 14.98$; $p = 0.000$; $r^2 = 0.481$) (Table 6.5):

- the composition of *Enneapogon* spp. (as a proportion of the total forage biomass),
- the composition of other grasses (as a proportion of the total forage biomass),

Figure 6.6. Relative density of kangaroos along the seven BOTANAL transects as measured by the distribution of kangaroo faecal pellets.

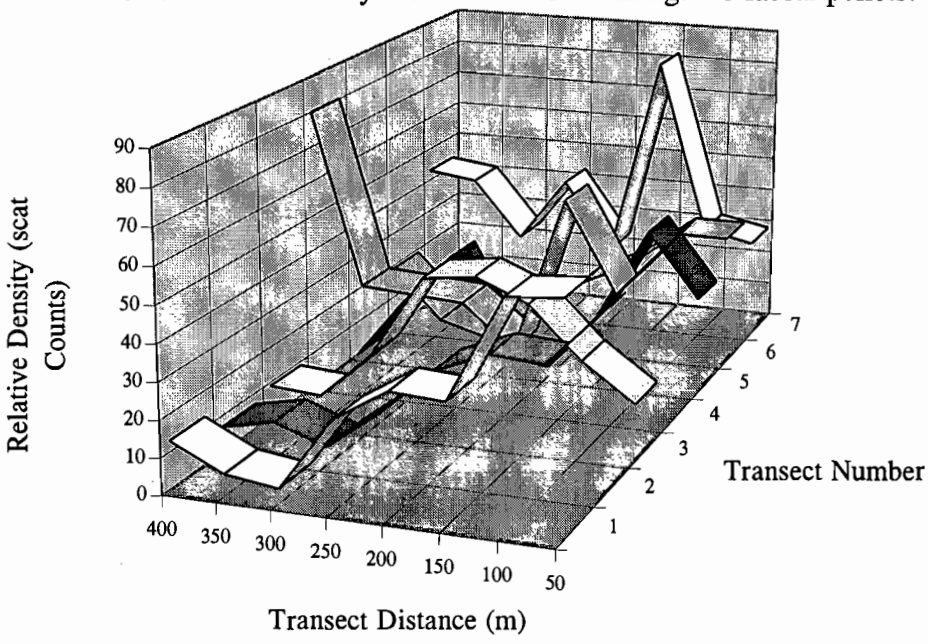
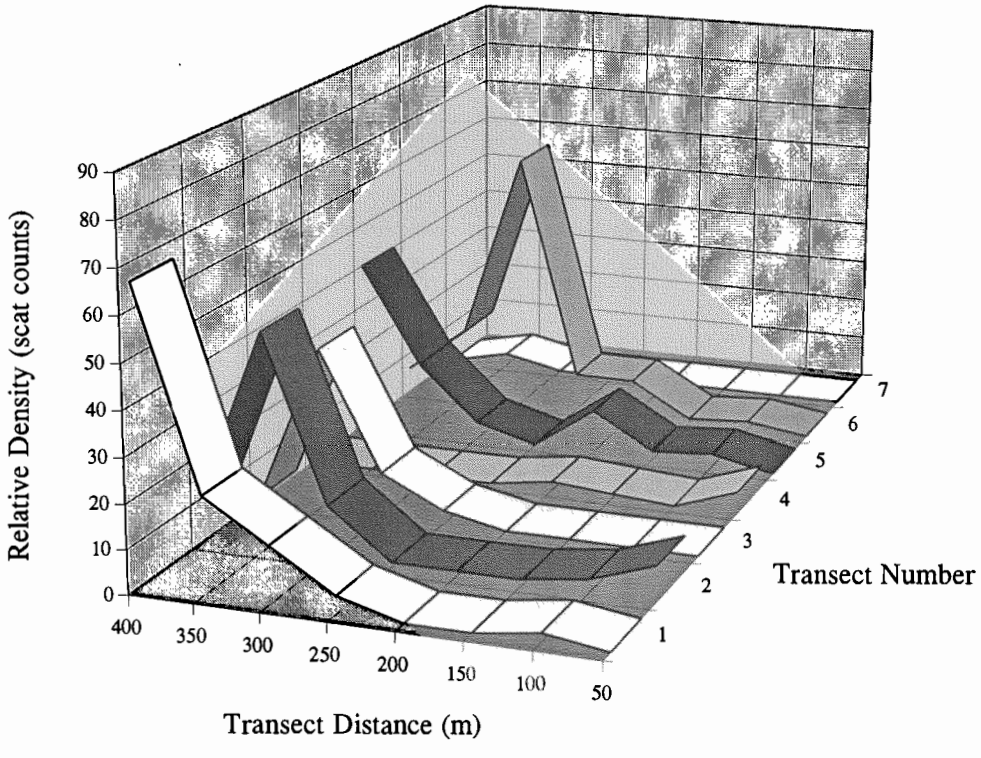


Figure 6.7. Relative density of wombats along the seven BOTANAL transects as measured by the distribution of wombat faecal pellets.



- the frequency of occurrence of *Chrysopogon fallax*,
- the frequency of occurrence of *Fimbristylis dichotoma*,
- the frequency of occurrence of other grasses,
- and ground cover.

The relationship was positive for composition of *Enneapogon* spp., frequency of *C. fallax* and frequency of other grasses, and negative for composition of other grasses, frequency of *F. dichotoma*, and ground cover. This indicated that where there was a high biomass of *Enneapogon* spp., high frequency of *C. fallax* and high frequency of other grasses; low biomass of other grasses, low frequency of *F. dichotoma* and low ground cover, the density of EG kangaroos (measured via the presence of faecal pellets) was likely to be high.

Table 6.5. Multiple linear regression model of kangaroo faecal pellet associations (kangaroo density index) with environmental variables including plant species composition and frequency, and ground cover ($F_{6,97} = 14.98$; $p = 0.000$; $r^2 = 0.481$).

Variable	Estimate	S.E. of Estimate	*t*-value	Significance
Enneapogon spp. Composition	0.52	0.18	2.86	0.005
Other Grasses Composition	-0.86	0.16	-5.96	0.000
Chrysopogon sp. Frequency	0.40	0.06	6.95	0.000
Fimbristylis sp. Frequency	-0.22	0.09	-2.45	0.016
Other Grasses Frequency	0.34	0.09	3.75	0.000
Ground Cover	-0.84	0.30	-2.83	0.006
Constant	10.46	2.65	3.95	0.000

NHN wombats

Multiple regression indicated that the distribution of the NHN wombat was influenced by the following variables ($F_{4,107} = 8.327$; $p = 0.000$; $r^2 = 0.237$) (Table 6.6):

- the composition of *Cenchrus ciliaris* (as a proportion of the total forage biomass),
- the composition of other grasses (as a proportion of the total forage biomass),
- the frequency of occurrence of *F. dichotoma*, and
- the frequency of occurrence of other grasses.

However, the low value of the coefficient of determination indicated that this relationship was negligible.

Table 6.6. Multiple linear regression model of wombat faecal pellet associations (wombat density index) with environmental variables including plant species composition and frequency, and ground cover ($F_{4,107} = 8.327$; $p = 0.000$; $r^2 = 0.237$).

Variable	Estimate	S.E. of Estimate	*t*-value	Significance
Cenchrus sp. Composition	0.76	0.24	3.19	0.002
Other Grasses Composition	0.93	0.17	5.52	0.000
Fimbristylis sp. Frequency	0.19	0.07	2.86	0.005
Other Grasses Frequency	-0.18	0.07	-2.59	0.012
Constant	-6.61	2.51	-2.64	0.010

6.4 DISCUSSION

6.4.1 Resource Overlap: The Diet of the Eastern Grey Kangaroo

Diet Composition of the Eastern Grey Kangaroo

The diet of the EG kangaroo at EFNP consists primarily of monocotyledons (Tables 6.1 and 6.2) with the dominant plant species consumed including *Aristida* spp., *Enneapogon* spp., *Cenchrus ciliaris*, *Chrysopogon fallax* and *Fimbristylis dichotoma*. Little temporal variation occurred in the botanical composition of the EG kangaroo diet and the design of sampling was such that little comment could be made on variation between individuals.

In comparison to smaller grazing macropod species (e.g. the black striped wallaby) the number of species comprising the bulk of the diet was similar but the types and parts of the grasses consumed are different (Ellis *et al.* 1992). Ellis *et al.* (1992) found that the black striped wallaby favoured young leaves whereas cattle consumed species higher in fibre such as *Aristida* spp. and *Heteropogon contortus* and a comparatively higher proportion of stem compared to leaf. The EG kangaroo at EFNP appears to forage in a manner intermediate between the smaller black striped wallaby (6.5 kg to 16 kg) and cattle, as it forages on species of high fibre concentration but favours the leaf portion (Table 6.3).

Dietary Overlap

Overlap of forage plants consumed by the EG kangaroo and the NHN wombat was high, both in botanical composition and plant part (Table 6.4). This overlap was consistent throughout the year, and displayed limited seasonal variation. The two methods of estimating overlap provided similar estimates, indicating that the diets of both herbivore species are almost identical.

In a comparison of Southern Hairy-nosed (SHN) wombats and western grey kangaroos (*Macropus fuliginosus*) foraging sympatrically in grassland, Dierenfeld (1984) found that western grey kangaroos foraged on a group of plants that were chemically and spatially different to SHN wombats. In the situation described by Dierenfeld (1984), environmental conditions were extreme and resources scarce through drought. Differences in the composition of the diet of each herbivore was attributed to the western grey kangaroo's inability to physically or physiologically switch to alternate resources such as comparatively unpalatable annual plant species. Overall, Dierenfeld (1984) concluded that the SHN wombat and western grey kangaroo utilised different forage types but both animals probably increased retention times to compensate for low intakes when food was limited. When food was not limited, both herbivores could increase their rates of intake and passage. The manner in which the western grey kangaroo and SHN wombat utilise the forage resources for nutrition during drought lead to a competitive advantage over other herbivores, such as the red kangaroo (*Macropus*

rufus) and the rabbit (*Oryctolagus cuniculus*), that behaviourally respond (e.g. through migration) to drought (Dierenfeld 1984).

The findings of Dierenfeld (1984) illustrate that when forage resources become limiting, both SHN wombats and western grey kangaroos will alter rates of intake. However, Chapter 2 has demonstrated that the forage environment of EFNP is not limiting in quantity, and the impact of vertebrate grazers on primary productivity is minimal. In addition, Chapter 3 and Table 6.2 demonstrated that neither the NHN wombat nor the EG kangaroo exhibit temporal variation in forage species consumed, although there is a high overlap of forage species consumed (Table 6.4). Consequently, during the course of this study it seems likely that both herbivores could maintain nutritional balance by maintaining a high forage intake, with the EFNP habitat able to support the grazing pressure of both herbivores without resources becoming limiting. However, the further effects of drought, management actions, and/or population expansions of grazing herbivores (invertebrate grazers, NHN wombats and/or EG kangaroos) at EFNP may alter the *status quo* thereby increasing the potential for resource competition.

6.4.2 Eastern Grey Kangaroos - Movements in Time and Space

6.4.2.1 Eastern Grey Kangaroo Population Dynamics

The EG kangaroo population densities in NHN wombat habitat are distinctively seasonal (Figure 6.3). Peak EG kangaroo densities occur between September and December, prior to wet season rains. There are two possible explanations for the seasonal variation in EG kangaroo densities. The first is that the seasonal trends may be an artefact of the survey methods. During the dry season, the biomass and height of the sward decrease with time which may lead to an increase in sightability of EG kangaroos. Conversely, following wet season rains (February to March), primary productivity and forage biomass increase reducing the probability of seeing EG kangaroos. Although this explanation seems feasible for the direct survey method, seasonal trends are also evident using the indirect method where the survey design is independent of sighting EG kangaroos. Some seasonal variability in the indirect method could be attributed to seasonal changes in the fibre concentration in the diet and hence defecation rates. However, changes

in rates of defecation are unlikely to significantly influence the density measurement.

The second, and more likely, explanation for seasonal variation in densities of EG kangaroos is that it is a function of the ranging behaviour of the EG kangaroo and the influence of changing forage resources beyond the EFNP boundaries. The major landuse on the properties surrounding EFNP district is pastoralism, specifically cattle grazing. Practices to improve the carrying capacity of cattle on pastoral properties have resulted in land clearing to promote the growth of grasses, the introduction of permanent watering points, and the limited control of predators such as dingoes. These land management practices create a foraging environment favourable to domestic stock but also to the larger kangaroo species (Robertson *et al.* 1987).

Since the study was conducted in a four year period of below average rainfall and, vertebrate grazing pressure on the surrounding land is considerably higher than for EFNP (E. Anderson, Queensland Department of Primary Industries, pers. comm.), the dynamics of the forage environment surrounding EFNP are more extreme (personal observations). Following summer rainfall (November to March) forage conditions beyond EFNP were potentially more favourable because of the relatively low proportion of non-growing biomass and higher proportion of new growth as a result of high levels of cattle grazing. At this time, EG kangaroo populations on EFNP were at a minimum with only animals judged to be resident animals (by distinctive facial characteristics) present in the population (pers. obs.). As the dry season progressed, and grazing pressure of cattle reduced forage availability in pastures beyond the EFNP boundary, the densities of EG kangaroos on EFNP increased, peaking prior to summer rainfall. This indicates that the foraging activities of EG kangaroos change in response to variation in their foraging environment. The second explanation is supported by the strong positive correlation of EG kangaroo population densities six months following peak values of forage biomass (Figure 6.5).

Additional support for the second explanation comes from a study by Hill (1982), who found that use of habitat by EG kangaroos was not constant but

followed a cyclic pattern, with EG kangaroos responding directly to seasonal changes in the environment. Hill (1982) concluded that the major factor influencing the distribution of EG kangaroos was changes in forage quality and quantity. Eastern grey kangaroos at EFNP appear to be equally sensitive to variation in forage quality and quantity, but over a significantly larger range than just NHN wombat habitat.

Accuracy of eastern grey kangaroo population estimates

Measurements and conclusions regarding the dynamics of EG kangaroos within NHN wombat habitat rely heavily on the ability of techniques used to measure EG kangaroo abundance. In a comprehensive review of the methodologies used to measure macropod abundance Southwell (1989) identified survey design, precision and sampling intensity, repeatability, and accuracy as the key considerations in assessing the suitability of each methodology. Assessing these considerations in the application of both direct and indirect EG kangaroo surveys (Table 6.7) reveals that while both techniques have their fundamental faults, their overall precision for estimating EG kangaroo abundance is comparable.

While both survey techniques provide comparable estimates of EG kangaroo density at EFNP, the issue of scale is paramount in the application of the appropriate survey technique in other situations. Southwell (1989) emphasises that although indirect counts are suitable for small areas (e.g. Perry and Braysher 1986, Johnson and Jarman 1987) broad scale surveys should use direct counts. From a management perspective for EFNP, the indirect survey method is probably more useful primarily because it can be done by untrained observers and is independent of weather conditions or human activity on the park.

6.4.2.2 Habitat Usage - A Comparison with the NHN Wombat

The distribution of EG kangaroos, inferred by faecal pellet distribution, is associated with a combination of habitat parameters. Habitat characteristics EG kangaroos positively select for include areas with high *Enneapogon* spp. composition, high frequency of *Chrysopogon fallax* and a diverse frequency of other grasses (not including *Aristida* spp. or *Cenchrus ciliaris*), with EG kangaroos avoiding areas of high ground cover, high frequency of *Fimbristylis*

Table 6.7. Major advantages and disadvantages of the two survey methods used to estimate the abundance of eastern grey kangaroos at Epping Forest National Park.

Consideration	Direct Survey		Indirect Survey	
	Advantages	Disadvantages	Advantages	Disadvantage
Survey Design	• samples entire wombat habitat passing along the sandy ancient water coarse • minimal time per individual survey	• follows a track (non random) • lack of replication • potential interference from vehicle noise • many surveys needed to provide accurate estimate	• random stratified design • sampling whole wombat habitat • uses established BOTANAL transects • kangaroos do not need to be sighted; weather/disturbance variables eliminated • 3 m circular design is time efficient • observer training is minimal	• samples a comparatively smaller area • uses individual pellets rather than pellet groups • uses assumption that pellet groups represent individual animals • heterogeneous habitat, with kangaroos showing preference for vegetation types • surveys need to be conducted regularly
Precision and Sampling Intensity	• measurement of error (standard deviation) • identification of age/species structure	• relatively small sampling area • low density population • inconsistent number of surveys per field trip (dependent on field trip duration)	• intensive sampling sufficient for wombat habitat • surveyed by inexperienced personnel • negligible costs • sampling can be completed in one day • high persistence of faecal pellets	• details on population structure remain unknown
Repeatability	• repeatable transect	• seasonal variability (time of day, weather conditions etc.) • observer inexperience • minimum number of surveys required	• highly repeatable • no seasonal variability	• potential for corruption of faecal pellets within counting area (eg blown in)
Accuracy	• accurate distance measurements (along transect) • individuals counted only once	• estimate of distance (to kangaroo) • habitat is heterogeneous	• comparable to direct survey method • allows spatial modelling of kangaroo habitat usage	• uses values of defecation and pellet group size from Johnson et al. (1987) • complex calculations to generate a measurement of error

dichotoma and high composition of other grasses (Table 6.5). The distribution of EG kangaroos was indifferent to areas of high or low *Cenchrus ciliaris* and *Aristida* spp. frequency, biomass and composition even though these species are major components of its diet (Table 6.1). The positive relationship which EG kangaroos have with *Chrysopogon fallax* indicate that it may be an important forage species. However, this remains only speculation for the reasons described in Chapter 3 when describing the merits of methods of diet analysis.

The defecation patterns of the EG kangaroo is likely to be a true reflection of habitat usage (Johnson *et al.* 1987). Eastern grey kangaroos usually defecate where they feed, therefore the distribution of faecal pellets is likely to have a high affinity for their forage preferences and overall habitat usage. However, some aspects of the analysis can cause a limited degree of bias, primarily through the collection of data. For example, the EG kangaroo's avoidance of areas with high ground cover may be an artefact of the collection methods combined with the foraging behaviour of the EG kangaroo. Eastern grey kangaroos are likely to feed on the fringe of a dense stand of tall grasses (e.g. *Cenchrus ciliaris*) and defecate around the periphery of the stand on areas with less ground cover (personal observation). Further, the chances of detecting and counting EG kangaroo faecal pellets are reduced when ground cover is high.

In comparison the spatial usage of habitat by the NHN wombat, inferred by the distribution of faecal pellets, is less clear. Feeding associations of NHN wombats cannot be inferred from faecal pellets because unlike the EG kangaroos they are not deposited where the NHN wombat feeds. Instead, the deposition of faecal pellets by all wombat species is a key component of their olfaction-based social system, used to mark territory and to advertise reproductive status (McIlroy 1973, Gaughwin 1981, Triggs 1988). Consequently the relationship between faecal pellet distribution and habitat parameters will not imply feeding patterns but may identify important spatial characteristics of the NHN wombat's social system. Habitat characteristics positively associated with NHN wombat faecal pellets include *Cenchrus ciliaris* composition, the composition of other grasses and the frequency of *Fimbristylis dichotoma*. These characteristics are strongly associated

with NHN wombat burrows and the immediate area surrounding NHN wombat burrows where the frequency of NHN wombat faecal pellets is highest. Areas with high *Aristida* spp. and *Enneapogon* spp. composition, frequency and biomass are characterised by absence of NHN wombat faecal pellets. However these species form the bulk of the NHN wombat's diet (Chapter 3), reinforcing the notion that the distribution of NHN wombat faecal pellets provides only limited information on the spatial use of habitat by the NHN wombat.

Overall it appears that there is some spatial overlap between the two herbivores. Figures 6.6 and 6.7 demonstrate that EG kangaroos do not feed (inferred via defecation patterns) in areas where the densities of NHN wombat faecal pellets are relatively high. These areas of high NHN wombat faecal pellet densities are characterised by the presence of NHN wombat burrows and soil disturbances in the immediate vicinity (30 metre radius) of the burrows that result in low ground cover. As such, the occurrence of forage species preferred by the EG kangaroo is low and foraging activities by the EG kangaroo are conducted beyond the immediate vicinity of NHN wombat burrows. Chapter 5 demonstrated that the foraging activity of the NHN wombat also extends beyond the immediate vicinity of NHN wombat burrows, and since the dietary overlap between the NHN wombat and the EG kangaroo is very high (Table 6.4), it appears that NHN wombats and EG kangaroos forage sympatrically.

6.4.3 Comparison of Feeding Activities

Although the diets of the EG kangaroo and the NHN wombat overlap extensively, both in botanical composition and parts of the plant consumed, feeding responses to a forage that varies in quality and quantity may differ. The NHN wombat must be considered a sedentary herbivore through its dependence on a burrow for shelter and thermoregulation. The dependence on a burrow, coupled with exceptionally low nutritional requirements and metabolic rate (Wells 1973, Barboza 1989, Barboza and Hume 1992, Barboza *et al.* 1993, Evans unpublished data), enables it to survive when forage resources are poor. In comparison, the EG kangaroo is also very efficient in surviving on nutritionally poor forages but its metabolic costs are slightly higher (Hume 1982, Dierenfeld 1984, Barboza 1989,

Barboza *et al.* 1993). Further, the EG kangaroo is a comparatively mobile herbivore. It can occupy home ranges of up to 317 ha in arid and semi-arid habitats (Jaremovic and Croft 1987) which almost eight times larger than the maximum home range of the NHN wombat (Chapter 5).

The foraging environment of the EG kangaroo extends beyond the bounds of EFNP and, unlike the NHN wombat, the EG kangaroo can respond to changes in forage on surrounding pastoral properties. Evidence for this response is the seasonal variability of EG kangaroo density in the geographic range of the NHN wombat. Eastern grey kangaroos move off the park during the wet season when forage conditions are better in surrounding pastoral properties and return to EFNP during the dry season when forage conditions are better at EFNP. Frith (1973) and Hill (1982) found that EG kangaroos selected forage that was grazed intensively by domestic stock and were attracted by new growth when it was available. This adds support to the notion that EG kangaroos feed on surrounding pastoral properties following seasonal rains.

Since EG kangaroos generally prefer to forage beyond EFNP during the wet season, this indicates that the forage quantity and/or quality at EFNP may be less than surrounding pastoral properties. Highest densities of EG kangaroos at EFNP during the dry season indicate that forage quantity and/or quality may be higher at EFNP than the surrounding pastoral properties at this stage. Although these trends are speculative and require further investigation, they do have implications for the conservation and management of the NHN wombat, particularly if reproduction is related to environmental variability.

CHAPTER 7

GENERAL DISCUSSION

7.1 THE FEEDING STRATEGY OF THE NORTHERN HAIRY-NOSED WOMBAT

This study has revealed much about the feeding ecology of the northern hairy-nosed (NHN) wombat. It has demonstrated that wombats have a suite of ecological and physiological characteristics that set them apart from all other herbivores. The feeding strategy of the NHN wombat is not constrained by diet selection or the food it eats, instead its feeding strategy aims to minimise the total energy it expends while foraging. This feeding strategy is reflected by the lack of temporal variation in the plants it consumes and the conservative number of hours per day it allocates for feeding. This strategy allows the NHN wombat to maintain consistent body composition and condition despite consuming food of poor nutritive value. The following chapter summarises the general findings of this thesis and provides an overview of the feeding strategy of the NHN wombat.

