ELLISON, GEORGE THOMAS HENRY

THERMOREGULATION AND ENERGY METABOLISM OF THE POUCHED MOUSE *SACCOSTOMUS CAMPESTRIS* PETERS FROM SOUTHERN AFRICA

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Thermoregulation and energy metabolism of the pouched mouse *Saccostomus campestris* Peters from southern Africa

by

George Thomas Henry Ellison

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For my parents

The person who receives education is like a man who has been given all the food available in a starving village in order that he might have the strength to bring back supplies from a distant place.

Julius Nyerere

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by

George Thomas Henry Ellison

Supervisor: Professor J.D. Skinner
Mammal Research Institute
University of Pretoria
Pretoria 0002
South Africa

ABSTRACT

Thermoregulation and energy metabolism of the pouched mouse (*Saccostomus campestris*) was studied to establish how this species copes with different climatic conditions throughout its broad distribution in southern Africa.

Nine burrows excavated at four localities in South Africa each contained a single nesting chamber, at least 200mm below ground, in which variation in temperature was minimal. Not all of the burrows contained bedding material or caches of food and there was no direct effect of photoperiod, temperature or locality on nest-building or food-hoarding behaviour in captivity which indicated that these activities do not respond to seasonal or geographical changes in climate. However, males and females were observed collecting seeds throughout the year in the wild, presumably to minimise the amount of time spent foraging outside their burrows where thermoregulatory costs and predation risks are higher.

There was a multiple correlation between body size and a variety of climatic factors which was largely due to a positive correlation with rainfall. These geographical differences in body size appeared to be heritable, and might represent an adaptation to reduce the energy requirements of pouched mice living in arid/semi-arid environments where food availability is low.

Although there were significant differences in energy metabolism between pouched mice from nine localities, their resting metabolism was not correlated to local climatic conditions, possibly because they do not experience geographical variation in temperature when they are at rest inside their nests. Nevertheless, they do experience some seasonal and geographical differences in ambient temperatures because they are active above ground at night when cold temperatures necessitate an increase in thermoregulatory heat production. This might explain why pouched mice display seasonal changes in heat production while animals from cooler localities exhibit a higher capacity for heat production than those from warmer areas. The significance of a correlation between locality temperature and the duration of spontaneous daily torpor remains unclear, because torpor was only observed at temperatures below those usually recorded within their burrows.

It therefore appears that pouched mice are able to survive under a wide variety of climatic conditions because their semi-fossorial lifestyle allows them to avoid most unfavourable temperatures. At the same time they also display ecotypic differences in body size, heat production and torpor which help them cope with geographical variation in food availability and nocturnal temperatures which they cannot escape.

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PREFACE

Some studies related to this work and cited herein have already been published or accepted for publication:

ELLISON, G.T.H. 1988. *Dorylus helvolus* L. (Formicidae: Dorylinae) preying on *Saccostomus campestris* (Rodentia: Cricetidae). *J. ent. Soc. sth. Afr.* 51: 296.

ELLISON, G.T.H. 1990. A note on the small mammal fauna of Vaalkop Dam nature reserve. *Koedoe* 33: 114-116.

ELLISON, G.T.H. & SKINNER, J.D. 1990. Noradrenaline thermogenesis in conscious and anaesthetised pouched mice (*Saccostomus campestris*). *Comp. Biochem. Physiol.* 97A: 23-26.

ELLISON, G.T.H. & SKINNER, J.D. 1991. Seasonal energy requirements and thermoregulation of growing pouched mice, *Saccostomus campestris* (Cricetidae). *Int. J. Biometeorol.* 35: 98-102.

ELLISON, G.T.H. & SKINNER, J.D. *in press*. The influence of ambient temperature on spontaneous daily torpor in pouched mice (*Saccostomus campestris*: Rodentia - Cricetidae) from southern Africa. *J. therm. Biol.*

ELLISON, G.T.H. & WESTLIN-VAN AARDE, L.M. 1990. Ringtail in the pouched mouse (*Saccostomus campestris*). *Lab. Anim.* 24: 205-206.

ELLISON, G.T.H., BRONNER, G.N. & TAYLOR, P.J. *in press*. Is the annual cycle in body weight of pouched mice (*Saccostomus campestris*) the result of seasonal changes in adult size or population structure? *J. Zool., Lond.*

HAIM, A., ELLISON, G.T.H. & SKINNER, J.D. 1988. Thermoregulatory circadian rhythms in the pouched mouse (*Saccostomus campestris*). *Comp. Biochem Physiol.* 91A: 123-127.

HAIM, A., RACEY, P.A., SPEAKMAN, J.R., ELLISON, G.T.H. & SKINNER, J.D. 1991. Seasonal acclimatization and thermoregulation in the pouched mouse *Saccostomus campestris*. *J. therm. Biol.* 16: 13-17.

Copies of these papers are included in Appendix I.

Two additional, unpublished manuscripts cited in the text are:

ELLISON, G.T.H., SKINNER, J.D. & FERGUSON, J.W.H. *in prep.* The effect of temperature and photoperiod on daily metabolism and activity of pouched mice (*Saccostomus campestris*) from contrasting habitats in southern Africa.

WESTLIN, L.M. & ELLISON, G.T.H. *in prep.* Reproductive flexibility in female pouched mice (*Saccostomus campestris*: Cricetomyinae).

Copies of these papers are included in Appendix II.

Other writings by the author, not directly related to the subject of this dissertation include:

ELLISON, G.T.H. & SKINNER, J.D. 1991. Thermoregulation and torpor in African woodland dormice, *Graphiurus murinus*, following cold acclimation. *Z. Säugetierk.* 56: 41-47.

GILLIES, A.C., ELLISON, G.T.H. & SKINNER, J.D. 1991. The effect of seasonal food restriction on activity, metabolism and torpor in the South African hedgehog (*Atelerix frontalis*). *J. Zool., Lond.* 223: 117-130.

CHAPTER 1: GENERAL INTRODUCTION

Energy metabolism as a factor limiting the distribution of mammals

There is growing concern at the increasing rate with which the World's species continue to disappear as a result of human activities and this has given the study of biological diversity ("biodiversity") an air of urgency (Prescott-Allen 1991). A primary factor determining the distribution and abundance of organisms is their ability to tolerate the physical and chemical characteristics of their environment (Mann 1967; Krebs 1978). Therefore, the study of these abilities represents a natural starting point in our understanding of biodiversity. One fundamental component of environmental compatibility is energy metabolism which indicates both the energy required for maintenance, and the amount necessary for successful reproduction (Thompson & Nicoll 1986). For this reason, ecological energetics has recently established itself as a coherent doctrine on its own (Grodzinski, Klekowski & Duncan 1975).

Among endothermic mammals and birds, energy metabolism is inextricably bound up with thermoregulation due to the large proportion of energy spent maintaining homeothermy. This is particularly true for small species (which are easier to handle and more commonly studied) because they have less insulation and higher rates of heat loss as a result of their larger surface area to volume ratio (Hart 1971). With this in mind Wunder (1978) proposed a hierarchy of energy use for small mammals in which thermoregulation assumed priority (Fig.1). His reasoning for this was that "if a mammal is not thermoregulating, then it is either hypo- or hyper-thermic or is becoming so, and ultimately cannot perform its normal functions; therefore, channeling (sic) energy to other functions would not be possible" (Wunder 1984). It is therefore not surprising that many of the studies which have investigated geographical variation in energy metabolism of small mammals have approached this question through an analysis of thermoregulation (Table 1).