7.1.1 The NHN Wombat: A Generalist Grazer

The NHN wombat must be considered a generalist grazing herbivore, as it consumes primarily grass species from more than 25 plant genera (Chapter 3). The diet of the NHN wombat is dominated by three plant groups: *Aristida* spp., *Enneapogon* spp. and *Cenchrus ciliaris*. The diversity of plant species consumed by the NHN wombat is confirmed by the temporal variation in the inventories of plant species consumed between the current study and the previous two studies by Flößer (1986) and Crossman (1988) (Tables 3.1 & 3.8). The long-term differences in the species composition of the diet of the NHN wombat appear to reflect temporal changes in the species composition of the foraging environment (Figure

2.8) and the ability of the NHN wombat to change principal foods in response to these changes.

7.1.2 Nutritional Quality: Consequences for the NHN Wombat

All three species of wombats are unusual among grazing herbivores. First, the maximum body mass of an adult common wombat or NHN wombat is 40 kg. While this falls into the category of being a large herbivore (> 10 kg; Johnson in press), it is considerably smaller than other colonic fermenters (Table 1.4). Since the wombat must be considered small among colonic fermenters, Parra (1978) suggests that metabolic costs should be higher and the digestive capacity should be comparatively smaller than in larger colonic fermenters. This could result in a relative inability to digest diets that are high in fibre and therefore an inability to meet the relatively higher energy requirements of being of small body size (Barboza 1989). Following this reasoning, wombats should inhabit habitats where the quantity of forage is high in order to maintain high rates of intake of forage (Duncan *et al.* 1990), and/or inhabit areas of where the quality of forage is high (Demment and Van Soest 1985). This is not the case for the two species of hairy-nosed wombats (*Lasiorhinus krefftii* and *L. latifrons*). Both live in semi-arid environments where the quality and quantity of available forage are highly dependent on rainfall. In these semi-arid habitats, the quality of the forage is poor: high in fibre and low in digestibility (Table 2.11). Further, these habitats are subjected to a high frequency of drought.

The SHN wombat lives in southern Australia (Figure 1.3) and consumes grasses with a C_3 photosynthetic pathway (Dierenfeld 1984). Temperate grasses consumed by SHN wombats are generally higher in nutritional quality than the tropical grasses consumed by the NHN wombat. Temperate grasses generally do not reach the same standing biomass (of dry matter) as tropical grasses, and generally have lower concentrations of fibre during the growing season (Table 2.11). As the semi-arid grasses consumed by the SHN wombat senesce, fibre concentrations of these temperate grasses become comparable to tropical forages consumed by the NHN wombat (Table 2.11). However, the NHN wombat

consumes plants that have high concentrations of fibre throughout the year (Table 2.11).

Barboza and Hume (1992a) found that both the SHN wombat and the common wombat had capacious proximal colons, with fibre being the main substrate fermented in this region of the digestive tract. Both wombat species absorbed short-chain fatty acids (SCFA) derived from microbial digestion of this fibre (Barboza and Hume 1992). Further, SCFA may provide wombats with enough energy to meet maintenance energy requirements from senescent forage, and when combined with digestible cell contents during the growing season, enough energy to meet the additional energy costs of growth and reproduction (Barboza and Hume 1992a). Therefore, it seems likely that both the SHN wombat and the common wombat may select for plant species with highly digestible cell contents and perhaps more soluble sugars.

It follows that the NHN wombat should have similar digestive characteristics to the other two wombat species; i.e. capacious colon, similar pattern of microbial fermentation and the ability to utilise SCFA for energy when consuming forage of high fibre concentrations. Although the NHN wombat is probably able to digest a diet high in fibre like the other species of wombat, the limited seasonal variability in the fibre concentration of grasses consumed by the NHN wombat may impose some limitations to their overall energy balance. The continuously high concentrations of fibre in grass species at Epping Forest National Park (EFNP) may limit the availability of highly digestible cell contents. Therefore the NHN wombat may not have the opportunity to achieve an energy balance above that of maintenance to the detriment of growth and reproduction. While only speculation, this may offer an explanation for low rates of reproduction during this study (Horsup 1997).

Of equal significance is the difference in the concentration of total nitrogen available to the SHN wombat and the NHN wombat. The mean concentrations of total nitrogen in grasses available to the NHN wombat range from $0.76 \pm 0.69\%$ during the wet season to $0.64 \pm 0.25\%$ during the dry season. While the total nitrogen concentration of some grass species falls below this range (e.g. minimum:

0.27% *Heteropogon contortus*, December 1994), the maximum concentration of nitrogen for a grass species at Epping Forest National Park was only 2.24% (*Chrysopogon fallax*, February 1995). This is considerably less than the concentrations of total nitrogen available to the SHN wombat. For example the total nitrogen concentration of the principal forage species of the SHN wombat, *Stipa nitida*, ranged from 3.01% during the growing season to 0.91% following senescence (Barboza 1989).

Barboza *et al.* (1993) investigated the nitrogen requirements of the common wombat and the SHN wombat. They found that maintenance requirements for nitrogen were lower than any other published examples for herbivorous marsupials. Barboza *et al.* (1993) suggest that this is an adaptation that allows both wombat species to utilise forages that have low digestible protein and high concentrations of fibre where the availability of forage varies seasonally. It follows that the NHN wombat should be equally able to digest these types of forages and have an equally low nitrogen requirement for maintenance. However, it is possible that for part of the year that the NHN wombat may enter a negative nitrogen balance, particularly because the principal forage species consumed by the NHN wombat are low in total nitrogen concentration during the dry season (July to November).

Related to the low nitrogen requirement for maintenance in the common wombat and SHN wombat is their low total energy requirements which is a consequence of a low basal metabolic rate (Hume 1982, Barboza *et al.* 1993). Wells (1978b) found that the SHN wombat had a standard metabolic rate of only 64% of the value for marsupials predicted by the Dawson and Hulbert (1970) model. Barboza (1989, 1993b) found that tritiated water did not equilibrate sufficiently to measure water space or turnover in wombats in order to estimate the maintenance energy requirements for nitrogen balance. Subsequently, an estimate of maintenance energy requirements for nitrogen balance for wombats was performed successfully by Barboza *et al.* (1993) without using water dilution. However, he supported the claim of Wells (1978b) that the standard metabolic rate of wombats was "significantly lower than that of other marsupials" (Barboza *et al.*

1993) because both species of wombat had low energy requirements for maintenance while in captivity (Barboza 1989, Barboza *et al.* 1993). Current unpublished data (M. Evans, pers. comm.) confirms the low field metabolic rate of the SHN wombat and the common wombat in comparison to other grazing marsupials. In addition, Evans found that the NHN wombat had a lower field metabolic rate than both the SHN wombat and the common wombat.

The inferred high digestive efficiency, capacious colon, low nitrogen requirement and long retention time of particulate matter (Barboza 1989, Barboza and Hume 1992a, Barboza and Hume 1992b, Barboza *et al.* 1993, Barboza 1993a), combined with a low field metabolic rate (M. Evans, unpublished data) are adaptations that allow the NHN wombat to live within the foraging environment of EFNP. These adaptations negate the effect of small body size and colonic fermentation. Indeed, the low standard metabolic rate and capacious colon of each species of wombat fit well with Demment and Van Soest's (1985) relationship between gut capacity and metabolic rate. Further, the high availability of forage to the NHN wombat allows the intake rates of forage to be unlimited throughout the year, maximising the efficiency of colonic fermentation (e.g. Duncan *et al.* 1990). However, the effects of a diet that is continuously high in fibre and the potential for a negative nitrogen balance during the dry season require further investigation. As this is not possible with the NHN wombat, the experiments of Barboza (1989) could be repeated with SHN wombats but with a diet of higher fibre concentration or inferred from a detailed anatomical study of the digestive tract of the NHN wombat.

7.1.3 Physiological Response to the Forage Environment

Despite four years of below average rainfall during this study, the NHN wombats maintained consistent body composition and body condition throughout this study. This is consistent with the findings of Gaughwin *et al.* (1984) for SHN wombats that during a 12 to 14 month period of drought, there was a significant change in mean body mass but no evidence of physiological change in an index of body fat or blood chemistry. They concluded that the SHN wombat was able to

survive periods of drought through low energy expenditure and modification of foraging behaviour in response to reduced availability of forage.

The study by Gaughwin *et al.* (1984) found no evidence of an ecological role for body fat. However, this study found that there may be an ecological role for body fat in the NHN wombat. Females may rely on stores of body fat to cope with the energetically demanding role of reproduction and lactation (Chapter 4). Apart from this possible use of body fat, there was no other evidence for the use of body fat in ecological function. In general, therefore, the NHN wombat does not store energy in the form of body fat as a physiological response to distinct seasonal trends in the availability of crude protein in plants consumed. The extremely low field metabolic rate of the NHN wombat may be sufficiently low that it rarely enters a negative energy balance.

7.1.4 Behavioural Responses to the Forage Environment

While the three species of wombat are small in comparison with other colonic fermenting herbivores, they are large in comparison to other burrowing herbivores (Johnson 1997). The benefits of the semi-fossorial lifestyle in the Vombatidae are clear: environmental shelter and behavioural thermoregulation, resulting in water and energy conservation (Wells 1973, 1978b, Johnson 1997). However, the burrowing lifestyle does restrict foraging activities.

Compared to large macropod species of similar body size such as *Macropus rufus* (Dawson *et al.* 1974, Croft 1991a), *M. giganteus* (Jaremovic and Croft 1987, Norbury *et al.* 1989) and *M. robustus* (Croft 1991b), the hairy-nosed wombats cannot increase their home range to compensate for changes in forage quality and quantity because of their dependence on burrows for thermoregulation (Wells 1978b). Change in the size of the population of eastern grey kangaroos at EFNP highlights the mobility of large macropods compared to wombats. These changes in the size of the population of eastern grey kangaroos may be in response to changes in the quality and quantity of forage at EFNP and surrounding pastoral properties. Minimum densities of eastern grey kangaroos occur during the wet season, when the quality of the forage in the surrounding pastoral properties is potentially higher than EFNP.

Rather than maintain a large home range like the large macropods, the lower energy requirements of the hairy-nosed wombats may allow them to maintain a very much smaller home range (Chapter 5). The NHN wombat in particular is able to meet its forage requirements because the quantity of forage is high at EFNP throughout the year, enabling it to maintain a high rate of forage intake. If the quantity of forage at EFNP became limiting it is likely that the NHN wombat would be able to compensate by altering the time devoted to foraging in a similar manner to SHN wombats (e.g. Gaughwin *et al.* 1984).

The total time devoted to foraging by the NHN wombat is low in comparison to non-burrowing grazing mammals (Table 5.3). Again this is a direct consequence of a low field metabolic rate, low nitrogen requirements and high digestive efficiency but also a consequence of its semi-fossorial lifestyle. The amount of time the NHN wombat spends foraging is comparable to other burrowing mammals that also have low energy expenditure but are considerably smaller in body size and consume more digestible diets (e.g. the yellow bellied marmot, *Marmota flaviventris*; Armitage 1991). However burrowing mammals, including wombats, are often thermally intolerant to above-ground conditions (e.g. Wells 1978b, Brown 1984, Brown and Taylor 1984, Melcher *et al.* 1989, Melcher *et al.* 1990). Consequently, the foraging activity of burrowing mammals may be influenced by temperature as well as food availability. For the NHN wombat, foraging activities are likely to be restricted to a window of optimum thermal conditions.

7.1.5 Resource Overlap with Other Grazers. Is it an Issue?

The principal foods of the NHN wombat and the eastern grey kangaroo are very similar (Chapter 6), but whether this produces resource competition remains unknown without experimental confirmation. However, the potential for resource competition does exist and must be considered a potential threat to the NHN wombat. Therefore the control and management of the eastern grey kangaroo population at EFNP will always be an issue, despite seasonal fluctuations in their population size.

Although this study has focussed primarily on plants consumed by the NHN wombat and the eastern grey kangaroo, the influence of invertebrate grazers on the forage environment remains unknown. Superficial observations during 1994 and 1995 found that plague locusts had a major influence on the frequency of *Waltheria indica*, and readily consumed *Cenchrus ciliaris*. The impact of locusts on other forage species was not noted but the potential for decimation of all forage species by plague locusts must be considered.

Likewise, the impact of termites was judged to be minimal. This may be a gross underestimation of the importance of termites as grazers at EFNP. Termite activity was assessed by evidence of above-ground activity but subterranean activity remained unquantified. Further, the importance of termites in nutrient cycling (particularly of nitrogen) remains unknown but may be of particular relevance to the quality of grass species at EFNP.

7.2 CONCLUSION

In conclusion, the NHN wombat, as with the other two species of wombats, should be considered as an extraordinary herbivore. Collectively, the conservative foraging behaviour of the three wombat species, their suite of nutritional adaptations and low energy requirements allow them to consume forage of low quality and variable quantity. The hairy-nosed wombats, in particular, inhabit forage environments that have low carrying capacities of large herbivores.

The feeding strategy of the NHN wombat is well-suited to the seasonal variability of its habitat, with individual NHN wombats maintaining good body condition despite four years of below average rainfall. The NHN wombat is a generalist grazer, selecting grass species in a similar ratio to their occurrence and selecting leaf over the coarser stem component of the plant. However, the forage environment of EFNP is poor, and rather than move off the Park in search of better quality food like the eastern grey kangaroo, low energy requirements combined with conservative ranging allow the NHN wombat to successfully endure extremes in the quality of forage. Overall, the NHN wombat appears well-adapted to its habitat, despite its critically endangered status.

7.3 MANAGEMENT AND RESEARCH DIRECTIONS

The investigation into the feeding ecology of the NHN wombat has left unanswered many questions relating to the ecology and physiology of the NHN wombat. Those issues specific for the continued research and management of the NHN wombat are described in Appendix 5.

Many of the issues discussed in this thesis are particularly relevant to herbivores in general with further research required to clarify some of these issues. A key component of this research has been the application of various methods to herbivore research. Some of these methods were very successful (e.g. near infra-red spectrometry, Chapter 2) and should be considered best practice for the conservation and management of the NHN wombat. Other methodologies require further refinement for their application in grazing herbivore research, but have considerable potential (e.g. bioelectrical impedance analysis, Chapter 4). By further research, development and refinement of the methodologies used in this thesis, a better understanding of the ecology of herbivores will be gained.

REFERENCES

Aastveit, A.H. and Marum, P. (1993). Near-infrared reflectance spectroscopy: different strategies for local calibrations in analyses of forage quality. *Applied Spectroscopy* **47**: 463-469.

Adjei, M.B., Mislevy, P. and West, R.L. (1988). Effect of stocking rate on the location of storage carbohydrates in the stubble of tropical grasses. *Tropical Grasslands* **22**: 50-56.

Aggrey, E.K. (1982). Seasonal changes in water content and turnover in cattle, sheep and goats grazing under humid tropical conditions in Ghana. In: *Use of tritiated water in studies of production and adaptation in ruminants* (eds International Atomic Energy Agency) pp 133-142. IAEA, Vienna.

Ambrose, S.H. and DeNiro, M.J. (1986). The isotopic ecology of East African mammals. *Oecologia* **69**: 395-406.

Anderson, A.E., Bowden, D.C. and Medin, D.E. (1990). Indexing the annual fat cycle in a mule deer population. *Journal of Wildlife Management* **54**: 550-56.

Angerbjörn, A. (1986). Reproduction of Mountain hares (*Lepus timidus*) in relation to density and physical condition. *Journal of Zoology* **208**: 559-568.

Anon (1995). Standard practices for infrared, multivariate, quantitative analysis. Designation: E1655-94. American Society for Testing and Materials.

ANZECC (1995). List of threatened Australian vertebrate fauna. Australian and New Zealand Environment and Conservation Council.

Aragones, L.V. (1996). Dugongs and green turtles: grazers in the tropical seagrass ecosystem. PhD thesis, James Cook University.

Archer, M. (1984). The Australian marsupial radiation. In: *Vertebrate Zoogeography and Evolution in Australasia. (Animals in Space and Time)* (eds M. Archer and G. Clayton) pp 633-807. Carlisle: Hesperian Press.

Armitage, K.B. (1991). Social and population dynamics of yellow-bellied marmots: Results from long-term research. *Annual Review of Ecological Systematics* **22**: 379-407.

Arnold, G.W. and Dudzinski, M.L. (1969). The effect of pasture density and structure on what the grazing animal eats and on animal productivity. In: *Intensive Utilization of Pastures* (ed B.J.F. James) pp 42-48. Angus and Robertson, Sydney.

Arnold, G.W. (1985). Regulation of forage intake. In: *Bioenergetics of wild herbivores* (eds R.J. Hudson and R.G. White) pp 81-101. CRC Press Incorporated, Florida.

Arnould, J.P.Y. (1995). Indices of body condition and body composition in female Antarctic fur seals (*Arctocephalus gazella*). *Marine Mammal Science* **11**: 301-13.

Atkinson, S.N. and Ramsay, M.A. (1995). The effects of prolonged fasting of the body composition and reproductive success of female Polar Bears (*Ursus maritimus*). *Functional Ecology* **9**: 559-567.

Back, P. (1997). The dynamics of the herbaceous vegetation at 5 sites on Epping Forest National Park. Unpublished Report to the Department of Environment.

Bailey, P.T. (1971). The red kangaroo, *Megaleia rufa* (Desmarest), in north-western New South Wales. I. Movements. *CSIRO Wildlife Research* **16**: 11-28.

Baker, C.W., Givens, D.I. and Deaville, E.R. (1994). Prediction of organic matter digestibility *in vivo* of grass silage by near infrared reflectance spectroscopy: effect of calibration method, residual moisture and particle size. *Animal Feed Science and Technology* **50**:17-26.

Bakker, H.R. and Main, A.R. (1980). Condition, body composition and total body water estimation in the Quokka, *Setonix brachyurus* (Macropodidae). *Australian Journal of Zoology* **28**: 395-406.

Banks, E.M., Brooks, R.J. and Schnell, J. (1975). A radiotracking study of home range and activity of the brown lemming (*Lemmus trimucronatus*). *Journal of Mammalogy* **56**: 888-901.

Barboza, P.S. (1989). The nutritional physiology of the Vombatidae. PhD thesis, University of New England, Armidale.

Barboza, P.S. (1993a). Digestive strategies of the wombats: feed intake, fibre digestion, and digesta passage in two grazing marsupials with hindgut fermentation. *Physiological Zoology* **66**: 983-999.

Barboza, P.S. (1993b). Effects of restricted water intake on digestion, urea recycling and renal function in wombats (Marsupialia: Vombatidae) from contrasting habitats. *Australian Journal of Zoology* **41**: 527-536.

Barboza, P.S. and Hume, I.D. (1992a). Hindgut fermentation in the wombats: two marsupial grazers. *Journal of Comparative Physiology B* **162**: 561-566.

Barboza, P.S. and Hume, I.D. (1992b). Digestive tract morphology and digestion in the wombats (Marsupialia: Vombatidae). *Journal of Comparative Physiology B* **162**: 552-560.

Barboza, P.S., Hume, I.D. and Nolan, J.V. (1993). Nitrogen metabolism and requirements of nitrogen and energy in the wombats (Marsupialia: Vombatidae). *Physiological Zoology* **66**: 807-828.

Barker, R.D. (1986a). An investigation into the accuracy of herbivore diet analysis. *Australian Wildlife Research* **13**: 559-568.

Barker, R.D. (1986b). A technique to simplify herbivore diet analysis. *Australian Wildlife Research* **13**: 569-573.

Barker, R.D. (1987). The diet of herbivores in the sheep rangelands. In: *Kangaroos, their ecology and management in the sheep rangelands* (eds G. Caughley, N. Shepherd and J. Short) pp 69-83. Cambridge studies in applied ecology and resource management, University Press, Cambridge.

Barnett, J.L., How, R.A. and Humphreys, W.F. (1979). Blood parameters in natural populations of *Trichosurus* species (Marsupialia: Phalangeridae) II. Influence of habitat and population strategies of *T. caninus* and *T. vulpecula*. *Australian Journal of Zoology* **27**: 927-938.

Batzli, G.O. and Essecks, E. (1992). Body fat as an indicator of nutritional condition for the brown lemming. *Journal of Mammalogy* **73**: 431-39.

Bell, B.D. (1981). Breeding and condition of possums *Trichosurus vulpecula* in the Orongorongo valley, near Wellington, New Zealand. 1966-1975. In: *Marsupials in New Zealand* (ed B.D. Bell) pp. 85-139. Wellington: Victoria University of Wellington.

Belovsky, G.E. (1984). Herbivore optimal foraging: a comparative test of three models. *American Naturalist* **124**: 97-115.

Belovsky, G.E. and Slade, J.B. (1986). Time budgets of grassland herbivores: body size similarities. *Oecologia* **70**: 53-62.

Berg, E.P. and Marchello, M.J. (1994). Bioelectrical impedance analysis for the prediction of fat-free mass in lambs and lamb carcasses. *Journal of Animal Science* **72**: 322.

Borges, P.A., Dominguez-Bello, M.G. and Herrera, E.A. (1996). Digestive physiology of wild capybara. *Journal of Comparative Physiology B* **166**: 55-60.

Brochu, L., Caron, L. and Bergeron, J. (1988). Diet quality and body condition of dispersing and resident voles (*Microtus pennsylvanicus*). *Journal of Mammalogy* **69**: 704-10.

Brooks III. J., Anderson, M and Urness, P.J. (1984). Infrared reflectance analysis of forage quality for elk. *Journal of Wildlife Management* **48**: 254-258.

Brown, G.D. (1984). Thermoregulation in the Common wombat *Vombatus ursinus* in and alpine habitat. In: *Thermal Physiology* (ed J.R.S. Hales) pp331-334. Raven Press, New York.

Brown, G.D, and Taylor, L.S. (1984). Radio-telemetry transmitters for use in studies of thermoregulation of unrestrained Common wombats, *Vombatus ursinus*. *Australian Wildlife Research* **11**: 289-298.

Brown, G. and Young, G. (1982). Wombats - amiable native lawn-mowers. *Australian Natural History* **20**: 279-283.

Brown, R.D., Hellgren, E.C., Abbot, M., Ruthven, D.C. and Bingham, R.L. (1995). Effects of dietary energy and protein restriction on nutritional indices of female white-tailed deer. *Journal of Wildlife Management* **59**: 595-609.

Bunnell, F.L. and Gillingham, M.P. (1985). Foraging behaviour: dynamics of dining out. In: *Bioenergetics of wild herbivores* (eds R.J. Hudson and R.G. White) pp 53-77. CRC Press Incorporated, Florida.

Burnham, K.P., Anderson, D.R. and Laake, J.L. (1980). Estimation of density from line transect sampling of biological populations. *Wildlife Monographs* No. 72.

Cameron, R.D., Smith, W.T., Fancy, S.G., Gerhart, K.L. and White, R.G. (1993). Calving success of female caribou in relation to body weight. *Canadian Journal of Zoology* **71**: 480-486.

Catchpole, W.R. and Wheeler, C.F. (1992). Review: Estimating plant biomass: A review of techniques. *Australian Journal of Ecology* **17**: 121-132.

Caughley, G. (1964). Density and dispersion of two species of kangaroo in relation to habitat. *Australian Journal of Zoology* **12**: 238-249.

Caughley, G., Short, J., Grigg, G.C and Nix, H. (1987). Kangaroos and climate: an analysis of distribution. *Journal of Animal Ecology* **56**: 751-761.

Cha, K., Hill, A.G., Rounds, J.D. and Wilmore, D.W. (1995). Multifrequency bioelectrical impedance fails to quantify sequestration of abdominal fluid. *Journal of Applied Physiology* **78**: 736-39.

Chacon, E. and Stobbs, T.H. (1976). Influence of progressive defoliation of a grass sward on the eating behaviour of cattle. *Australian Journal of Agricultural Research* **27**: 709-727.

Chesson, J. (1978). Measuring preference in selective predation. *Ecology* **59**: 211-215.

Choo, G.M., Waterman, P.G., McKey, D.B. and Gartlan, J.S. (1981). A simple enzyme assay for dry matter digestibility and its value in studying food selection by generalist herbivores. *Oecologia* **49**:170-178.

Clark, D.H., Johnson, D.A., Kephart, K.D. and Jackson, N.A. (1995). Near infrared reflectance spectroscopy estimation of ^{13}C discrimination in forages. *Journal of Range Management* **48**: 132-136.

Clarke, J.L., Jones, M.E. and Jarman, P.J. (1989). A day in the life of a kangaroo: activities and movements of eastern grey kangaroos at Wallaby Creek. In: *Kangaroos, Wallabies and Rat-Kangaroos* (eds G. Grigg, P. Jarman and I. Hume) pp 611-618. Surry Beatty & Sons, Sydney.

Coleman, S.W., Barton II, F.E. and Meyer, R.D. (1985). The use of near-infrared reflectance spectroscopy to predict species composition of forage mixtures. *Crop Science* **25**: 834-837.

Contreras, L.C. and McNab, B.K. (1990). Thermoregulation and energetics of subterranean mammals. In: *Evolution of subterranean mammals at the organismal and molecular levels*. Eds: E. Nevo and O.A. Reig. Wiley-Liss, New York.

Copley, P.B. and Robinson, A.C. (1983). Studies on the yellow-footed rock-wallaby, *Petrogale xanthopus* Gray (Marsupialia: Macropodidae) II. Diet. *Australian Wildlife Research* **10**: 63-76.

Coppock, D.L., Detling, J,K., Ellis, J.E. and Dyer, M.I. (1983). Plant-herbivore interactions in a north American mixed-grass prairie. I. Effect of black-tailed prairie dogs on intraseasonal aboveground plant biomass and nutrient dynamics and plant species diversity. *Oecologia* **56**: 1-9.

Corbett, L.K. (1995). *The Dingo in Australia and Asia*. New South Wales University Press, Sydney.

Craighead, J.J., Craighead Jr, F.C., Ruff, R.L. and O'Gara, B.W. (1973). Home ranges and activity patterns of nonmigratory elk of the Madison drainage herd as determined by biotelemetry. *Wildlife Monographs* **33**: 1-50.

Croft, D.B. (1991a). Home range of the red kangaroo, *Macropus rufus*. *Journal of Arid Environments* **20**: 83-98.

Croft, D.B. (1991b). Home range of the euro, *Macropus robustus erubescens*. *Journal of Arid Environments* **20**: 99-112.

Crossman, D.G. (1988). Population ecology and diet of the Northern Hairy-nosed Wombat (*Lasiorhinus krefftii* (Owen)). Report to World Wildlife Fund, Australia.

Crossman, D.G., Johnson, C.N. and Horsup, A.B. (1994). Trends in the population of the northern hairy-nosed wombat in Epping Forest National Park. *Pacific Conservation Biology* **1**: 141-149.

Dailey, T.V., Hobbs, N.T. and Woodward, T.N. (1984). Experimental comparison of diet selection by mountain goats and mountain sheep in Colorado. *Journal of Wildlife Management* **48**: 799-806.

Dawson, L. (1983). The taxonomic status of small fossil wombats (Vombatidae: Marsupialia) from Quaternary deposits, and of related modern wombats. *Proceedings of the Linnean Society of New South Wales* **107**: 99-121.

Dawson, T.J., Denny, M.J.S., Russell, E.M. and Ellis, B. (1974). Water usage and diet preference of free ranging kangaroos, sheep and feral goats in the arid zone during summer. *Journal of Zoology* **177**: 1-23.

Dawson, T.J. and Ellis, B.A. (1979). Comparison of the diets of yellow-footed rock-wallabies and sympatric herbivores in western New South Wales. *Australian Wildlife Research* **6**: 245-254.

Dawson, T.J. and Hulbert, A.J. (1970). Standard metabolism, body temperature, and surface areas of Australian marsupials. *American Journal of Physiology* **218**: 1233-1238.

Dawson, T.J., Tierney, P.J. and Ellis, B.A. (1992). The diet of the bridled nailtail wallaby (*Onychogalea fraenata*). II. Overlap in dietary niche breadth and plant preferences with the black-striped wallaby (*Macropus dorsalis*) and domestic cattle. *Wildlife Research* **19**: 79-87.

Dearden, B.L., Pegau, R.E. and Hansen, R. M. (1975). Precision of microhistological estimates of ruminant food habits. *Journal of Wildlife Management* **39**: 402-407.

Del Giudice, G.D., Mech, L.D., Kunkel, K.E., Gese, E.M. and Seal, U.S. (1992). Seasonal patterns of weight, haematology, and serum characteristics of free-ranging female white-tailed deer in Minnesota. *Canadian Journal of Zoology* **70**: 974-983.

Dellow, D.W. (1982). Studies on the nutrition of macropodine marsupials. III. The flow of digesta through the stomach and intestine of macropodines and sheep. *Australian Journal of Zoology* **30**: 751-765.

Dellow, D.W. and Hume, I.D. (1982). Studies on the nutrition of macropodine marsupials. I. Intake and digestion of lucrene hay and fresh grass, *Phlaris aquatica*. *Australian Journal of Zoology* **30**: 391-398.

Dellow, D.W., Nolan, J.V. and Hume, I.D. (1983). Studies on the nutrition of macropodine marsupials. V. Microbial fermentation in the forestomach of *Thylogale thetis* and *Macropus eugenii*. *Australian Journal of Zoology* **31**: 433-443.

Demment, M.W. and Greenwood, G.B. (1988). Forage ingestion: effects of sward characteristics and body size. *Journal of Animal Science* **66**: 2380-2392.

Demment, M.W. and Van Soest, P.J. (1985). A Nutritional explanation for body-size patterns of ruminant and nonruminant herbivores. *American Naturalist* **125**: 641-672.

DeNiro, M.J. and Epstein, S. (1978). Influence of diet on the distribution of carbon isotopes in animals. *Geochimica et Cosmochimica Acta* **42**: 495-506.

Dennis, A.J. and Johnson, P.M. (1995). The rufous bettong. In: *The Mammals of Australia* (ed R. Strahan) pp 285-287. Reed Books, Australia.

De Vis, C.W. (1900). A new species of hairy-nosed wombat. *Annals of the Queensland Museum* **5**: 14-16.

Dickens, R.K. (1976). A comparison of haematological values from koalas in different locations and the effect of stress on those values. In: *The Koala: Proceedings of the Taronga Symposium on Koala Biology* (ed T.J. Bergin) pp 183-197. Sydney, Australia.

Dierenfeld, E.S. (1984). Diet quality of sympatric wombats, kangaroos and rabbits during severe drought. PhD thesis, Cornell University.

Dixon, K.R. and Chapman, J.A. (1980). Harmonic mean measure of animal activity areas. *Ecology* **61**: 1040-1044.

Dove, H. (1992). Using the n-alkanes of plant cuticular wax to estimate the species composition of herbage mixtures. *Australian Journal of Agricultural Research* **43**: 1711-1724.

Dove, H. and Mayes, R.W. (1991). The use of plant wax alkanes as marker substances in studies of nutrition in herbivores: a review. *Australian Journal of Agricultural Research* **46**: 913-952.

Dove, H. and Mayes, R.W. (1996). Plant wax components: a new approach to estimating intake and diet composition in herbivores. *Journal of Nutrition* **126**: 13-26.

Dove, H. and Moore, A.D. (1995). Using a least-squares optimisation procedure to estimate botanical composition based on the alkanes of plant cuticular wax. *Australian Journal of Agricultural Research* **46**: 1535-1544.

Dove, H., Mayes, R.W. and Freer, M. (1996). Effects of species, plant part, and plant age on the n-alkane concentrations in the cuticular wax of pasture plants. *Australian Journal of Agricultural Research* **47**: 1333-1347.

Duncan, P. (1992). *Horses and Grasses: The nutritional ecology of Equids and their impact on the Camargue.* Ecological studies Vol. 87. Springer-Verlag, New York.

Duncan, P., Foose, T.J., Gordon, I.J., Gakahu, C.G. and Lloyd, M. (1990). Comparative nutrient extraction from forages by grazing bovids and equids: a test of the nutritional model of equid/bovid competition and coexistence. *Oecologia* **84**: 411-418.

Dyson, W.G. and Herbin, G.A. (1968). Studies on plant cuticular waxes IV. Leaf wax alkanes as a taxonomic discriminant for cypresses grown in Kenya. *Phytochemistry* **7**: 1339-1344.

Dyson, W.G. and Herbin, G.A. (1970). Variation in leaf wax alkanes in cypress trees grown in Kenya. *Phytochemistry* **9**: 585-589.

Eckert, R., Randall, D.J. and Augustine, G. (1988). *Animal Physiology. Mechanisms and Adaptations*. W.H. Freeman and Company, New York.

Eglington, G., Hamilton, R.J. and Martin-Smith, M. (1962). The alkane constituents of some New Zealand plants and their possible taxonomic implications. *Phytochemistry* **1**: 137-145.

Ellis, B.A., Russell, E.M., Dawson, T.J. and Harrop, C.J.F. (1977). Seasonal changes in free-ranging red kangaroos, euros and sheep in western New South Wales. *Australian Wildlife Research* **4**: 127-144.

Ellis, B.A., Tierney, P.J. and Dawson, T.J. (1992). The diet of the bridled nailtail wallaby (*Onychogalea fraenata*). I. Site and seasonal influences and dietary overlap with the black-striped wallaby (*Macropus dorsalis*) and domestic cattle. *Wildlife Research* **19**: 65-78.

Erikstad, K.E. (1979). Effects of radio packages on reproductive success of willow grouse. *Journal of Wildlife Management* **43**: 170-175.

Evans, M. (1992). The bridled nailtail wallaby: ecology and management. MSc thesis, University of New England.

Farley, S.D. and Robbins, C.T. (1994). Development of two methods to predict body composition of bears. *Canadian Journal of Zoology* **72**: 220-26.

Feinsinger, P., Spears, E.E. and Poole, R.W. (1981). A simple measure of niche breadth. *Ecology* **62**: 27-32.

Fitzgerald, A.E. and Waddington, D.C. (1979). Comparison of two methods of fecal analysis of herbivore diet. *Journal of Wildlife Management* **42**: 468-473.

Fitzpatrick, E.A. (1967). Part IV. Climate of the Nogoa-Belyando Area. In*: Lands of the Nogoa-Belyando Area* (eds R.H. Gunn, R.W. Galloway, L. Pedley and E.A. Fitzpatrick) pp 68-82. CSIRO Australia Land Research Series No. 18.

Flannery, T.F. (1994). *The future eaters: an ecological history of the Australasian lands and people*. Reed Books, Australia.

Flinn, P.C., Windham, W.R. and Dove, H. (1994). Pasture intake by grazing sheep estimated using natural and dosed *n*-alkanes - a place for NIR? In: NIRS - Bridging the gap between data analysis and NIR application (eds K.I. Hildrum, T. Isaksson, T. Naer and A. Tandberg) pp 173-178. Ellis Horwood.

Flößer, R. (1986). Futterpräferenz-Analyse durch bestimmung von grasblattfragmenten im kot des Nördlichen Haarnasenwombats *Lasiorhinus latifrons barnardi* (Longman, 1939). *Säugetierkundliche Mitteilungen* **33**: 53-70.

Foley, W.J., McIlwee, A.P., Lawler, I., Aragones, L., Woolnough, A.P. and Berding, N. (1998) Ecological application of near-infrared spectroscopy - a tool for rapid, cost-effective prediction of the composition of plant and animal tissue and aspects of animal performance. *Oecologia.*

Frith, H.J. (1973). *Wildlife Conservation*. Angus and Robertson, Sydney.

Fryxell, J.M., Greever, J. and Sinclair, A.R.E. (1988). Why are migratory ungulates so abundant? *American Naturalist* **131**: 781-798.

Gales, R., Renouf, D. and Worthy, G.A.J. (1994). Use of bioelectrical impedance analysis to assess body composition of seals. *Marine Mammal Science* **10**: 1-12.

Gallivan, G.J., Culverwell, J. and Girdwood, R. (1995). Body condition indices of Impala, *Aepyceros melampus* - effect of age class, sex, season and management. *South African Journal of Wildlife Research* **25**: 23-31.

García-Ciudad, A., García-Ciudad, B. and Pérez-Corona, M.E. (1991). Prediction of botanical composition in grassland herbage by near infrared reflectance spectroscopy. *Journal of Science Food and Agriculture* **57**:507-515.

García-Ciudad, A., García-Ciudad, B., Pérez-Corona, M.E., Vazquez de Aldana, B.R. and Ruano-Ramos, A.M. (1993). Application of near-infrared reflectance spectroscopy to chemical analysis of heterogeneous and botanically complex grassland samples. *Journal of Science Food and Agriculture* **63**:419-426.

Garrott, R.A., Bartmann, R.M. and White, G.C. (1985). Comparison of radio-transmitter packages relative to deer fawn mortality. *Journal of Wildlife Management* **49**: 758-759.

Gaughwin, M.D. (1981). Socio-ecology of the Hairy-nosed Wombat (*Lasiorhinus latifrons*) in the Blanche Town region of South Australia. PhD thesis, University of Adelaide.

Gaughwin, M.D., Judson, G.J, Macfarlane, W.V. and Siebert, B.D. (1984). Effect of drought on the health of wild hairy-nosed wombats, *Lasiorhinus latifrons*. *Australian Wildlife Research* **11**: 455-463.

Georgiadis, N.J. and McNaughton, S.J. (1990). Elemental and fibre contents of savanna grasses: variation with grazing, soil type, season and species. *Journal of Applied Ecology* **27**: 623-634.

Giraldeau, L. (1984). Group foraging: skill pool effect and frequency-dependent learning. *American Naturalist* **124**: 72-79.

Gordon, G., Riney, T., Toop, J., Lawrie, B.C. and Godwin, M.D. (1985). Observations on the Queensland Hairy-nosed Wombat *Lasiorhinus krefftii* (Owen). *Biological Conservation* **33**: 165-195.

Gordon, I.J. and Illius, A.W. (1996). The nutritional ecology of African ruminants: a reinterpretation. *Journal of Animal Ecology* **65**: 18-28.

Gordon, I.J., Illius, A.W. and Milne, J.D. (1996). Sources of variation in the foraging efficiency of grazing ruminants. *Functional Ecology* **10**: 219-226.

Greenwood, R.J. and Sargeant, A.B. (1973). Influence of radio packs on captive mallards and blue-winged teal. *Journal of Wildlife Management* **37**: 3-9.

Griffiths, M. and Barker, R. (1966). The plants eaten by sheep and by kangaroos grazing together in a paddock in south-western Queensland. *CSIRO Wildlife Research* **11**: 145-167.

Griffiths, M., Barker, R. and MacLean, L. (1974). Further observations on he plants eaten by sheep and by kangaroos grazing together in a paddock in south-western Queensland. *Australian Wildlife Research* **1**: 27-43.

Gross, J.E., Alkon, P.U. and Demment, M.W. (1996). Nutritional ecology of dimorphic herbivores: digestion and passage rates in Nubian ibex. *Oecologia* **107**: 170-178.

Gwynne, M.D. and Bell, R.H.V. (1968). Selection of vegetation components by grazing ungulates in the Serengeti National Park. *Nature* **220**: 390-393.

Hamilton, R. (1997). Faecal steroid analysis as a technique for monitoring reproductive status in the hairy-nosed wombat. B.Sc.(Hons) thesis, Monash University.

Hammond, K.A. and Wunder, B.A. (1991). The role of diet quality and energy need in the nutritional quality of a small herbivore, *Microtus ochrogaster*. *Physiological Zoology* **64**: 541-567.

Hanley, T.A. (1997). A nutritional view of understanding and complexity in the problem of diet selection by deer (Cervidae). *Oikos* **79**: 209-218.

Hansson, L. (1970). Methods of morphological diet micro-analysis in rodents. *Oecologia* **21**: 255-266.

Harestad, A.S. and Bunnell, F.L. (1979). Home range and body weight - a reevaluation. *Ecology* **60**: 389-402.

Hartigan, J.A. (1987). Estimation of convex density contour in two dimensions. *Journal of the American Statistical Association* **82**: 267-270.

Haydock, K.P. and Shaw, H.H. (1975). The comparative yield method for estimating dry matter yield of pasture. *Australian Journal of Experimental Agriculture and Animal Husbandry* **15**: 663-670.

Helrich, K. (1990). Official Methods of Analysis of the Association of Official Analytical Chemists. Arlington, Virginia.

Herbin, G.A. and Robins, P.A. (1969). Patterns of variation and development in leaf wax alkanes. *Phytochemistry* **8**: 1985-1998.

Hilderbrand, M. (1988) *Analysis of vertebrate function*. John Wiley and Sons, New York.

Hill, G.J.E. (1982). Seasonal movement patterns of the eastern grey kangaroo in southern Queensland. *Australian Wildlife Research* **9**: 373-387.

Hines, J.E. and Zwickel, F.C. (1985). Influence of radio packages on young blue grouse. *Journal of Wildlife Management* **49**: 1050-1054.

Hirakawa, H. (1997). Digestion-constrained optimal foraging in generalist mammalian herbivores. *Oikos* **78**: 37-47.

Histϕl, T. and Hjeljord, O. (1993). Winter feeding strategies of migrating and nonmigrating moose. *Canadian Journal of Zoology* **71**: 1421-1428.

Holand, Ø. (1992). Fat indices versus ingesta-free body fat in European roe deer. *Journal of Wildlife Management* **56**, 241-45.

Hollis, C.J., Robertshaw, J.D. and Harden, R.H. (1986). Ecology of the swamp wallaby (*Wallabia bicolor*) in north-eastern New South Wales. I. Diet. *Australian Wildlife Research* **13**: 355-365.

Holecheck, J.L., Vavra, M. and Piper, R.D. (1982). Botanical composition determination of range herbivore diets: A review. *Journal of Range Management* **35**: 309-315.

Horsup, A. (1997). A trapping survey of the northern hairy-nosed wombat. Proceedings of the 'Wombats in Australia: biology, history, art and literature' Conference. Adelaide, September 1994. Surrey Beatty and Sons, Sydney.

Horsup, A. (1998). Recovery plan for the Northern Hairy-nosed Wombat (*Lasiorhinus krefftii*). A Report Submitted to Threatened Species and Communities Section, Environment Australia (Project 191).

Horsup, A. and Marsh, H. (1992). The diet of the allied rock-wallaby, *Petrogale assimilis*, in the wet-dry tropics. *Wildlife Research* **19**: 17-33.

Hoyle, S.D., Horsup, A.B., Johnson, C.N., Crossman, D.G. and McCallum, H. (1995). Live-trapping of the Northern Hairy-nosed Wombat (*Lasiorhinus krefftii*): Population-size estimates and effects on individuals. *Wildlife Research* **22**: 741-755.

Hume, I.D. (1978). Evolution of the Macropodidae digestive system. *Australian Mammalogy* **2**: 37-42.

Hume, I.D. (1982). *Digestive physiology and nutrition of marsupials.* Cambridge University Press, New York.

Hume, I.D. (1989). Optimal digestive strategies in mammalian herbivores. *Physiological Zoology* **62**: 1145-1163.

Illius, A.W. and Gordon, I.J. (1992). Modelling the nutritional ecology of ungulate herbivores: evolution of body size and competitive interactions. *Oecologia* **89**: 428-434.

Illius, A.W. and Gordon, I.J. (1993). Diet selection in mammalian herbivores: constraints and tactics. In: *Diet selection: an interdisciplinary approach to foraging behaviour* (ed R.N. Hughes) pp 157-181. Blackwell Scientific Publications, Oxford.

Illius, A.W. and Gordon, I.J. (1999). The physiological ecology of mammalian herbivory. In: *Vth International Symposium on the Nutrition of Herbivores* (eds H.J.G. Jung and G.C. Fahey Jr.) American Society of Animal Science, Savoy Il.

Ivlev, V.S. (1961). *Experimental ecology of the feeding of fishes.* Yale University Press, Connecticut, USA.

Janis, C. (1976). The evolutionary strategy of the Equidae and the origins of rumen and cecal digestion. *Evolution* **30**: 757-774.

Jaremovic, R.V. and Croft, D.B. (1987). Comparison of techniques to determine eastern grey kangaroo home range. *Journal of Wildlife Management* **51**: 921-930.

Jarman, P.J. (1971). Diets of large mammals in the woodlands around Lake Kariba, Rhodesia. *Oecologia* **8**: 157-178.

Jarman, P.J. (1974). The social organisation of antelope in relation to their ecology. *Behaviour* **48**: 215-266.

Jarman, P.J. (1987). Group size and activity in eastern grey kangaroos. *Animal Behaviour* **35**: 1044-1050.

Jarman, P.J. (1993). Plant communities and the social organization and community structure of herbivorous mammals. In: *Mutualism and Community Organization. Behavioural, Theoretical, and Food-Web Approaches* (eds H. Kawanabe, J.E. Cohen and K. Iwasaki) pp 109-132. Oxford University Press, Oxford.

Jarman, P.J. (1994). The eating of seedheads by species of Macropodidae. *Australian Mammalogy* **17**:51-63.

Jarman, M.V. and Jarman, P.J. (1976). Daily activity of impala. *East African Wildlife Journal* **11**: 75-92.

Jarman, P.J., Johnson, C.N., Southwell, C.J. and Stuart-Dick, R. (1987). Macropod studies at Wallaby Creek. I. The area and animals. *Australian Wildlife Research* **14**: 1-14.

Jarman, P.J. and Sinclair, P.J. (1979). Feeding strategy and the pattern of resource partitioning in ungulates. In: *Serengeti. Dynamics of an Ecosystem* (eds A.R.E. Sinclair and M. Norton-Griffiths) pp 130-163. University of Chicago Press, Chicago.

Johannesen, E., Andreassen, H.P. and Steen. H. (1997). Effect of radiolcollars on survival of root voles. Journal of Mammalogy **78**: 638-42.

Johnson, C.N. (1991a). Behaviour and Ecology of the Northern Hairy-nosed Wombat, *Lasiorhinus krefftii*. Internal Report, Queensland National Parks and Wildlife Service.

Johnson, C.N. (1991b). Utilisation of habitat by the northern hairy-nosed wombat, *Lasiorhinus krefftii*. *Journal of Zoology* **225**: 495-507.

Johnson, C.N. (1997). The evolutionary ecology of wombats. Proceedings of the 'Wombats in Australia: biology, history, art and literature' Conference. Adelaide, September 1994. Surrey Beatty and Sons, Sydney.

Johnson, C.N. and Crossman, D.G. (1991). Dispersal and social organisation of the northern hairy-nosed wombat *Lasiorhinus krefftii*. *Journal of Zoology* **225**: 605-613.

Johnson, C.N. and Jarman, P.J. (1987). Macropod studies at Wallaby Creek VI. A validation of the use of dung-pellet counts for measuring absolute densities of populations of macropodids. *Australian Wildlife Research* **14**: 139-145.

Johnson, C.N., Jarman, P.J. and Southwell, C.J. (1987). Macropod studies at Wallaby Creek V. Patterns of defecation by eastern grey kangaroos and red-necked wallabies. *Australian Wildlife Research* **14**: 133-138.

Johnson, R.N. and Berner, A.H. (1980). Effects of radio transmitters on released cock pheasants. *Journal of Wildlife Management* **44**: 686-689.

Jones, R.J., Ludlow, M.M., Troughton, J.H. and Blunt, C.G. (1979). Estimation of the proportions of C_3 and C_4 plant species in the diet of animals from the ratio of natural ^{12}C and ^{13}C isotopes in the faeces. *Journal of Agricultural Science* **92**: 91-100.

Kean, L.E. (1990). A study on the diets of wombats and rabbits on Brookfield Conservation Park, via faecal pellet analysis. B.Sc.(Hons) thesis, Flinders University of South Australia.

Kellaway, R.C. and Stimson, C. (1993). Near infrared reflectance spectroscopy of fibre. In: *Dietary Fibre and Beyond - Australian Perspectives* (eds S. Samman and G. Annison) pp 95-106. Nutritional Society of Australia Occasional Publications Volume 1.

Kershaw, J.A. (1909). Notes on the hairy-nosed wombat, *Phascolomys latifrons* Owen. *Victorian Naturalist* **26**: 118-119.

Khan, M.K.M. (1985). *Asian Rhinos. An action plan for their conservation.* IUCN, Switzerland.

Kirkpatrick, T.H. (1965). Studies of Macropodidae in Queensland. 1. Food preferences of the grey kangaroo (*Macropus major* Shaw). *Queensland Journal of Agricultural and Animal Sciences* **22**: 89-93.

Klieber, M. (1961). *The fire of life: an introduction to animal energetics.* John Wiley and Sons, New York, USA.

Kohlmann, S.G. and Risenhoover, K.L. (1994). Spatial and behavioural response of white-tailed deer to forage depletion. *Canadian Journal of Zoology* **72**: 506-513.

Komarek, A.R., Robertson, J.B. and Van Soest, P.J. (1994a). Comparison of the filter bag technique to conventional filtration in the Van Soest NDF analysis of 21 feeds. Abstract: National Conference on Forage Quality, Evaluation, and Utilization, University of Nebraska.

Komarek, A.R., Robertson, J.B. and Van Soest, P.J. (1994b). A comparison of methods for determining ADF using the filter bag technique versus conventional filtration. Abstract: ASAS/ADSA Meetings, USA.

Krausman, P.R. (1978). Forage relationships between two deer species in Big Bend National Park, Texas. *Journal of Wildlife Management* **42**: 101-107.

Krebs, C.J. and Singleton, G.R. (1993). Indices of condition for small mammals. *Australian Journal of Zoology* **41**: 317-23.

Kushner, R.F. (1992). Bioelectrical Impedance Analysis: A review of principles and applications. *Journal of the American College of Nutrition* **11**, 199-209.

Langbein, J., Sheibe, K.M., Eichorn, K., Lindner, U., Striech and W.J. (1996). An activity data logger for monitoring free-ranging animals. *Applied Animal Behaviour Science* **48**: 115-124.

Laredo, M.A., Simpson, G.D., Minson, D.J. and Orpin, C.G. (1991). The potential for using n-alkanes in tropical forages as a marker for the determination of dry matter intake by grazing ruminants. *Journal of Agricultural Science* **117**: 355-361.

LeFeuvre, R.P. and Jones, R.J. (1988). The static combustion of biological samples sealed in glass tubes as a preparation for $\delta^{13}C$ determination. *Analyst* **113**: 817-823.

Lechowicz, M.J. (1982). The sampling characteristics of electivity indices. *Oecologia* **52**: 22-30.

Loeb, S.C., Schwab, R.G. and Demment, M.W. (1991). Responses of pocket gophers (*Thomomys bottae*) to changes in diet quality. *Oecologia* **86**: 542-551.

Lukaski, H.C., Bolonchuk, W.W., Hall C.B. and Siders, W.A. (1986). Validation of tetrapolar bioelectrical impedance method to assess human body composition. *Journal of Applied Physiology* **60**:1327-32.

Lyons, K.R., Stuth, J.W. and Angerer, J.P. (1995). Technical note: fecal NIRS equation field validation. *Journal of Range Management* **48**: 380-382.

Maclean, G.S. (1981). Factors influencing the composition of respiratory gases in mammal burrows. *Comparative Biochemistry and Physiology* **69**: 373-380.

Maddock, L. (1979). The "Migration" and grazing succession. In: : *Serengeti. Dynamics of an Ecosystem* (eds A.R.E. Sinclair and M. Norton-Griffiths) pp 104-129. University of Chicago Press, Chicago.

Mallet, K.J. and Cooke, B.D. (1986). *The ecology of the Common wombat in South Australia*. Nature Conservation Society of South Australia Incorporated, Adelaide.

Mannetje, L. 't, and Haydock, K.P. (1963). The dry-weight-rank method for the botanical analysis of pasture. *Journal of the British Grasslands Society* **18**: 268-275.

Mantel, M. (1967). The detection of disease clustering and a generalised regression approach. *Cancer Research* **27**: 209-220.

Marchello, M.J. and Slanger, W.D. (1994). Bioelectrical impedance can predict skeletal muscle of beef cows and their carcasses. *Journal of Animal Science* **72**: 3118-23.

Mayes, R.W., Lamb, C.S. and Colgrove, P.M. (1986). The use of dosed and herbage n-alkanes as markers for the determination of herbage intake. *Journal of Agricultural Science* **107**: 161-170.

McIllwee, A.P. (1994). The dietary ecology of three mycophagous marsupials in north-eastern Queensland. B.Sc. (Hons) thesis, James Cook University.

McIlroy, J.C. (1973). Aspects of the ecology of the common wombat, *Vombatus ursinus* (Shaw 1800). PhD thesis, Australian National University, Canberra.

McIntosh, D.L. (1966). The digestibility of two roughages and the rates of passage of their residues by the red kangaroo, *Megaleia rufa* (Desmarest), and the merino sheep. *CSIRO Wildlife Research* **11**: 125-135.

McNab, B.K. (1963). Bioenergetics and the determination of home range size. *American Naturalist* **97**: 133-140.

McNab, B.K. (1979). The influence of body size on the energetics and distribution of fossorial and burrowing mammals. *Ecology* **60**: 1010-1021.

McNab, B.K. (1988). Complications inherent in scaling the basal rate of metabolism in mammals. *Quarterly Review of Biology* **63**: 25-54.

McNaughton, S.J. (1979). Grassland-Herbivore dynamics. In: *Serengeti. Dynamics of an Ecosystem* (eds A.R.E. Sinclair and M. Norton-Griffiths) pp 46-81. University of Chicago Press, Chicago.

McNaughton, S.J. (1985). Ecology of a grazing system: the Serengeti. *Ecological Monographs* **55**: 259-294.

McNaughton, S.J. and Banyikwa, F.F. (1995). Plant communities and herbivory. In: *Serengeti II. Dynamics, Management and Conservation of an Ecosystem* (eds A.R.E. Sinclair and P. Arcese) pp 49-70. University of Chicago Press, Chicago.

McNaughton, S.J., Tarrants, J.L., McNaughton, M.M. and Davis, R.H. (1985). Silica as a defence against herbivory and a growth promoter in African grasses. *Ecology* **66**: 528-535.

Melcher, J.C., Armitage, K.B. and Porter, W.P. (1989). Energy allocation by yellow-bellied marmots. *Physiological Zoology* **62**: 429-448.

Melcher, J.C., Armitage, K.B. and Porter, W.P. (1990). Thermal influences on the activity and energetics of yellow-bellied marmots (*Marmota flaviventris*). *Physiological Zoology* **63**: 803-820.

Merchant, J.C. (1995). The swamp wallaby. In: *The Mammals of Australia* (ed R. Strahan) pp 404-405. Reed Books, Australia.

Meuret, M. Dardenne, P., Biston, R. and Poty, O. (1993). The use of NIR in predicting nutritive value of Mediterranean tree and shrub foliage. *Journal of Near Infrared Spectroscopy* **1**: 45-54.

Minson, D.J., Ludlow, M.M. and Troughton, J.H. (1975). Differences in natural carbon isotope ratios of milk and hair from cattle grazing tropical and temperate pastures. *Nature* **256**: 602.

Murray, M.G. (1995). Specific nutrient requirements and migration of wildebeest. In: *Serengeti II. Dynamics, Management and Conservation of an Ecosystem* (eds A.R.E. Sinclair and P. Arcese) pp 231-256. University of Chicago Press, Chicago.

Nagy, K.A. (1980). CO_2 production in animals: analysis of potential error in the doubly labelled water method. *American Journal of Physiology* **238**, R466-73.

Nagy, K.A. and Costa, D.P. (1980). Water flux in animals: analysis of potential errors in the tritiated water method. *American Journal of Physiology* **238**, R454-65.

Newman, J.A., Thompson, W.A., Penning, P.D. and Mayes, R.W. (1995). Least-squares estimation of diet composition from n-alkane herbage and faeces using matrix mathematics. *Australian Journal of Agricultural Research* **46**: 793-805.

Newsome, A.E. (1965). The abundance of red kangaroos, *Megaleia rufa* (Desmarest), in central Australia. *Australian Journal of Zoology* **13**: 269-287.

Norberg, R. Å. (1977). An ecological theory on foraging time and energetics and choice of optimal food-searching method. *Journal of Animal Ecology.* **46**: 511-529.

Norbury, G.L. (1988a). A comparison of stomach and faecal samples for diet analysis of grey kangaroos. *Australian Wildlife Research* **15**: 249-255.

Norbury, G.L. (1988b). Microscopic analysis of herbivore diets - a problem and a solution. *Australian Wildlife Research* **15**: 51-57.

Norbury, G.L. and Sanson, G.D. (1992). Problems with measuring diet selection of terrestrial, mammalian herbivores. *Australian Journal of Ecology* **17**: 1-7.

Norbury, G.L., Sanson, G.D. and Lee, A.K. (1989). Feeding ecology of the Macropodoidea. In: *Kangaroos, Wallabies and Rat-Kangaroos* (eds G. Grigg, P. Jarman and I. Hume) pp 169-178. Surry Beatty & Sons, Sydney.

Norgan, N.G. (1994). Population differences in body composition in relation to the body mass index. *European Journal of Clinical Nutrition* **48** (Supplement 3), S10-27.

Norris, K.H., Barnes, R.F., Moore, J.E. and Shenk, J.S. (1976). Predicting forage quality be infrared reflectance spectroscopy. *Journal of Animal Science* **43**:879-897.

Nortcote, K.H. (1971). *A factual key for the recognition of Australian soils.* Rellim Technical Publications, Glenside, South Australia.

Nowak, R.M. (1991). *Walker's Mammals of the World.* The Johns Hopkins University Press, Baltimore.

Oftedahl, O.T. (1985). Pregnancy and lactation. In: *Bioenergetics of wild herbivores* (eds R.J. Hudson and R.G. White) pp 215-238. CRC Press Incorporated, Florida.

Olubajo, F.O., Van Soest, P.J. and Oyenuga, V.A. (1974). Comparison and digestibility of four tropical grasses grown in Nigeria. *Journal of Animal Science* **38**: 149-153.

Orell, P. (1989). An assessment of microscopic techniques for the analysis of herbivore diets. B.Sc.(Hons) thesis, Flinders University of South Australia.

Orton, R.K., Hume, I.D. and Leng, R.A. (1985). Effects of exercise and level of dietary protein on digestive function in horses. *Equine Veterinary Journal* **17**: 386-390.

Osborne, B,G., Fearn, T. and Hindle, P.H. (1993) Practical NIR spectroscopy with applications in food and beverage analysis. Longman Scientific and Technical, Singapore.

Owen-Smith, N. (1982). Factors influencing the consumption of plant products by large herbivores. In: *Ecology of Tropical Savannas* (eds B.J. Huntley and B.H. Walker) pp 359-404. Springer-Verlag, Berlin.

Owen-Smith, N. (1992). *Megaherbivores. The influence of very large body size on ecology*. Cambridge University Press, Cambridge.

Owen-Smith, N. (1993). Assessing the constraints for optimal diet models. *Evolutionary Ecology* **7**: 530-531.

Pace, N. and Rathbun, E.N. (1945). Studies on body composition III. The body water and chemically combined nitrogen content in relation to fat content. *Journal of Biological Chemistry* **158**, 685-91.

Para, R. (1978). Comparison of foregut and hindgut fermentation in herbivores. In: *The ecology of arboreal folivores* (ed G.G. Montgomery) pp 205-229. The symposia of the National Zoological Park, Smithsonian Institution Press, Washington D.C.

Parker, K.L., Gillingham, M.P., Hanley, T.A. and Robbins, C.T. (1993). Seasonal patterns in body mass, body composition, and water transfer rates of free-ranging and captive black-tailed deer (*Odocoileus hemionus sitkensis*) in Alaska. *Canadian Journal of Zoology* **71**: 1397-1404.

Pellew, R.A. (1984). Food consumption and energy budgets of the Giraffe. *Journal of Applied Ecology* **21**: 141-159.

Penry, D.L. (1993). Digestive constraints on diet selection. In: *Diet selection: an interdisciplinary approach to foraging behaviour* (ed R.N. Hughes) pp 32-55. Blackwell Scientific Publications, Oxford.

Penry, D.L. and Jumars, P.A. (1986). Chemical reactor analysis and optimal digestion. *BioScience* **36**: 311-315.

Penry, D.L. and Jumars, P.A. (1987). Modelling animal guts as chemical reactors. *American Naturalist* **36**: 310-315.

Perry, R.J. and Braysher, M.L. (1986). A technique for estimating the number of eastern grey kangaroos, *Macropus giganteus*, grazing in a given area of pasture. *Australian Wildlife Research* **13**: 335-338.

Petrides, G.A. (1975). Principal foods versus preferred foods and their relations to stocking rate and range condition. *Biological Conservation* **7**: 161-169.

Pond, C.M. (1978). Morphological aspects and the ecological and mechanical consequences of fat deposition in wild vertebrates. *Annual Review of Ecology and Systematics* **9**: 519-70.

Poppi, D.P. (1996). Predictions of food intake in ruminants from analyses of food composition. *Australian Journal of Agricultural Research* **47**: 489-504.

Poppi, D.P., Hendricksen, R.E. and Minson, D.J. (1985). The relative resistance to escape of leaf and stem particles from the rumen of sheep and cattle. *Journal of Agricultural Science* **105**: 9-14.

Pratt, D.J and Gwynne, M.D. (1977). *Rangeland Management and Ecology in East Africa*. Hodder and Stoughton, London.

Price, M.A. and White, R.G. (1985). Growth and development. In: *Bioenergetics of wild herbivores* (eds R.J. Hudson and R.G. White) pp 183-213. CRC Press Incorporated, Florida.

Pyke, G.H., Pulliam, H.R. and Charnov, E.L. (1977). Optimal foraging: a selective review of theory and tests. *Quarterly Review of Biology* **52**:137-154.

Rabotnikof, C.M., Planas, G.M., Silva Colomer, J. and Stritzler, N.P. (1995). Near infrared reflectance spectroscopy (NIRS) for predicting forage quality of perennial warm-season grasses in La Pampa, Argentina. *Annals of Zootech* **44**: 97-100.

Radojevic, I., Simpson, R.J., St. John, J.A. and Humphreys, M.O. (1994). Chemical composition and *in vitro* digestibility of lines of *Lolium perenne* selected for high concentrations of water-soluble carbohydrate. *Australian Journal of Agricultural Research* **45**: 901-912.

Ransom, A.B. (1978). Kidney and marrow fat as indicators of white-tailed deer condition. *Journal of Wildlife Management* **29**: 397-98.

Reichman, O.J. and Smith, S.C. (1990). Burrows and burrowing behaviour by mammals. In: *Current Mammalogy, Volume 2* (ed H.H. Genoways) pp 197-244. Plenum Press, New York and London.

Rishworth, C., McIlroy, J.C. and Tanton, M.T. (1995a). Diet of the Common Wombat, *Vombatus ursinus*, in plantations of *Pinus radiata*. *Wildlife Research* **22**: 333-339.

Rishworth, C., McIlroy, J.C. and Tanton, M.T. (1995b). Factors affecting population densities of the Common Wombat, *Vombatus ursinus*, in plantations of *Pinus radiata*. *Forest Ecology and Management* **76**: 11-19.

Robbins, C.T. (1983). *Wildlife feeding and nutrition*. Academic Press, New York.

Robertson, G. (1987). Plant dynamics. In*: Kangaroos, their ecology and management in the sheep rangelands* (eds G. Caughley, N. Shepherd and J. Short) pp 50-68. Cambridge studies in applied ecology and resource management, University Press, Cambridge.

Robertson, G.G. and Gepp, B. (1982). Capture of kangaroos by 'stunning'. *Australian Wildlife Research* **9**: 393-96.

Robertson, G., Short, J. and Wellard, G. (1987). The environment of the Australian sheep rangelands. In*: Kangaroos, their ecology and management in the sheep rangelands* (eds G. Caughley, N. Shepherd and J. Short) pp 14-34. Cambridge studies in applied ecology and resource management, University Press, Cambridge.

Rumpler, W.V., Allen, M.E., Ullrey, D.E., Earle, R.D., Schmitt, S.M. and Cooley, T.M. (1987). Body composition of white-tailed deer estimated by deuterium oxide dilution. *Canadian Journal of Zoology* **65:** 204-208.

Salt, C.A., Mayes, R.W., Colgrove, P.M., and Lamb, C.S. (1994). The effects of season and diet composition on the radiocaesium intake by sheep grazing heather moorland. *Journal of Applied Ecology* **31**: 125-136.

Sanson, G.D. (1989). Morphological adaptations of teeth to diets and feeding in the Macropodidae. In: *Kangaroos, wallabies and rat-kangaroos* (eds G. Grigg, P. Jarman and I. Hume) pp 151-168. Surrey Beatty and Sons, Sydney.

Santomá, G., De Blas, J.C., Carabaño and Fraga, M.J. (1989). Nutrition of Rabbits. In: *Recent advances in animal nutrition* (eds W. Haresign and D.J.A. Cole) pp 109-138. Butterworths, London.

Schoener, T.W. (1971). Theory of feeding strategies. *Annual Review of Ecology and Systematics* **11**: 369-404.

Schoener, T.W. (1979). Generality of the size-distance relation in models of optimal feeding. *American Naturalist* **114**: 902-914.

Seebeck, J.H. and Rose, R.W. (1989). Potoroidae. In: *Fauna of Australia. Mammalia* (eds D.W. Walton and B.J. Richardson) pp 716-739. Canberra: Australian Government Publishing Service Vol. 1B.

Senanayake, S.G.J.N. (1995). The effect of different light levels on the nutritive quality of four natural tropical grasses. *Tropical Grasslands* **29**: 111-114.

Shenk, J.S., Westerhaus, M.O. and Hoover, M.R. (1979). Analysis of forages by infrared reflectance. *Journal of Dairy Science* **62**:807-812.

Shenk, J.S. and Westerhaus, M.O. (1991). New standardisation and calibration procedures for near infrared reflectance spectroscopy. *Crop Science* **31**: 469-474.

Shenk, J.S. and Westerhaus, M.O. (1992). NIRS 3. Version 3.00. Routine operation, calibration and network system management software for Near Infrared instruments. Infrasoft International, Port Matilda, Pa, USA.

Shenk, J.S. and Westerhaus, M.O. (1994). The application of near infrared spectroscopy (NIRS) to forage analysis. In: *National Conference on Forage Quality, Evaluation and Utilization, University of Nebraska* (eds G.C. Fahey Jr., L.E. Mosser, D.R. Martens and M. Collins) pp 406-449. American Society of Agronomy Inc., Crop Science Society of America Inc. and Soil Science Society of America Inc., Madison, WI, USA.

Shenk, J.S., Workman Jr., J.J. and Westerhaus, M.O. (1992). Application of NIR spectroscopy to agricultural products. In: *Handbook of Near-infrared analysis* (eds D.A. Burns and E.W. Ciurczak) pp 383-431. Marcel Dekker Inc., New York.

Sheng, H.P. and Huggins, R.A. (1979). A review of body composition with emphasis on total body water and fat. *American Journal of Clinical Nutrition* **32**: 630-47.

Shipley, L.A. and Spalinger, D.E. (1992). Mechanics of browsing in dense food patches: effects of plant and animal morphology on intake rate. *Canadian Journal of Zoology* **70**: 1743-1752.

Short, J. (1987). Factors affecting food intake of rangeland herbivores. In: *Kangaroos, their ecology and management in the sheep rangelands* (eds G. Caughley, N. Shepherd and J. Short) pp 84-99. Cambridge studies in applied ecology and resource management, University Press, Cambridge.

Sinclair, A.R.E. (1977). *The African Buffalo: A Study of Resource Limitation of Populations*. The University of Chicago Press, Chicago.

Sinclair, A.R.E. (1985). Does interspecific competition or predation shape the African ungulate community? *Journal of Animal Ecology* **54**: 899-918.

Sinclair, A.R.E. (1995). Serengeti past and present. In: *Serengeti II. Dynamics, Management and Conservation of an Ecosystem* (eds A.R.E. Sinclair and P. Arcese) pp 3-30. University of Chicago Press, Chicago.

Smith, K.F. and Flynn P.C. (1991). Monitoring the performance of a broad-based calibration for measuring the nutritive value of two independent populations of pasture using near infrared reflectance (NIR) spectroscopy. *Australian Journal of Experimental Agriculture* **31**:205-210.

Spalinger, D.E. and Hobbs, N.T. (1992). Mechanisms of foraging in mammalian herbivores: new models of functional response. *American Naturalist* **140**: 325-348.

Spinage, C.A. (1982). *A Territorial Antelope: The Uganda Waterbuck*. Academic Press, London.

Southwell, C. J. (1984). Variability in grouping in the eastern grey kangaroo, Macropus giganteus. II. Dynamics of group formation. *Australian Wildlife Research* **11**: 437-449.

Southwell, C. (1989). Techniques for monitoring the abundance of kangaroo and wallaby populations. In: *Kangaroos, wallabies and rat kangaroos* (eds G. Grigg, P. Jarman and I. Hume) pp 659-693. Surrey Beatty and Sons, New South Wales.

Southwell, C.J., Weaver, K.E., Cairns, S.C., Pople, A.R., Gordon, A.N., Sheppard, N.W. and Broers, R. (1995). Abundance of macropods in north-eastern New South Wales, and the logistics of broad-scale ground surveys. *Wildlife Research* **22**: 757-766.

St John, B.J. and Saunders, G.M. (1989). Plan of management for the hairy-nosed wombat in South Australia. Unpublished Report, South Australian National Parks and Wildlife.

Steele, V.R., Magrath, M.J.L. and Temple-Smith, P.D. (1995). Activity monitoring of southern hairy-nosed wombats (*Lasiorhinus latifrons*) using microchip identification. Proceedings of the Australian Mammal Society, Townsville.

Steinbeck C. (1994). A spatial analysis and assessment of the habitat of the Northern Hairy-nosed Wombat (*Lasiorhinus krefftii*). Postgraduate Diploma of Science thesis, James Cook University.

Stevens, C.E. and Hume, I.D. (1996). *Comparative physiology of the vertebrate digestive system*. Cambridge University Press, Cambridge.

Stewart, D.R.M. (1967). Analysis of plant epidermis in faeces: A technique for studying the food preference of grazing herbivores. *Journal of Applied Ecology* **4**: 83-111.

Stobbs, T.H. (1973a). The effect of plant structure on the intake of tropical pastures. I. Variation in the bite size of grazing cattle. *Australian Journal of Agriculture Research* **24**: 809-819.

Stobbs, T.H. (1973b). The effect of plant structure on the intake of tropical pastures. II. Differences in sward structure, nutritive value, and bite size of animals grazing *Setaria anceps* and *Chloris gayana* at various stages. *Australian Journal of Agriculture Research* **24**: 821-829.

Strahan, R. (1995). *The Mammals of Australia*. Reed Books, Australia.

Swantek, P.M., Crenshaw, J.D., Marchello, M.J. and Lukaski, H.C. (1992). Bioelectrical Impedance: A nondestructive method to determine fat-free mass of live market swine and pork carcasses. *Journal of Animal Science* **70**: 169-77.

Taylor, A.C. (1995). Molecular ecology of the endangered Northern Hairy-nosed Wombat *Lasiorhinus krefftii*, and application to conservation and management. PhD thesis, University of New South Wales, Sydney.

Taylor, A.C., Sherwin, W.B. and Wayne, R.K. (1994). Genetic variation of microsatellite loci in a bottlenecked species: the northern hairy-nosed wombat *Lasiorhinus krefftii*. *Molecular Ecology* **3**: 277-290.

Taylor, G.K. (1977). Moorunde Wildlife Reserve: wombat population study progress report. Unpublished Report, South Australian National Parks and Wildlife Service.

Taylor, R. (1994). Plants have fingerprints too. *Rural Research* **165**: 10-15.

Tieszen, L.L. and Boutton, T.W. (1988). Stable carbon isotopes in terrestrial ecosystem research. In: *Stable Isotopes in Ecological Research* (eds P.W. Rundel, J.R. Ehleringer and K.A. Nagy) pp 167-195. Ecological Studies Volume 68. Springer-Verlag, New York.

Tieszen, L.L., Boutton, T.W., Tesdahl, K.G. and Slade, N.A. (1983). Fractionation and turnover of stable carbon isotopes in animal tissue: Implications for $\delta^{13}C$ analysis of diet. *Oecologia* **57**: 32-37.

Tieszen, L.L., Hein, D., Qvortrup, S.A., Troughton, J.H. and Imbamba, S.K. (1979). Use of $\delta^{13}C$ values to determine vegetation selectivity in East African herbivores. *Oecologia* **37**: 337-350.

Tixier, H., Duncan, P., Scehovic, J., Yani, A., Gleizes, M. and Lila, M. (1997). Food selection by European roe deer (*Capreolus capreolus*): effects of plant chemistry, and consequences for the nutritional value of their diets. *Journal of Zoology* **242**: 229-245.

Tothill, J.C., Hargreaves, J.N.G., Jones, R.M. and McDonald, C.K. (1992). BOTANAL - A comprehensive sampling and computing procedure for estimating pasture yield and composition. 1. Field Sampling. Tropical Agronomy Technical Memorandum Number 78, CSIRO Tropical Crops and Pasture, St. Lucia, Australia.

Triggs, B. (1988). *The Wombat - Common Wombats in Australia.* New South Wales University Press, Sydney.

Trudell, J. and White, R.G. (1981). The effect of forage structure and availability on food intake, biting rate, bite size and daily eating time of reindeer. *Journal of Applied Ecology* **18**: 63-81.

Vanderploeg, H.A. and Scavia, D. (1979). Calculation and use of selectivity coefficients of feeding: zooplankton grazing. *Ecological Modelling* **7**: 135-149.

Van Rooyen, A.F. (1992). Diets of impala and nyala in two game reserves in Natal, South Africa. *South African Journal of Wildlife Research* **22**: 98-101.

Van Rooyen, A.F. (1993). Variation in body condition of impala and nyala in relation to social status and reproduction. *South African Journal of Wildlife Research* **23**: 36-38.

Van Soest, P.J. (1964). Symposium on nutrition and forage and pastures: new chemical procedures for evaluating forages. *Journal of Animal Science* **123**: 838-845.

Van Soest, P.J. (1965). Symposium on factors influencing the voluntary intake of herbage by ruminants: voluntary intake in relation to chemical composition and digestibility. *Journal of Animal Science* **124**: 834-843.

Van Soest, P.J., Robertson, J.B. and Lewis, B.A. (1991). Methods for dietary fiber, neutral detergent fiber and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science* **74**:3583-3597.

Vaughan, B.E. and Boling, E.A. (1961). Rapid assay procedures for tritium-labelled water in body fluids. *Journal of Laboratory and Clinical Medicine* 57: 159-64.

Vázquez de Aldana, B.R., García-Criado, B., García-Criado, A. and Pérez-Corona, M.E. (1996). Non-destructive method for determining ash content in pasture samples: application of near infrared reflectance spectroscopy. *Community, Soil Science and Plant Analysis* **27**: 795-802.

Vernes, K. (1995). The diet of the red-legged pademelon *Thylogale stigmatica* (Gould) (Marsupialia: Macropodidae) in fragmented tropical rainforest, north Queensland, Australia. *Mammalia* **59**: 517-525.

Vleck, D. (1978). The energy cost of burrowing by the pocket gopher *Thomomys bottae*. *Physiological Zoology* **52** 122-136.

Vogel, S., Ellington Jr., C.P. and Kilgore Jr., D.L. (1973). Wind-induced ventilation of the burrow of the Prairie-Dog, *Cynomys ludovicianus*. *Journal of Comparative Physiology* **85**: 1-14.

Volesky, J.D. and Coleman, S.W. (1996). Estimation of botanical composition of esophogeal extrusa samples using near infrared reflectance spectroscopy. *Journal of Range Management* **49**: 163-166.

Vulich, S.A. (1994). A note on the use of alternative saponification procedures for the determination of n-alkanes in faeces. *Irish Journal of Agricultural and Food Research* **33**: 71-73.

Vulich, S.A., Hanrahan, J.P. and Crowley, B.A. (1995). Modification of the analytical procedures for the determination of herbage and faecal n-alkanes used in the estimation of herbage intake. *Journal of Agricultural Science* **124**: 71-77.

Vulich, S.A., O'Riordan, E.G. and Hanrahan, J.P. (1991). Use of n-alkanes for the estimation of herbage intake in sheep: accuracy and precision of the estimates. *Journal of Agricultural Science* **116**: 319-323.

Walker, J. and Gillson, A.N. (1982). Australian Savannas. In: *Ecology of Tropical Savannas* (eds B.J. Huntley and B.H. Walker) pp 5-24. Springer-Verlag, Berlin.

Wallis de Vries, M.F. and Schippers, P. (1994). Foraging in a landscape mosaic: selection for energy and minerals in free-ranging cattle. *Oecologia* **100**: 107-117.

Ward, R.G., Smith, G.S., Wallace, J.D., Urquhart, N.S. and Shenk, J.S. (1982). Estimates of intake and quality of grazed range forage by near infrared reflectance spectroscopy. *Journal of Animal Science* **54**:399-402.

Weckerly, F.W. (1994). Selective feeding by black-tailed deer: forage quality or abundance? *Journal of Mammalogy* **75**: 905-913.

Wells, R.T. (1973) Physiological and behavioural adaptations of the Hairy-nosed wombat (*Lasiorhinus latifrons* Owen) to its arid environment. PhD thesis, University of Adelaide.

Wells, R.T. (1978a). Field observations of the Hairy-nosed Wombat (*Lasiorhinus latifrons* Owen). *Australian Wildlife Research* **5**: 299-303.

Wells, R.T. (1978b). Thermoregulation and activity rhythms in the Hairy-nosed Wombat *Lasiorhinus latifrons* (Owen) (Vombatidae). *Australian Journal of Zoology* **26**: 639-651.

Wells, R.T. (1989). Vombatidae. In: *Fauna of Australia. Mammalia* (eds D.W. Walton and B.J. Richardson) pp 755-768. Canberra: Australian Government Publishing Service Vol. 1B.

White, G.C. and Garrott, R.A. (1990). *Analysis of wildlife radio-tracking data.* Academic Press, San Diego, USA.

Wickstrom, M.L., Robbins, C.T., Hanley, T.A., Spalinger, D.E. and Parish, S.M. (1984). Food intake and foraging energetics of elk and mule deer. *Journal of Wildlife Management* **48**: 1285-1301.

Williams, W.T. (1971). Strategy and tactics in the acquisition of ecological data. *Proceedings of the Ecological Society of Australia* **6**: 57-62.

Wilson, A.D. (1974). Nutrition of sheep and cattle in Australian arid areas. In: *Studies of the Australian Arid Zone. II. Animal Production* (ed A.D. Wilson) pp 74-84. CSIRO, Australia.

Wilson, K.J. and Ellington Jr., C.P. (1978). The effects of location and design on the diffusion of respiratory gases in mammal burrows. *Journal of Theoretical Biology* **71**:73-101.

Withers, P.C. (1978). Models of diffusion-mediated gas exchange in animal burrows. *American Naturalist* **112**: 1101-1112.

Worden, K.A. and Pekins, P.J. (1995). Seasonal change in feed intake, body composition, and metabolic rate of white-tailed deer. *Canadian Journal of Zoology* **73**: 452-457.

Yamuchi, A. and Iwasa, Y. (1995). Coupling of fermentation and foraging strategies of herbivorous mammals. *Journal of Theoretical Biology* **172**: 1-11.

Yemm, E.W. and Willis, A.J. (1954). The estimation of carbohydrates in plant extracts by anthrone. *Biochemistry Journal* **57**: 508-514.

APPENDIX 1.

Near Infrared (NIR) predicted quality of plant species collected from Northern Hairy-nosed wombat habitat at Epping Forest National Park during this study.

Abbreviations - N: total nitrogen (%); WSC: water soluble carbohydrate (%); AL: acid-lignin (%); OM: organic matter; NDF: neutral detergent fibre; ADF: acid detergent fibre; IVDMD: *in vitro* dry matter digestibility. Species marked with an asterix are outliers, with the observed data collected by wet chemistry being significantly different from the NIR predicted data.

Species	Collection Date	N	WSC	AL	OM	NDF	ADF	IVDMD
Abutilon sp.	16/02/95	1.61	2.99	18.85	91.24	64.03	45.53	42.08
Abutilon sp.	17/05/95	1.33	1.65	16.44	88.23	69.6	49.56	50.15
Abutilon sp.	24/11/95	1.38	4.14	12.77	93.51	58.89	40.04	45.58
Abutilon sp.	30/11/93	2.71	3.54	13.12	76.5	43.66	29.14	63.98
Abutilon sp.	13/10/94	1.9	4.22	8.72	84.06	45.63	29.17	69.69
Aristida browniana	14/04/94	0.69	4.13	3.71	94.94	76.28	41.68	31.09
Aristida sp.	27/05/94	0.56	1.76	8.49	92.83	78.85	46.53	26.33
Aristida sp.	27/05/94	0.51	2.41	8.21	93.27	79.18	44.47	26.64
Aristida sp.	27/05/94	0.78	2.26	9.95	94.48	78.26	46.46	29.73
Aristida sp.	30/11/93	1.12	0.98	7.91	93.17	80.92	46.42	30.18
Aristida sp.	27/05/94	0.47	2.93	8.28	93.87	78.74	45.18	24.56
Aristida sp.	29/07/94	0.51	3.53	7.62	95.06	78.8	46.62	29.74
Aristida sp.	13/10/94	0.64	3.39	8.69	92.12	75.03	43.93	28.61
Aristida sp.	7/12/94	0.43	4.81	6.63	95.96	75.01	42.16	33.76
Aristida sp.	16/02/95	0.85	0.98	5.86	93.46	84.66	46.08	30.07
Aristida sp.	12/04/95	0.5	1.3	6.71	97.38	86.84	47.56	22.64
Aristida sp.	17/05/95	0.76	2.15	6.53	93.69	77.99	45.32	32.54
Aristida sp.	31/07/95	0.48	3.08	7.21	94.67	77.66	45.6	32.79
Aristida sp.	22/09/95	0.48	2.12	6.22	97.68	82.67	49.21	26.43
Aristida sp.	24/11/95	0.83	1.81	5.47	94.44	81.28	46.57	29.55
Aristida sp.	23/02/96	0.58	2.21	6.2	95.37	80.08	47.72	25.89
Bothriochloa ewartina	1/12/93	0.96	1.53	7.46	86.61	72.68	42.35	34.44
Bothriochloa ewartina	13/04/94	0.74	3.14	6.68	93.21	76.24	42.31	32.67
Bothriochloa ewartina	16/02/95	1.01	1.61	9.4	91.55	76.56	44.19	33.09
Bothriochloa ewartina	22/09/95	0.56	1.77	6.47	92.85	77.4	46.93	32.58
Brachiaria sp.	27/05/94	0.81	1.33	8.1	84.71	72.32	40.31	45.38
Brachiaria sp.	29/07/94	0.67	2.22	9.69	88.26	71.72	42.47	42.11
Brachiaria sp.	16/02/95	2.01	2.6	7.22	85.95	70.08	36.94	51.73
Brachiaria sp.	12/04/95	0.83	0.98	7.88	89.63	79.65	42.24	42.76
Brachiaria sp.	17/05/95	1.27	1.72	8.5	87.51	71.93	40.51	49.11
Brachiaria sp.	31/07/95	0.8	1.02	8.07	92.45	74.1	42.07	45.41
Brachiaria sp.	13/10/94	0.65	0.83	6.69	87.02	69.41	39.95	45.19
*Carissa ovata**	27/11/93	2.09	9.68	17.22	92.28	42.39	36.94	51.31

Appendix 1 (continued)

Species	Collection Date	N	WSC	AL	OM	NDF	ADF	IVDMD
Cenchrus ciliaris	27/10/93	1.2	4.45	5.57	91.76	67.67	35.52	48.76
Cenchrus ciliaris	27/11/93	2.15	2.64	5.95	88.58	65.78	38.79	52.06
Cenchrus ciliaris	13/10/94	0.47	3.61	4.7	88.06	66.7	37.48	46.09
Cenchrus ciliaris	7/12/94	0.58	3.28	6.59	92.91	73.98	40.89	38.76
Cenchrus ciliaris	16/02/95	1.6	1.58	7.81	85.83	73.29	41.52	45.87
Cenchrus ciliaris	12/04/95	0.59	2.08	6.65	92.36	79.55	44.14	38.33
Cenchrus ciliaris	17/05/95	0.86	3.77	5.83	91.9	74.15	35.63	44.45
Cenchrus ciliaris	31/07/95	0.73	1.62	7.03	89.37	79.5	43.07	41.13
Cenchrus ciliaris	22/09/95	0.87	2.47	6.3	92.33	77.91	40.88	44.11
Cenchrus ciliaris	24/11/95	0.69	1.75	6.79	93.83	79.07	45.27	42.08
Cenchrus ciliaris	23/02/96	0.77	4.14	6.26	93.27	74.29	36.25	40.95
Cenchrus ciliaris leaf	10/09/93	1.9	0.33	8.91	80.35	70.46	41.55	50.16
Cenchrus ciliaris leaf	26/01/94	1.57	2.06	6.45	86.63	72.8	41.49	46.44
Cenchrus ciliaris leaf	26/01/94	1.14	1.93	6.11	82.86	77.45	40.68	43.46
Cenchrus ciliaris leaf	1/03/94	1.17	2.73	6.55	88.81	75.14	41.82	45.57
Cenchrus ciliaris leaf	1/03/94	1.6	2.67	5.99	85.8	73.22	42.45	51.87
Cenchrus ciliaris leaf	13/04/94	1.94	0.56	5.37	84.32	72.36	42.28	50.75
Cenchrus ciliaris leaf	25/05/94	0.86	1.53	6.97	78.17	77.12	43.52	45.49
Cenchrus ciliaris leaf	12/10/94	0.56	1.88	6.29	87.49	73.64	42.48	42.67
Cenchrus ciliaris leaf	12/10/94	0.87	2.55	8.17	88.51	73.11	43.08	46.68
Cenchrus ciliaris leaf	16/02/95	1.8	1.06	6.06	84.46	71.18	40.55	50.41
Cenchrus ciliaris sheath	10/09/93	1.71	2.01	8.54	86.16	77.27	39.42	39.74
Cenchrus ciliaris sheath	26/01/94	0.82	1.9	4.84	85.27	75.36	40.83	43.72
Cenchrus ciliaris sheath	1/03/94	0.81	1.97	4.71	85.45	75.83	42.26	45.98
Cenchrus ciliaris sheath	13/04/94	0.85	0.34	3.94	84.96	77.88	43.7	44.94
Cenchrus ciliaris sheath	25/05/94	0.38	1.57	6.35	85.01	73.84	41.7	38
Cenchrus ciliaris stem	10/09/93	0.58	4.35	9.94	91.68	78.11	42.9	28.17
Cenchrus ciliaris stem	26/01/94	0.64	4.67	7.32	93.53	74.81	38.64	35.74
Cenchrus ciliaris stem	1/03/94	0.64	4.96	7.46	96.31	75.22	34.8	35.62
Cenchrus ciliaris stem	13/04/94	0.6	2.77	8.23	89.9	81.99	47.26	28.45
Cenchrus ciliaris stem	25/05/94	0.5	5.26	6.91	92.31	74.34	39.16	36.16
Cenchrus ciliaris stem	12/10/94	0.51	5.15	7.43	94.96	75.54	36.74	34.32
Cenchrus ciliaris stem	16/02/95	0.56	2.98	8.34	91.21	80.19	45.94	28.33
Chrysopogon fallax	13/04/94	1.21	1.18	10.81	89.5	76.25	44.74	37.41
Chrysopogon fallax	29/07/94	0.6	1.89	7.73	92.05	77.55	41.35	34.11
Chrysopogon fallax	13/10/94	0.91	2.17	8.93	90.09	70.82	39.67	37.1
Chrysopogon fallax	13/10/94	0.83	3.09	10.35	91.81	71.32	41.32	35.08
Chrysopogon fallax	16/02/95	2.28	1.11	11.34	91.83	75.17	40.84	37.53
Chrysopogon fallax	12/04/95	0.75	0.9	9.15	92.98	82.93	48.25	32.35
Chrysopogon fallax	17/05/95	0.6	1.42	8.9	92.98	77.95	45.38	32.98
Chrysopogon fallax	31/07/95	0.72	1.4	8.58	91.12	77.54	43.62	33.03
Chrysopogon fallax	22/09/95	1.12	1.51	8.5	94.3	74.49	41.31	38.37
Chrysopogon fallax	24/11/95	0.81	0.16	5.95	95.88	79.49	44.11	29.98
Chrysopogon fallax	23/02/96	0.92	3.46	5.78	93.04	74.55	40.24	32.13

Appendix 1 (continued)

Species	Collection Date	N	WSC	AL	OM	NDF	ADF	IVDMD
Dactyloctenium radulans	13/04/94	1.66	2.51	7	87.61	62.85	35.27	57.66
Enneapogon sp.	27/05/94	0.66	2.11	9.56	93.6	78.49	45.67	31.57
Enneapogon sp.	24/05/94	0.91	1.56	8.18	90.21	76.09	41.76	36.51
Enneapogon sp.	27/05/94	0.68	2.52	9.19	94.84	76.45	45.21	32.71
Enneapogon sp.	29/07/94	0.45	1.88	6.44	98.36	80.86	46.66	29.42
Enneapogon sp.	13/10/94	0.32	1.75	6.92	96.19	75.77	46.55	35.19
Enneapogon sp.	7/12/94	0.34	2.86	7.59	95.96	76.19	46.89	31.75
Enneapogon sp.	16/02/95	1.14	1.14	7.4	95.67	80.52	44.05	31.73
Enneapogon sp.	12/04/95	0.78	0.43	7.32	94.24	81.77	49.82	30.49
Enneapogon sp.	17/05/95	0.57	1.11	7.44	94.24	81.76	48.14	28.81
Enneapogon sp.	31/07/95	0.51	1.22	6.56	97.61	81.99	49.29	27.06
Enneapogon sp.	22/09/95	0.49	1.34	6.8	99.27	81.57	48.07	28.61
Enneapogon sp.	24/11/95	1.11	1.46	5.47	94.78	81.18	45.49	36.41
Enneapogon sp.	23/02/96	0.68	4.48	6.93	96.64	73.51	42.78	32.73
Eragostris lacunaria	30/11/93	0.5	1.08	8.33	83.99	76.59	43.99	35.25
Eragostris lacunaria	29/07/94	0.73	3.49	10.84	85.12	74.51	42.3	29.7
Eragostris lacunaria	12/10/94	0.77	3.43	13.21	81.16	73.63	42.6	28.65
Eragostris lacunaria	7/12/94	0.78	3.71	10.65	92.03	73.68	42.21	32.03
Eragostris lacunaria	16/02/95	1.55	1.8	11.02	92.73	77.2	43.16	29.39
Eragostris lacunaria	12/04/95	0.84	2.24	8.56	93.46	79.29	44.05	25.71
Eragostris lacunaria	17/05/95	0.87	3.39	8.09	93.61	74.82	40.79	31.51
Eragostris lacunaria	31/07/95	0.73	3.93	7.36	93.95	76.09	40.1	30.72
Eragostris lacunaria	22/09/95	0.83	2.54	9.24	94.52	78.13	41.69	26.72
Eragostris lacunaria	24/11/95	0.95	2.71	6.63	93	77.87	39.78	31.26
Eragostris lacunaria	23/02/96	0.79	5.94	7.08	94.82	72.28	37.78	31.97
Eragostris schultzii	25/05/94	0.53	2.58	8.65	90.22	78.36	45.2	33.45
Fimbristylis dichotoma	30/11/93	2.52	1.16	12.06	86.57	71.12	35.94	54.28
Fimbristylis dichotoma	27/05/94	1.69	2.06	13.84	88.3	66.35	38.08	48.42
Fimbristylis dichotoma	29/07/94	1.14	4.53	8.44	90.01	63.26	34.16	46.86
Fimbristylis dichotoma	13/10/94	1.3	2.25	9.46	89.8	67.71	45.69	48.22
Fimbristylis dichotoma	7/12/94	1.37	1.11	7.66	87.04	66.58	43.94	45.52
Fimbristylis dichotoma	16/02/95	1.95	1.23	13.45	81.59	76.47	44.29	42.52
Fimbristylis dichotoma	12/04/95	2.3	0.72	13.25	89.25	71.33	39.11	53.08
Fimbristylis dichotoma	17/05/95	1.57	1.9	13.52	84.15	65.42	39.88	48.75
Fimbristylis dichotoma	31/07/95	1.42	1.46	11.2	83.84	66.45	40.53	45.75
Fimbristylis dichotoma	22/09/95	1.47	2.13	12.13	80.1	65.82	42.86	48.79
Fimbristylis dichotoma	24/11/95	1.54	3.08	7.2	90.29	67.48	35.58	46.1
Fimbristylis dichotoma	23/02/96	1.24	6.79	9.04	91.53	57.3	30.44	54.1
Fimbristylis neilsonii	27/05/94	0.85	1.58	15	87.05	73.27	46.42	37.11
Helichrysum sp*.	30/11/93	2.49	1.62	18.67	83.15	64.33	47.22	52.00
Heteropogon contortus	13/04/94	0.7	2.85	7.52	90.55	73.99	42.22	33.56
Heteropogon contortus	29/07/94	0.4	5	5.61	94.22	72.97	41.67	38.71
Heteropogon contortus	13/10/94	0.28	4.64	6.77	94.32	71.64	41.92	35.31
Heteropogon contortus	7/12/94	0.27	5.89	6.27	95.96	71.38	44.65	37.83

Appendix 1 (continued)

Species	Collection Date	N	WSC	AL	OM	NDF	ADF	IVDMD
Heteropogon contortus	24/11/95	0.6	2.02	6.29	94.44	77.02	45.66	35.11
Heteropogon contortus	23/02/96	0.63	3.52	5.87	93.32	71.32	41.16	35.23
Malvaceae	29/07/94	1.43	3.45	9.7	87.95	55.64	36.35	55.47
Malvaceae	31/07/95	1.11	1.77	9.87	89.4	63.28	40.72	47.99
Malvaceae - *Abutilon*	22/09/95	1.36	5.62	13.96	93.88	56.27	37.97	45.05
Malvaceae - *Abutilon*	23/02/96	1.48	6.23	12.97	92.83	54.05	36.92	41.91
Melinis repens	14/04/94	0.74	3.9	4.01	93.72	72.55	41.27	39.85
Perotis rara	30/11/93	2.19	0.84	11.73	79.92	76.57	48.29	41.45
Perotis rara	13/04/94	0.8	1.85	6.24	92.04	78.59	43.67	30.86
Perotis rara	29/07/94	0.75	1.98	7.84	0.04	79.27	44.4	35.46
Perotis rara	12/04/95	1.42	1.27	8.91	88.77	76.41	43.52	37.68
Perotis rara	31/07/95	0.55	1.56	8.28	90.82	71.99	56.09	32.92
Salsola kalii	27/11/93	2.35	1.57	7.26	79.92	48.56	38.47	69.06
Salsola kalii	29/07/94	1.02	3.05	7.96	75.03	54.21	40.39	60.33
*Salsola kalii**	22/09/95	2.92	1.67	5.51	73.40	46.69	36.26	84.53
*Salsola kalii**	24/11/95	2.05	1.96	4.46	81.84	51.93	35.53	78.93
Salsola kalii	23/02/96	2.27	5.16	4.85	81.16	42.02	35.49	73.66
Sida sp.	7/12/94	1.94	5.82	7.39	86.41	37.38	24.1	57.53
Sida sp.	12/04/95	0.69	0.66	11.19	95.75	75.89	51.28	31.18
Sida sp.	24/11/95	1.63	1.91	9.76	91.56	55.82	36.49	47.2
Themeda triandra	13/10/94	0.32	2.99	8.06	88.92	70.81	41.23	35.34
Themeda triandra	14/04/94	0.81	2.62	5.04	90.43	73.09	41.21	37.77
Themeda triandra	7/12/94	0.41	4.78	5.82	89.68	67.55	38.4	39.29
Tribulus terrestris	27/11/93	2.5	2.41	12.09	75.24	54.91	38.17	65.13
Triraphis mollis	25/05/94	0.35	2.25	7.17	97.53	79.33	50.36	31.02
Triraphis mollis	29/07/94	0.49	1.4	7.43	97.81	84.59	51.58	28.63
Triraphis mollis	24/11/95	1.04	2.66	5.99	94.03	76.61	46.7	38.4
Waltheria indica	13/04/94	1.8	8.52	13.48	90.56	50.75	35.21	50.19
Waltheria indica	29/07/94	1.59	8.4	12.38	95.31	52.69	35.69	46.23
Waltheria indica	13/10/94	1.59	7.02	11.09	92.22	50.62	33	48.71
Waltheria indica	7/12/94	2.05	8.34	12.41	94.81	45.77	30.79	46.94
Waltheria indica	16/02/95	2.29	2.68	19.71	91.32	59.1	43.86	47.95
Waltheria indica	12/04/95	0.94	4.78	14.44	95.61	62.18	45.94	34.21

APPENDIX 2

Principal Component Analysis of Plant Species by Long-chain Alkane Analysis

Figure A2.1. First two principal components of long-chain alkane concentrations for plants collected in a) November 1993; b) April 1994; c) May 1994; d) July 1994. Species Abreviations are: Ab - *Abutilon* spp.; Ar - *Aristida* spp. (and species A, B, C & D); Br - *Brachiaria* sp.; Bo - *Bothriochloa ewartina*; Ca - *Carissa ovata*; Ce - *Cenchrus ciliaris*; Ch - *Chrysopogon fallax*; Da - *Dactyloctenium radulans*; En - *Enneapogon* spp. (and species 2 & X); Er - *Eragostris lacunaria;* Fi - *Fimbristylis* sp.; He - *Heteropogon contortus*; Hl - *Helichrysum* sp.; Ma - Malvaceae; Pe - *Perotis rara*; Sa - *Salsola kalii*; Si - *Sida* sp.; Tb - *Tribulus terrestris;* Th - *Themeda triandra*; Tr - *Triraphis mollis*; Wa - *Waltheria indica.*

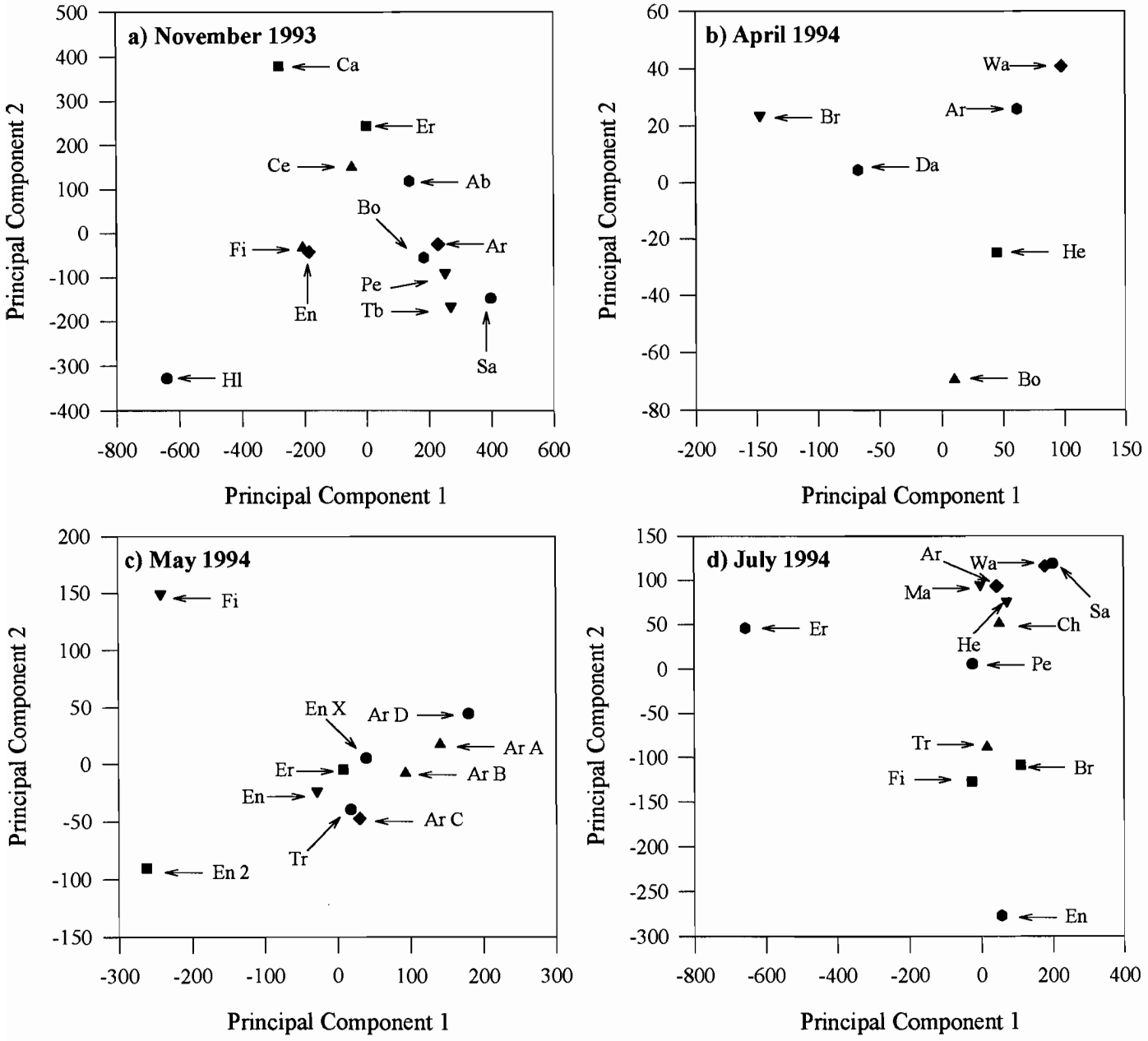


Figure A2.1 (continued). First two principal components of long-chain alkane concentrations for plants collected in e) October 1994; f) December 1994; g) February 1995; h) April 1995.
Species Abreviations are: Ab - *Abutilon* spp.; Ar - *Aristida* spp. (and species A, B, C & D); Br - *Brachiaria* sp.; Bo - *Bothriochloa ewartina*; Ca - *Carissa ovata*; Ce - *Cenchrus ciliaris*; Ch - *Chrysopogon fallax*; Da - *Dactyloctenium radulans*; En - *Enneapogon* spp. (and species 2 & X); Er - *Eragostris lacunaria;* Fi - *Fimbristylis* sp.; He - *Heteropogon contortus*; Hl - *Helichrysum* sp.; Ma - Malvaceae; Pe - *Perotis rara*; Sa - *Salsola kalii*; Si - *Sida* sp.; Tb - *Tribulus terrestris;* Th - *Themeda triandra*; Tr - *Triraphis mollis*; Wa - *Waltheria indica.*

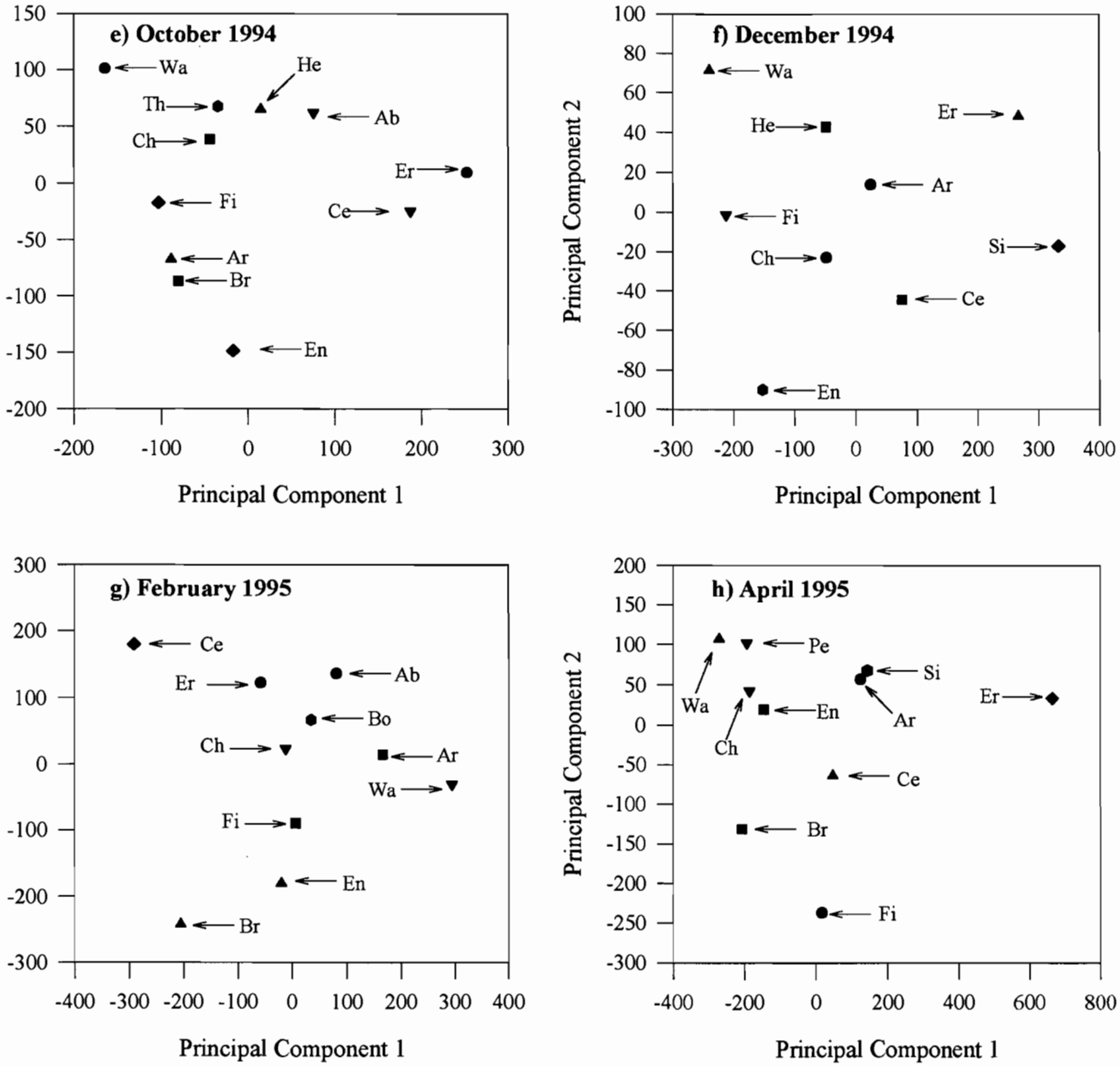


Figure A2.1 (continued). First two principal components of long-chain alkane concentrations for plants collected in i) May 1995; j) July 1995; k) September 1995; l) November 1995.

Species Abreviations are: Ab - *Abutilon* spp.; Ar - *Aristida* spp. (and species A, B, C & D); Br - *Brachiaria* sp.; Bo - *Bothriochloa ewartina*; Ca - *Carissa ovata*; Ce - *Cenchrus ciliaris*; Ch - *Chrysopogon fallax*; Da - *Dactyloctenium radulans*; En - *Enneapogon* spp. (and species 2 & X); Er - *Eragostris lacunaria;* Fi - *Fimbristylis* sp.; He - *Heteropogon contortus*; Hl - *Helichrysum* sp.; Ma - Malvaceae; Pe - *Perotis rara*; Sa - *Salsola kalii*; Si - *Sida* sp.; Tb - *Tribulus terrestris;* Th - *Themeda triandra*; Tr - *Triraphis mollis*; Wa - *Waltheria indica.*

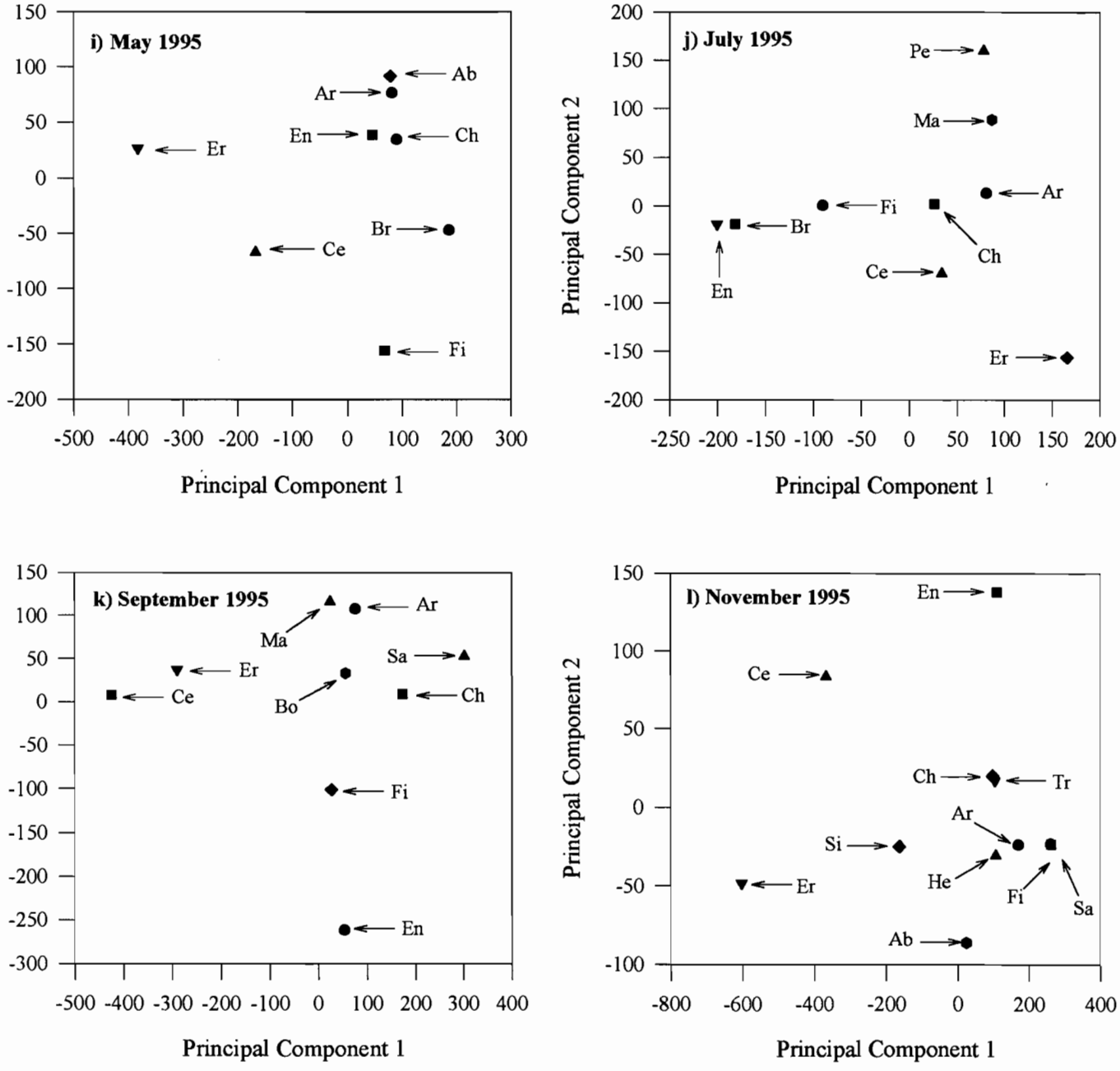
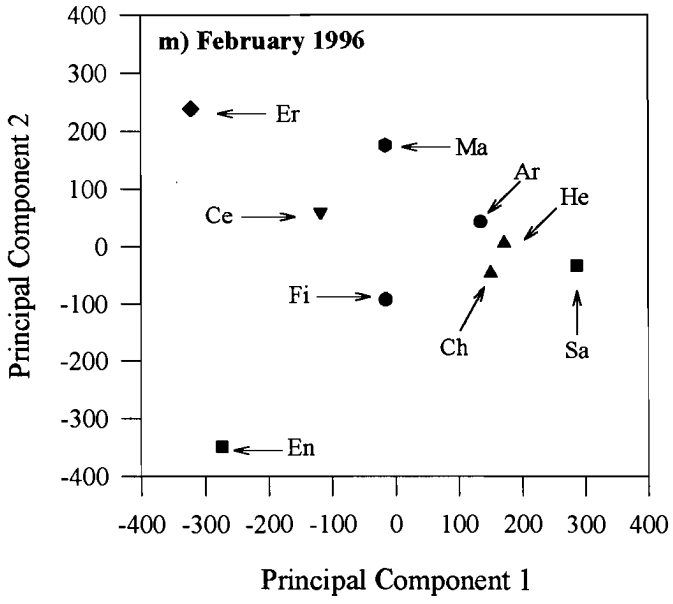
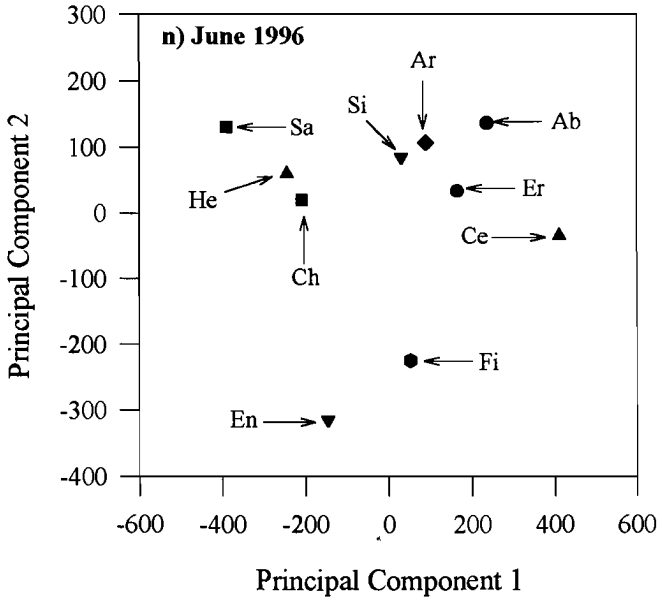


Figure A2.1 (continued). First two principal components of long-chain alkane concentrations for plants collected in m) February 1996; n) June 1996. Species Abreviations are: Ab - *Abutilon* spp.; Ar - *Aristida* spp. (and species A, B, C & D); Br - *Brachiaria* sp.; Bo - *Bothriochloa ewartina*; Ca - *Carissa ovata*; Ce - *Cenchrus ciliaris*; Ch - *Chrysopogon fallax*; Da - *Dactyloctenium radulans*; En - *Enneapogon* spp. (and species 2 & X); Er - *Eragostris lacunaria;* Fi - *Fimbristylis* sp.; He - *Heteropogon contortus*; Hl - *Helichrysum* sp.; Ma - Malvaceae; Pe - *Perotis rara*; Sa - *Salsola kalii*; Si - *Sida* sp.; Tb - *Tribulus terrestris;* Th - *Themeda triandra*; Tr - *Triraphis mollis*; Wa - *Waltheria indica.*





APPENDIX 3

Temporal variation in the diets of the northern hairy-nosed wombat and the eastern grey kangaroo reconstructed by long-chain alkane analysis.

Table A3.1. Mean temporal trends in diet composition of the NHN wombat predicted by EatWhat using the first method; i.e. all plant species and all alkanes were used in the EatWhat prediction. Diet composition expressed as a proportion of 1.00.

Plant Species	Nov-93	Apr-94	May-94	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma	0.00		0.04	0.37	0.02	0.28	0.77	0.05	0.16	0.00	0.01	0.00	0.00	0.27
Aristida spp.	0.34	0.31	0.55	0.03	0.05	0.23	0.00	0.05	0.04	0.15	0.00	0.00	0.03	0.27
Bothriochloa ewartina	0.00	0.01					0.00				0.03			
Brachiaria spp.		0.12		0.07	0.00		0.17	0.46	0.30	0.00				
Cenchrus ciliaris	0.00				0.05	0.04	0.00	0.00	0.00	0.12	0.03	0.00	0.01	0.00
Chrysopogon fallax				0.03	0.04	0.04	0.03	0.14	0.00	0.01	0.00	0.25	0.47	0.06
Dactlyoctenium radulans		0.52												
Enneapogon spp.	0.00		0.26	0.13	0.06	0.01	0.03	0.00	0.00	0.59	0.60	0.65	0.22	0.08
Eragostris lacunaria	0.10		0.03	0.11	0.32	0.31	0.00	0.01	0.00	0.12	0.14	0.01	0.02	0.00
Heteropogon contortus		0.02		0.12	0.00	0.00						0.00	0.01	0.04
Perotis rara	0.16			0.10				0.00		0.01				
Themeda triandra					0.00									
Triraphis mollis			0.01	0.02								0.00		
Carissa ovata	0.00													
Helichrysum sp.	0.31													
Salsola kalii	0.07			0.00							0.00	0.10	0.11	0.02
Malvaceae				0.00						0.00	0.19		0.03	
Abutilon sp.	0.11				0.00		0.00		0.50			0.00		0.12
Sida sp.						0.01		0.00				0.00		0.13
Waltheria indica		0.03		0.01	0.46	0.09	0.00	0.16						
Tribulus terrestris	0.05													

Table A3.2. Mean temporal trends in diet composition of the NHN wombat predicted by EatWhat using the second method: selection of plant species using spatial separation of the first and second principal components. Diet composition expressed as a proportion of 1.00.

Plant Species	Nov-93	Apr-94	May-94	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma			0.01	0.42	0.00	0.27	0.80	0.05	0.31	0.04	0.61		0.00	0.04
Aristida spp.	0.67	0.61	0.79	0.55	0.01	0.03	0.00	0.12		0.20	0.21	0.00	0.29	0.34
Bothriochloa ewartina		0.00												
Brachiaria spp.		0.32					0.19	0.81	0.12					
Cenchrus ciliaris	0.00				0.65	0.15	0.01	0.00	0.05	0.15	0.01	0.00	0.00	0.00
Chrysopogon fallax					0.00							0.26		0.60
Enneapogon spp.	0.01		0.20	0.08	0.29	0.05	0.00	0.00	0.51	0.54	0.23	0.72	0.49	0.00
Eragostris lacunaria				0.00		0.43	0.00	0.01	0.01	0.07		0.01	0.01	
Heteropogon contortus		0.07				0.03								
Perotis rara														
Themeda triandra														
Triraphis mollis														
Carissa ovata	0.00													
Helichrysum sp.	0.32													
Salsola kalii														
Malvaceae										0.01			0.21	
Abutilon sp.												0.02		
Sida sp.						0.00						0.00		
Waltheria indica					0.29	0.05								
Tribulus terrestris														

Table A3.3. Mean temporal trends in diet composition of the NHN wombat predicted by EatWhat using the third method: selection of plant species using spatial separation of the first and second principal components, and considering only C_4 plants and plants detected by microscopic analysis. Diet composition expressed as a proportion of 1.00.

Plant Species	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma	0.49	0.00	0.21	0.79	0.99	0.42	0.05	0.08	0.13	0.00	0.05
Aristida spp.	0.35	0.00	0.01	0.00	0.00	0.24	0.20	0.04	0.00	0.59	0.34
Bothriochloa ewartina											
Brachiaria spp.											
Cenchrus ciliaris		0.32	0.19	0.00	0.00	0.00	0.15	0.20	0.00	0.01	0.00
Chrysopogon fallax	0.13	0.35	0.12	0.01			0.00	0.00	0.24		0.60
Dactlyoctenium radulans											
Enneapogon spp.	0.03	0.08	0.02	0.20	0.01	0.34	0.54	0.61	0.63	0.39	0.02
Eragostris lacunaria	0.00	0.25	0.45	0.00	0.00	0.00	0.07	0.07	0.01	0.00	0.00
Heteropogon contortus											
Perotis rara											
Themeda triandra											
Triraphis mollis											
Carissa ovata											
Helichrysum sp.											
Salsola kalii											
Malvaceae											
Abutilon sp.											
Sida sp.											
Waltheria indica											
Tribulus terrestris											

Table A3.4. Mean temporal trends in diet composition of the Eastern Grey Kangaroo predicted by EatWhat using the first method: All plant species and all alkanes were used in the EatWhat prediction. Diet composition expressed as a proportion of 1.00.

Plant Species	Nov-93	Apr-94	May-94	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma	0.14		0.07	0.24	0.06	0.26	0.13	0.32	0.00	0.09	0.05	0.00	0.00	0.00
Aristida spp.	0.00	0.29	0.42	0.00	0.07	0.32	0.00	0.05	0.25	0.00	0.00	0.00	0.05	0.27
Bothriochloa ewartina	0.00	0.00					0.00				0.04			
Brachiaria spp.		0.07		0.02	0.00		0.06	0.40	0.51	0.00				
Cenchrus ciliaris	0.20				0.03	0.08	0.06	0.03	0.00	0.00	0.15	0.00	0.00	0.00
Chrysopogon fallax				0.08	0.09	0.02	0.16	0.00	0.00	0.13	0.26	0.46	0.70	0.00
Dactlyoctenium radulans		0.55												
Enneapogon spp.	0.03		0.19	0.35	0.09	0.18	0.18	0.01	0.00	0.38	0.31	0.47	0.16	0.31
Eragostris lacunaria	0.09		0.13	0.12	0.30	0.02	0.35	0.00	0.00	0.22	0.141	0.00	0.00	0.00
Heteropogon contortus		0.09		0.02	0.00	0.00						0.04	0.00	0.16
Perotis rara	0.02			0.06				0.04		0.09				
Themeda triandra					0.00									
Triraphis mollis			0.19	0.11								0.00		
Carissa ovata	0.00													
Helichrysum sp.	0.34													
Salsola kalii	0.00			0.00							0.00	0.02	0.00	0.00
Malvaceae				0.00						0.09	0.08		0.08	
Abutilon sp.	0.17				0.07		0.06		0.24			0.00		0.09
Sida sp.						0.04		0.00				0.00		0.17
Waltheria indica		0.00		0.00	0.28	0.06	0.00	0.14						
Tribulus terrestris	0.00													

Table A3.5. Mean temporal trends in diet composition of the Eastern Grey Kangaroo predicted by EatWhat using the second method: selection of plant species using spatial separation of the first and second principal components. Diet composition expressed as a proportion of 1.00.

Plant Species	Nov-93	Apr-94	May-94	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma			0.03	0.48	0.00	0.27	0.31	0.29	0.00	0.32	0.54		0.00	0.00
Aristida spp.	0.20	0.63	0.82	0.36	0.11	0.34	0.22	0.16		0.06	0.27	0.00	0.30	0.09
Bothriochloa ewartina		0.00												
Brachiaria spp.		0.29					0.00	0.55	0.56					
Cenchrus ciliaris	0.37				0.59	0.18	0.11	0.00	0.00	0.00	0.06	0.00	0.00	0.00
Chrysopogon fallax					0.00							0.46		0.91
Dactlyoctenium radulans														
Enneapogon spp.	0.14		0.15	0.16	0.06	0.14	0.13	0.00	0.39	0.24	0.13	0.50	0.46	0.00
Eragostris lacunaria				0.01		0.00	0.23	0.00	0.05	0.12		0.00	0.00	
Heteropogon contortus		0.09				0.03								
Perotis rara														
Themeda triandra														
Triraphis mollis														
Carissa ovata	0.00													
Helichrysum sp.	0.29													
Salsola kalii														
Malvaceae										0.25			0.24	
Abutilon sp.												0.04		
Sida sp.						0.01						0.00		
Waltheria indica					0.24	0.00								
Tribulus terrestris														

Table A3.6. Mean temporal trends in diet composition of the Eastern Grey Kangaroo predicted by EatWhat using the third method: selection of plant species using spatial separation of the first and second principal components, and considering only C_4 plants and plants detected by microscopic analysis. Diet composition expressed as a proportion of 1.00.

Plant Species	Jul-94	Oct-94	Dec-94	Feb-95	Apr-95	May-95	Jul-95	Sep-95	Nov-95	Feb-96	Jun-96
Fimbristylis dichotoma	0.51	0.00	0.10	0.26	0.89	0.45	0.28	0.16	0.15	0.00	0.00
Aristida spp.	0.29	0.00	0.14	0.22	0.11	0.00	0.40	0.00	0.00	0.64	0.09
Bothriochloa ewartina											
Brachiaria spp.											
Cenchrus ciliaris		0.19	0.38	0.07	0.00	0.55	0.00	0.25	0.00	0.00	0.00
Chrysopogon fallax	0.05	0.45	0.29	0.08			0.12	0.25	0.44		0.91
Dactlyoctenium radulans											
Enneapogon spp.	0.14	0.14	0.08	0.13	0.00	0.00	0.20	0.28	0.41	0.36	0.00
Eragostris lacunaria	0.01	0.22	0.01	0.24	0.00	0.00	0.00	0.06	0.010	0.00	0.00
Heteropogon contortus											
Perotis rara											
Themeda triandra											
Triraphis mollis											
Carissa ovata											
Helichrysum sp.											
Salsola kalii											
Malvaceae											
Abutilon sp.											
Sida sp.											
Waltheria indica											
Tribulus terrestris											

APPENDIX 4

Profiles of Activity and Temperature for Individual NHN Wombats

Figure A4.1. Daily trends of activity for Male #32. Activity represents the cummulative activity over a fifteen minute period recorded by the data logger, that is, each cummulative measure of activity represents three individual activity measurements. Twenty-four hour time period is from 12 pm on the 16th to 12 pm on the17th of each month in 1995.

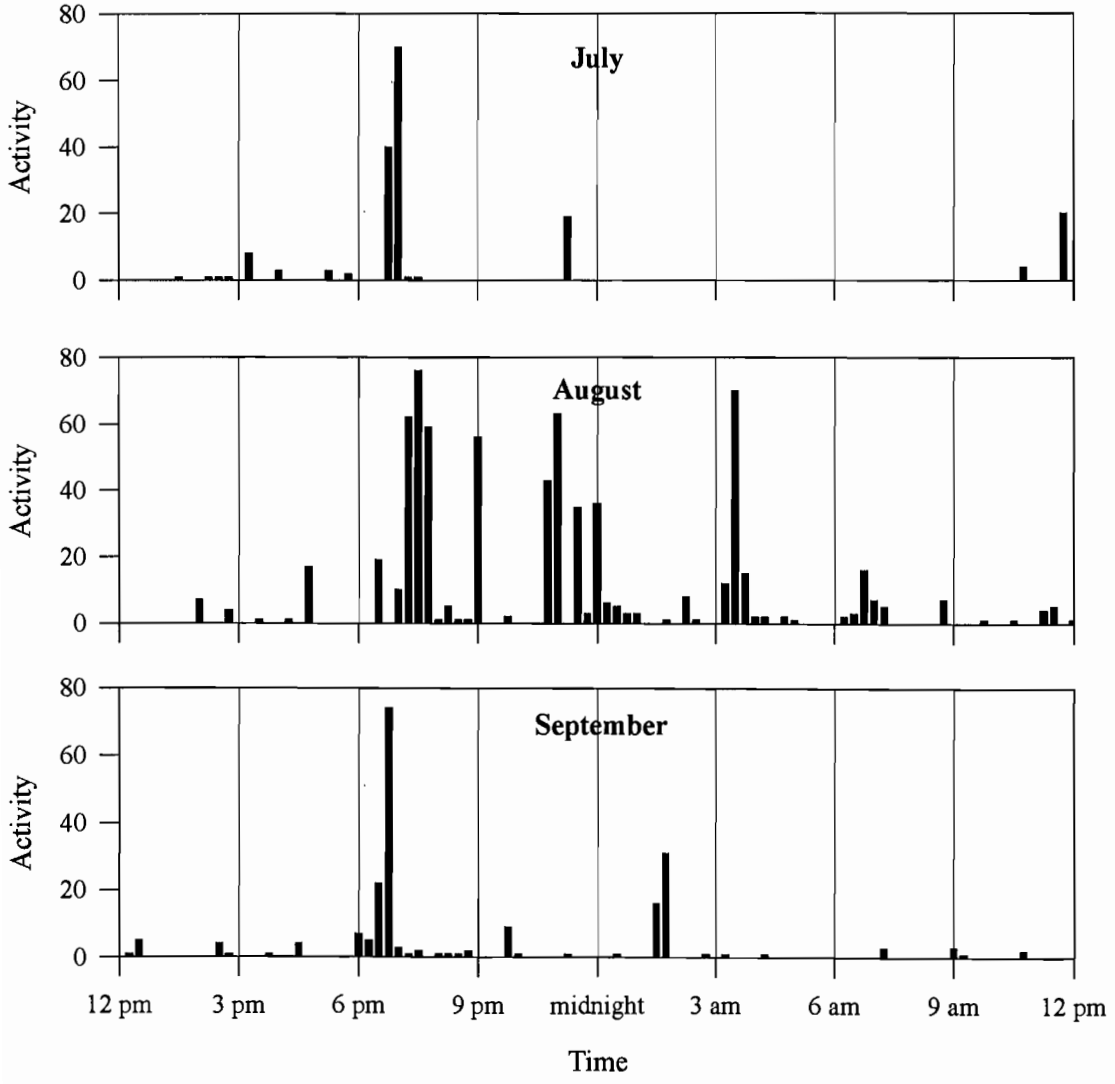


Figure A4.2. Daily trends in temperature from the data logger attached to Male #32. Temperature records are from 12 pm on the 16th to 12 pm on the 17th of each month, and represent the mean temperature for a fifteen minute period.

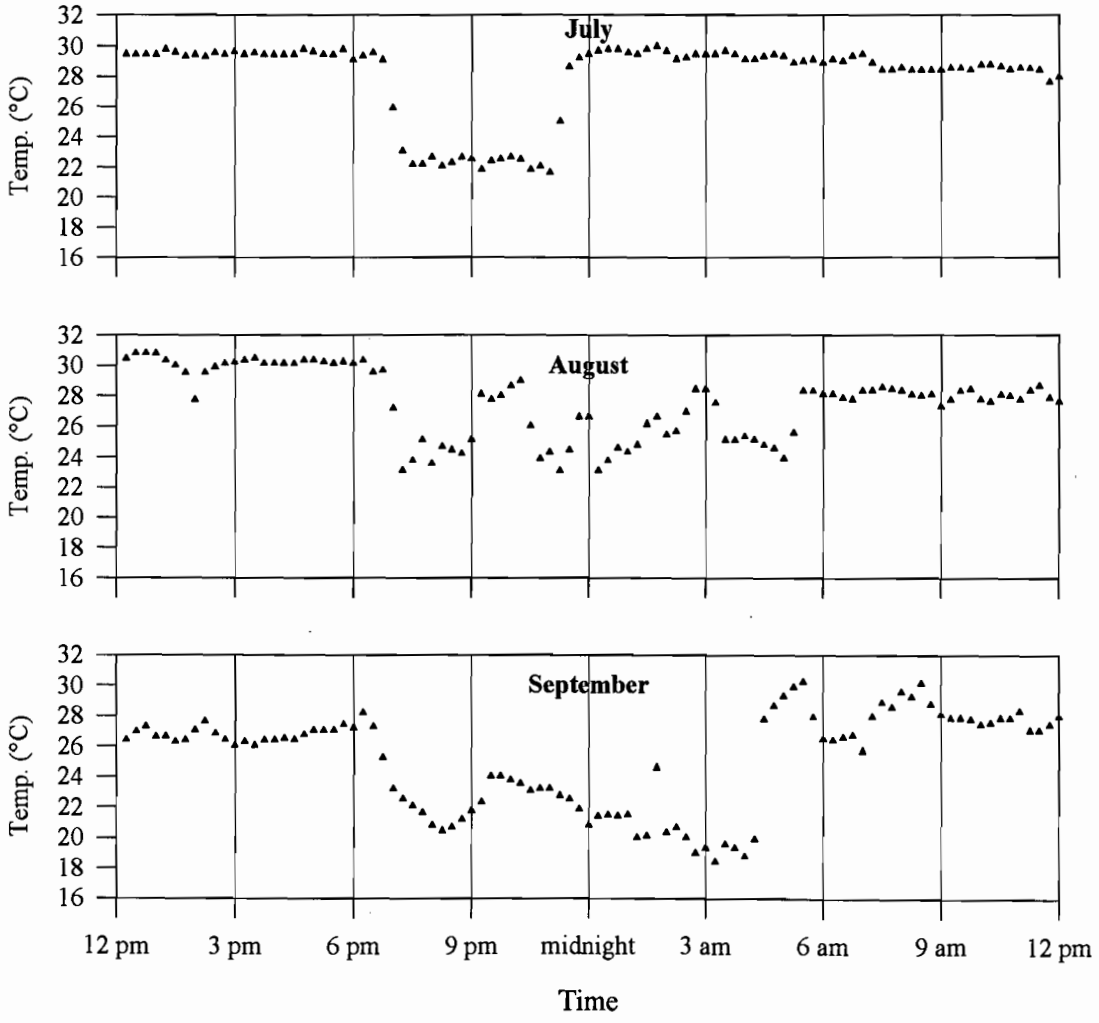


Figure A4.3. Daily trends of activity for Male #41. Activity represents the cummulative activity over a fifteen minute period recorded by the data logger, that is, each cummulative measure of activity represents three individual activity measurements. Twenty-four hour time period is from 12 pm on the 16th to 12 pm on the17th of each month in 1996.

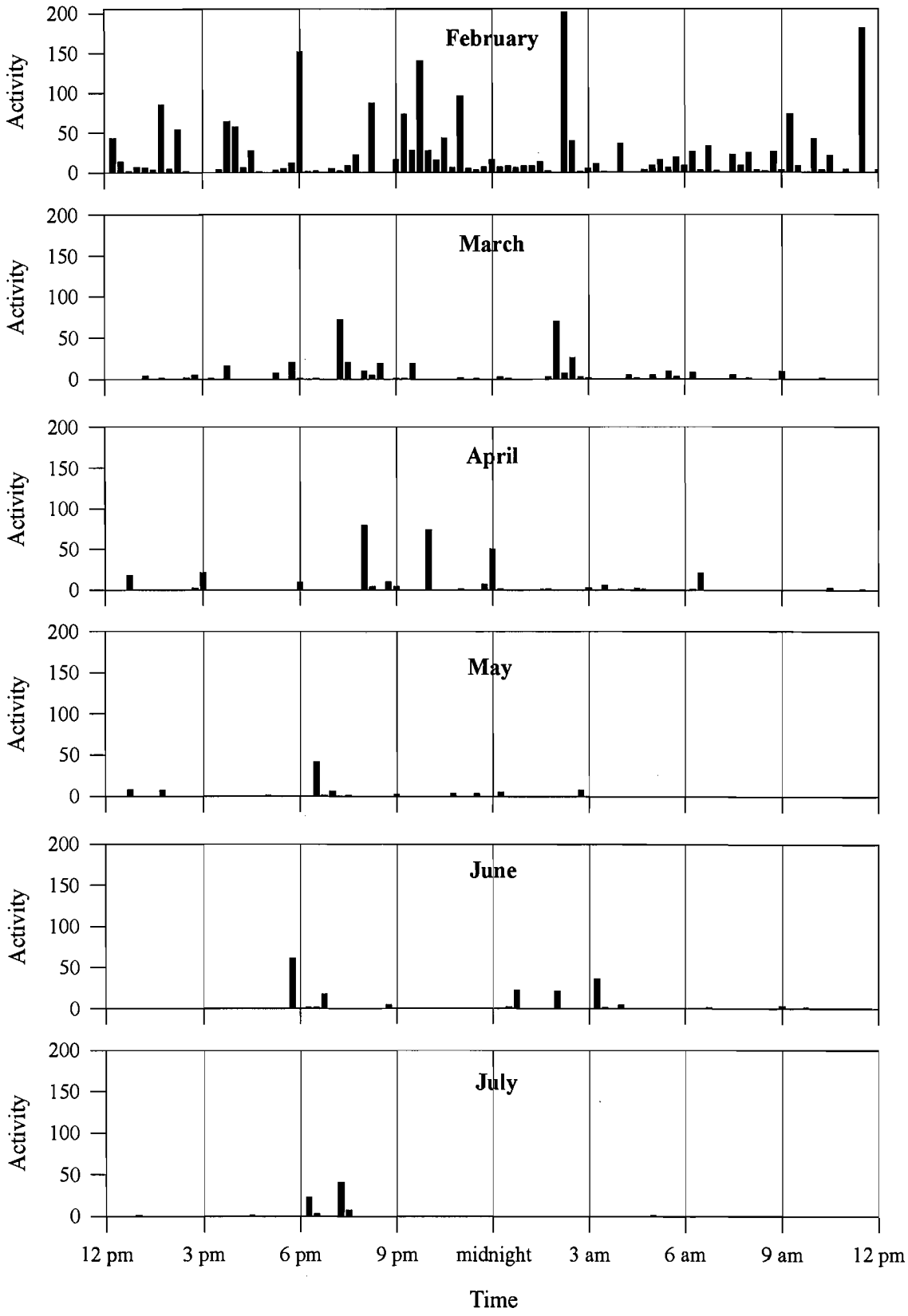


Figure A4.4. Daily trends in temperature from the data logger attached to Male #41. Temperature records are from 12 pm on the 16th to 12 pm on the 17th of each month, and represent the mean temperature for a fifteen minute period.

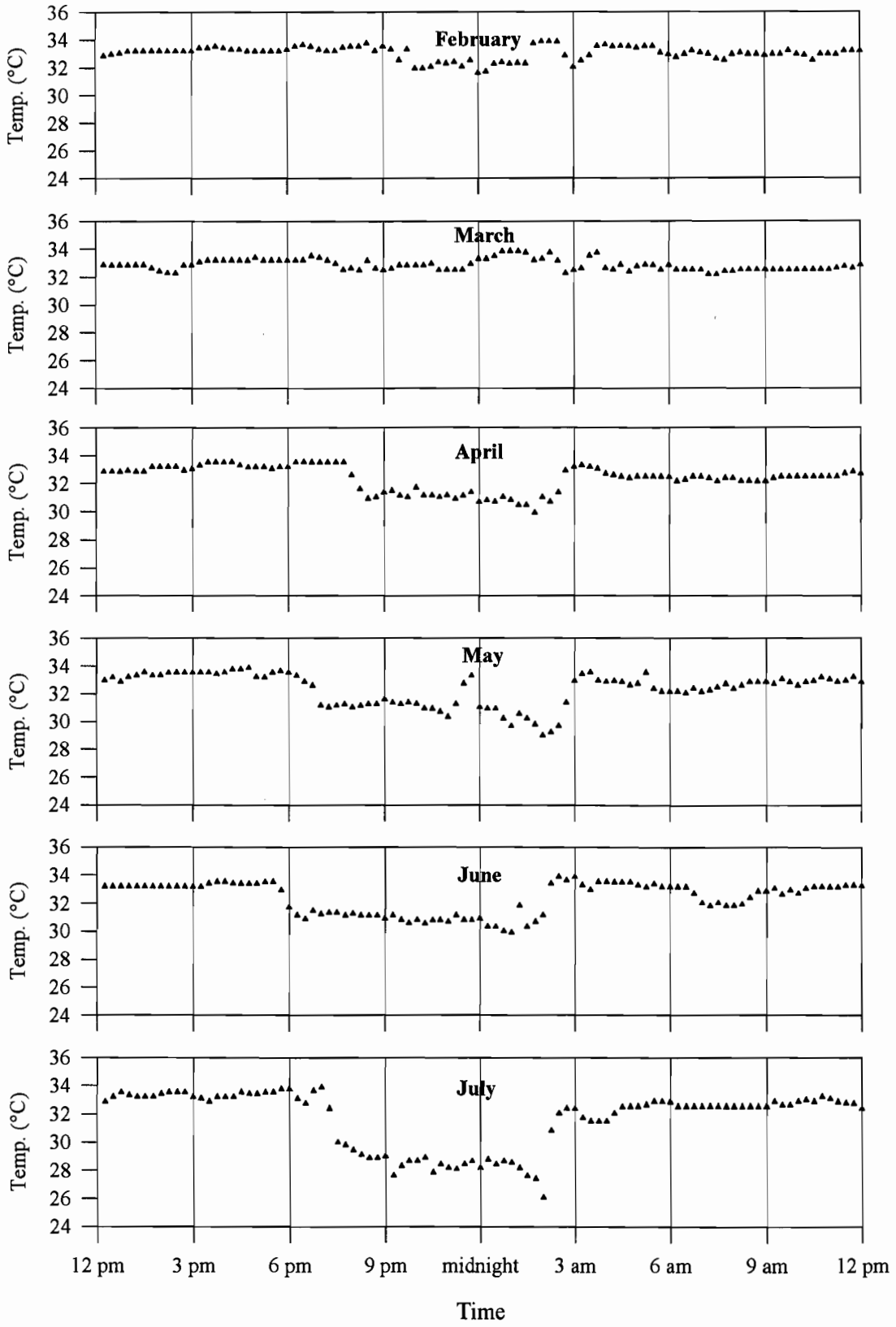


Figure A4.5. Daily trends of activity for Female #61. Activity represents the cummulative activity over a fifteen minute period recorded by the data logger, that is, each cummulative measure of activity represents three individual activity measurements. Twenty-four hour time period is from 12 pm on the 16th to 12 pm on the17th of each month in 1996.

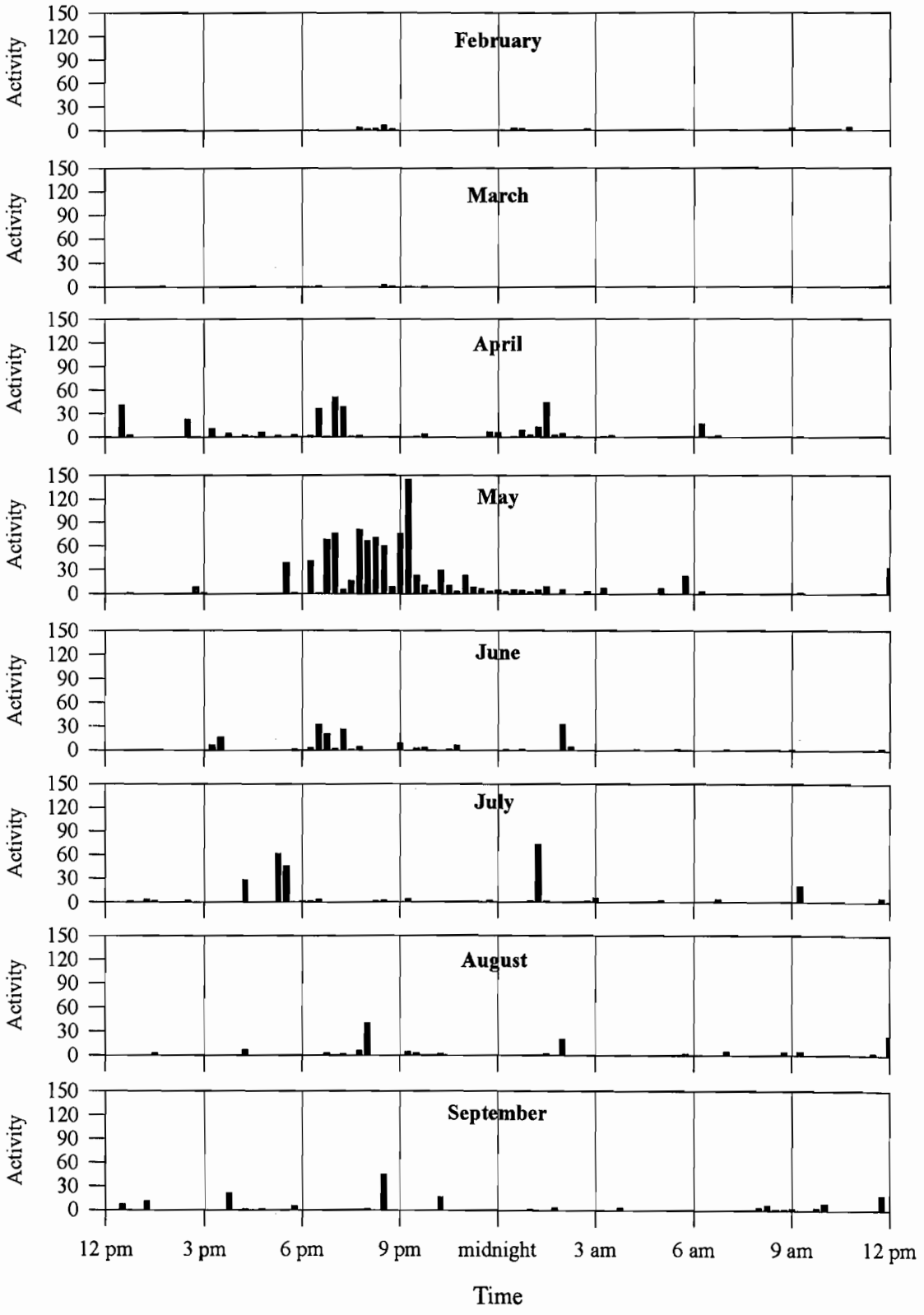
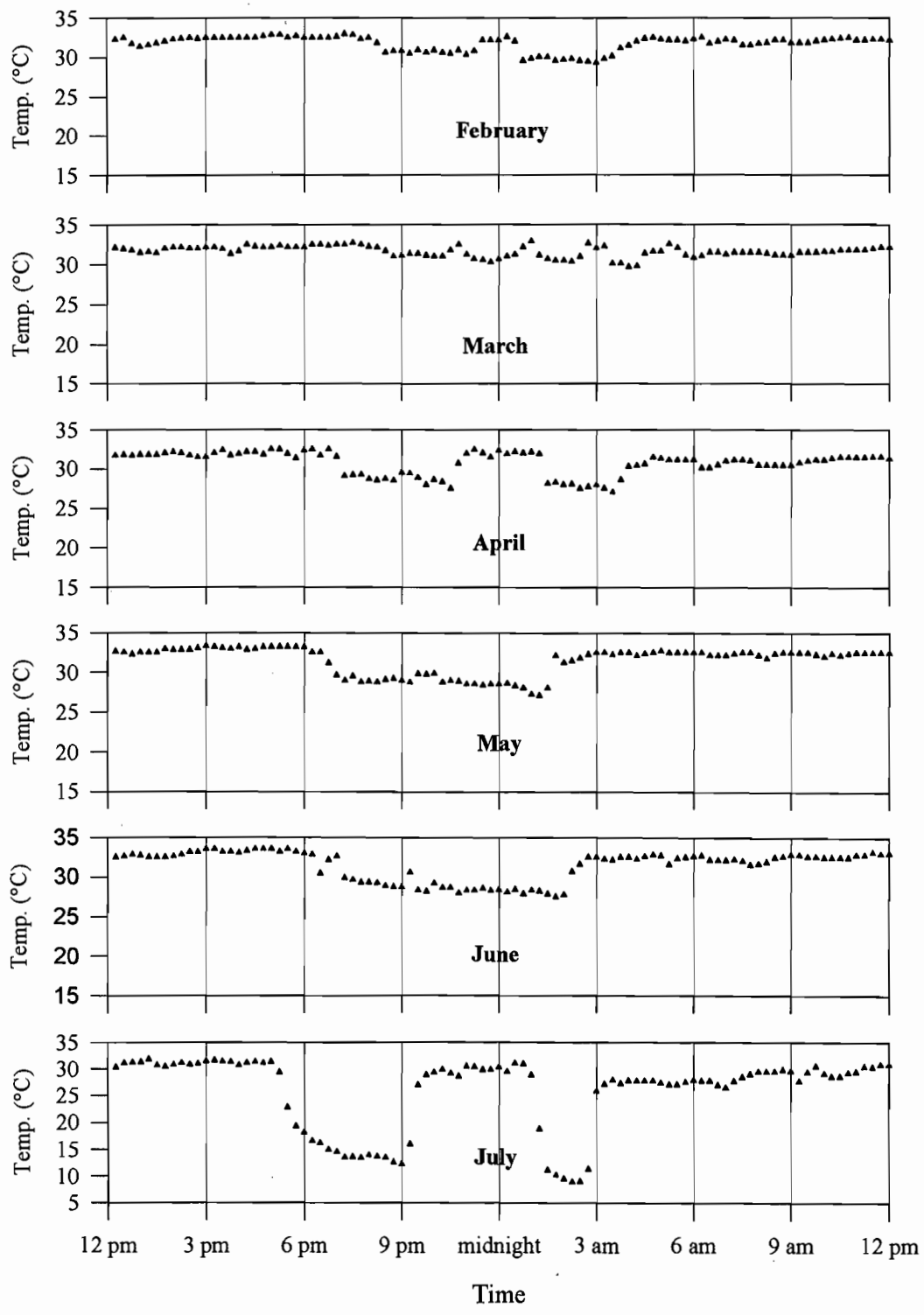


Figure A4.6. Daily trends in temperature from the data logger attached to Female #61. Temperature records are from 12 pm on the 16th to 12 pm on the 17th of each month, and represent the mean temperature for a fifteen minute period.



APPENDIX 5

RESEARCH AND MANAGEMENT RECOMMENDATIONS FOR THE NORTHERN HAIRY-NOSED WOMBAT

This Appendix describes suggested research and management recommendations based on the findings of this thesis. These include:

- **Forage Improvement Practices**

Irrigation Experiments

Chapter 2 demonstrated a positive correlation between forage quality and rainfall, specifically in crude protein concentration. Low crude protein concentrations of tropical forages during the dry season can have a detrimental impact on animal production (Owen-Smith 1982). Since the aim of the recovery process is to increase the population size of NHN wombats, implementing a scheme to increase the crude protein content of the forage can only benefit the NHN wombat population.

Two primary approaches can be used to increase the crude protein content of the forage: artificial fertilisation or irrigation. Experimentation with the former has already begun at EFNP as part of the NHN Recovery Plan (Horsup and Davidson 1994). However, I would emphasise caution with this approach mainly because the response of each forage species, particularly native grass species, to fertilisers is unknown. Fertilisation may encourage production of undesirable grass and browse species that may out-compete native forage species if the levels of nutrients becomes elevated.

The alternative that should be investigated is implementation of an irrigation experiment coupled with current forage management practices. An

experimental plot comparing forage quality and quantity under this method with current forage improvement management practices (fire, slashing and fertilising) would greatly add to direction of the management plan in improving forage quality. Although the initial capital outlay for an experimental irrigation system may be substantial, potential benefits may far exceed other forage improvement practices, particularly if the effects of high drought frequency can be negated.

Drought needs to be an important consideration in the management plan to improve the available forage. Rainfall is not reliable in the district (Chapter 2) and forage improvement actions such as fire, slashing and fertilisation all require rainfall to be effective.

Slashing

The cyclic population trends of eastern grey kangaroos suggest that the availability of new grass growth on surrounding pastoral properties following rain may present a comparatively better forage resource than EFNP. However, during the dry season the availability of forage in the range of the NHN wombat is higher than surrounding pastoral properties (regardless of below average rainfall). It is at this time when the population of eastern grey kangaroos on EFNP reached its maximum density.

Managing forage to promote new green growth (by slashing) may be advantageous during the wet season if rains are favourable. However, slashing in poor seasons may decrease the quantity of forage not only during the wet season, but, more importantly, during the dry season. Since wombats have a 'bulk feed' strategy to compensate for reduced quality of forage, reducing the availability of forage by slashing may actually be to the detriment of the NHN wombat. Also the potential for resource competition with eastern grey kangaroos may become a significant factor. Therefore, caution should be heeded with a slashing approach to forage improvement.

Seeding

Native grass seeds are available commercially. Sowing seeds in the range of the NHN wombat can only be of benefit to the wombat, and to the native grass

community. Species that should be selected include *Enneapogon* spp. and *Aristida* spp., which are the principal forage species consumed by the NHN wombat (Chapter 3). In addition, I would strongly encourage the increase of *Chrysopogon fallax* by seeding. Although *C. fallax* does not appear to be a major component of the diet, most *C. fallax* plants are heavily grazed in the range of the NHN wombat (personal observations). *Chrysopogon fallax* therefore could be classified as a preferred forage species rather than a principal forage species (Petrides 1975, Dawson *et al.* 1992). In addition, *C. fallax* was not detected by microscopic analysis as it was often confused with other plant species including *Enneapogon* species. Finally, *C. fallax* also has a comparatively higher concentration of total nitrogen than other native grasses.

- **Continued Monitoring of Forage Quality and Quantity**

Chapter 2 illustrated short-term trends in the quality and quantity of the forage environment at Epping Forest National Park. However, it considers only a short period of time in relation to the dynamics of grassland communities in the Australian tropical savanna. In comparison to the work of Crossman (1988) and Anderson and Back (unpublished data, Queensland Department of Primary Industries) the species composition, frequency and biomass of forage species in the NHN wombat habitat have changed significantly over a ten year period. Long term changes in the quantity of forage are poorly documented with no data on seasonal changes. Further, documentation of temporal changes in the quality of forage has been limited to the current study despite the possible implications for the nutritional ecology of the NHN wombat. Long-term seasonal monitoring of the quality and quantity of forage should therefore be a high management priority.

Species composition, frequency and biomass should be monitored biannually (wet season: February; and dry season: September) to provide long term data about forage quantity. While the scale of the monitoring need not be as comprehensive as in this study, monitoring should consider a representative selection of the whole NHN wombat habitat. I would recommend that three transects established in this study (transects 1, 5 & 7 representing typical NHN wombat habitat) be selected for long-term monitoring using established monitoring

techniques (either BOTANAL or QGRAZE). The transect approach allows a profile across the NHN wombat habitat (sandy alluvial soils) to be investigated, increasing the potential of encountering the range of forages preferred by the wombat.

Likewise forage quality is also very important in any future monitoring practices, not only in the current range of the NHN wombat but also for potential re-introduction sites and free-ranging captive populations. Near infrared spectrometry as a tool for measuring forage quality at EFNP (Chapter 2) should be adopted as a routine monitoring technique as well as to quantitatively assess the success of forage improvement procedures. The time and cost saving of the NIRS method will allow comprehensive nutritional mapping of forage species in the current and former range of the NHN wombat. The use of NIRS should be considered an essential tool for routine monitoring of forage quality.

- **Herbivore Responses to the forage environment of Epping Forest National Park**

With the implementation of any forage improvement plans it is essential that the response of all herbivores to 'improved' pasture be monitored. Northern hairy-nosed wombats, eastern grey kangaroos and invertebrates are dominant herbivores at EFNP. Faecal pellet monitoring is efficient in determining the feeding activity and density of eastern grey kangaroos, and to a lesser extent NHN wombats. However the presence of NHN wombat faecal pellets does not indicate feeding activity, unlike the eastern grey kangaroo (Chapter 6). Consequently, I would recommend that a non-invasive technique be developed to determine if and how wombats use pasture improved by experimental manipulations. Given the relatively small size of current experimental plots and their clear bias toward open areas of habitat, tower-mounted infrared video monitoring combined with activity sensors (similar to those used in home security systems) may provide information on habitat utilisation but resolution and detail may be limited. Although details on specific NHN wombat behaviour maybe limited by such an approach, information gained on habitat utilisation is far more important than specific NHN wombat behaviours. It is also important to monitor the botanical composition of NHN

wombat diet (via microscopic analysis) in the vicinity of experimental pastures. Clearly if wombats are not consuming species dominating the composition of pastures improved by experimental manipulations, the value of the experimental procedure should be questioned.

Grazing pressure from invertebrates (primarily termites and locusts) is also a key component of the EFNP grazing system and should be quantified. The potential for a catastrophic event caused by pestilence exists and should be considered with appropriate contingency plans devised. Likewise the importance of termites in nutrient cycling at EFNP is an unknown that should be quantified.

The dietary overlap between the NHN wombat and the eastern grey kangaroo is extremely high (Chapter 6). While this study does not suggest that resource competition currently exists, the potential for resource competition is real, particularly with forage improvement and management practices. Populations of eastern grey kangaroos throughout the range of the NHN wombat should therefore be continually monitored. The indirect method described in Chapter 6 will provide a better indication of actual macropod density because factors such as weather conditions and human activity are reduced. It also has the advantage of requiring little training or cost. The indirect survey task could be conducted while performing activities with a high level of disturbance such as trapping of NHN wombats or maintenance tasks. The continuation of the current survey design or the establishment of a new survey design but using the same procedure, with a quarterly monitoring schedule, will provide reliable data on eastern grey kangaroo population dynamics.

- **Geographic Information Systems (GIS) as a forage monitoring and management tool**

Steinbeck (1994) developed a GIS for Epping Forest National Park. His GIS was originally designed as a mapping tool for vegetation communities, soil types and locations of wombat burrows. Steinbeck (1994) developed the GIS as an evolving management tool, expanding as new information became available. Rather than focusing specifically on wombat habitat, the GIS in its original form considered EFNP a single entity. The potential exists for the expansion of the GIS

to become a management tool with a component specifically to examine forage dynamics. The application of the GIS to incorporate forage dynamics should include a systematic survey of forage species, incorporating quality and quantity. Areas to be selected should incorporate the current NHN wombat habitat and the historical range. Initial surveys (one in the wet season and one in the dry season) could describe species composition in a pre-determined mapping unit of NHN wombat habitat. A representative sample of each species in each mapping unit could then be harvested to measure attributes of forage quality by NIR spectroscopy. After the initial surveys representative mapping units could be selected for routine biannual surveys. This information, once in the GIS, could provide vital information on the state of the forage environment. Also it could provide valuable references for experimental forage enhancement programs and the detection of invasive species. Finally a GIS incorporating forage parameters and habitat characteristics could be a valuable tool to identify and maintain potential sites for establishment of a second NHN wombat population.

- **Range expansion**

Crossman (1988) described the former range of the NHN wombat, demonstrating that it extended to the northern boundary of the Park in the 1960's (Figure 2.3). The decline of the NHN wombat within this range has been dramatic but Johnson (1991a) estimates the carrying capacity of EFNP is up to 300 individuals. Given the increase in population size from the original assessment of some 20 to 30 individuals by Gordon *et al.* (1985), to a population estimate of approximately 65 individuals (Hoyle *et al.* 1996) there appears to be no evidence of re-establishment of its northern range. Why the population has not moved into its former habitat is unknown.

The mapping of forage quantity and quality and incorporation into a GIS for forage management, as described above, may provide some answers. It may highlight differences in species composition and quality between the currently occupied habitat and the former range. By comparing aspects of the forage between the current and former ranges, potential factors exacerbating the range

decline (currently attributed to cattle grazing and human interference) may become evident and appropriate management practices can be implemented.

Differences between the current and former ranges may not appear using the methods described above, and indeed may not exist. Limits in population size or behavioural causes may be impeding range expansion northward along the sandy alluvial soils (Figure 2.6). While it is most likely that the current wombat population represents a low density and there is no need to expand the range, human intervention may help in range expansion if it is required. Northern Hairy-nosed wombats favour pathways and fire-tracks to move large distances (personal observations). By establishing a fire track from the northern end of the current range to the northern boundary through the former habitat, range expansion may be enhanced by offering an easy to follow path. Further, the creation of the new fire track may stimulate construction of burrows near or on the verge of the new track.

- **NHN Wombat Social Behaviour**

Synchronous Emergences

Chapter 5 describes the conservative behaviour of NHN wombats and emphasises that little time is spent either actively foraging or in other activities. A key finding of this study was that the timing of above-ground activities appeared to be synchronous. Gaughwin (1981) inferred that the timing of emergences was cued physiologically rather than behaviourally, with this study supporting this assessment. However, this study failed to separate the link between above-ground activity and temperature or forage quality and quantity. In addition, the synchronisation of emergence may be a key component of wombat social behaviour. Unfortunately, the use of data loggers to determine emergence timing was limited by the accuracy of the temperature and light probes. The temperature probe was successful in determining emergence times but only during cooler winter months. A less invasive tool is therefore required.

Taylor (1977) recorded SHN wombats leaving and entering the burrow using an analogue counting device fixed to the entrance of a burrow. With recent advances in technology, Steele *et al.* (1995) successfully used microchip implants

and reading antennas to record exit and entrance movements in captive SHN wombats. A system such as that described by Steele *et al.* (1995) provides data at a higher resolution than either radio telemetry (Johnson 1991a, current study) or activity loggers can provide, and more importantly, has less impact on the animal. A feasibility study of the Steele *et al.* (1995) system should be assessed in the field on SHN wombats. If successful, its application to the NHN wombat would be very beneficial.

Reproductive Behaviour

Of specific concern is the low rate of reproduction in the population. One factor contributing to the poor rate of reproduction could be the low apparent rate of encounter between individuals. This would be exacerbated if home ranges did not overlap as a consequence of edge effects created by a small population.

If the encounter frequency with other wombats is low, reproduction may also be low. Gaughwin (1981) suggested that olfaction is an important component of wombat behaviour and is widely used in marking behaviours. The use of olfactory signals is likely to reduce the necessity for direct social interactions, and hence the encounter frequency can be low. Steele (pers. comm.) suggests that if the home range of an adult male overlaps with an adult female, the male will be able to detect the state of oestrous from olfactory cues. However, knowledge of the olfaction-driven social system and social interactions between individuals is superficial and requires further investigation for all wombat species, as does the reproductive cycle in the NHN wombat.

- **Trapping and Handling of Wombats**

Trapping is a necessary component of all wombat research and is required to add to the dearth of information known about the NHN wombat, particularly its reproductive biology. The 'smart trap' developed by Vern Steele (Department of Anatomy, Monash University) should be fully developed and implemented for NHN wombats. The 'smart trap' targets individual animals by reading implanted microchips and incorporates a more sophisticated trap design than that of Crossman (1988). Its application to the trapping of NHN wombats will allow

targeted animals to be trapped, minimising potential distress to non-targeted animals.

It is important that when a NHN wombat is trapped, the maximum potential information is gathered. One of the shortcomings of the Hoyle *et al.* (1995) study was the lack of available information on the life history of the wombat that their population predictions hinged upon. By continuing trapping, even at reduced levels, the data base can continue to evolve. Further, I would strongly recommend the continued use of bioelectrical impedance analysis by experienced operators to examine temporal changes in body composition during any trapping procedure. Although not as successful as stable isotopes to determine *in vivo* body compositions, trends (or lack of trends) in body composition measured by BIA will provide valuable information on how wombats respond to changes in the quality of forage. It is equally important in establishing if females use body fat as a means to cope with the energetically demanding process of lactation.

Clearly trapping does have a detrimental effect on NHN wombat behaviour (Chapter 5), but Hoyle *et al.* (1995) overestimated the significance of changes in NHN wombat body mass attributed to trapping. Body mass changes described by Hoyle *et al.* (1995) represent only minor changes in total body mass and are not significant in terms of the body mass of a 'bulkfeeding' herbivore (e.g. the difference between a full gastrointestinal tract and a half-full gastrointestinal tract). However, although I question the interpretation by Hoyle *et al.* (1995), I do not question their general conclusion that trapping is detrimental to wombats. Trapping should be kept to a bare minimum and the attachment of collars of any description should be avoided, primarily because of the adjustment period (Chapter 5) and the potential risk of injury to the wombat.

- **Comparative Study with SHN Wombats**

Like each of the three species of wombats, the SHN wombat has received limited research attention, yet its ecological similarities with the NHN wombat make it ideal for comparative studies. The large population size of the SHN wombat and its status as a pest species in some regions makes it an ideal species to

perform ecological experiments where habitat or animals are experimentally manipulated. In addition, it would be an ideal species to use in order to establish if translocation is suitable in hairy-nosed wombats.

The following publication is submitted in support of this thesis.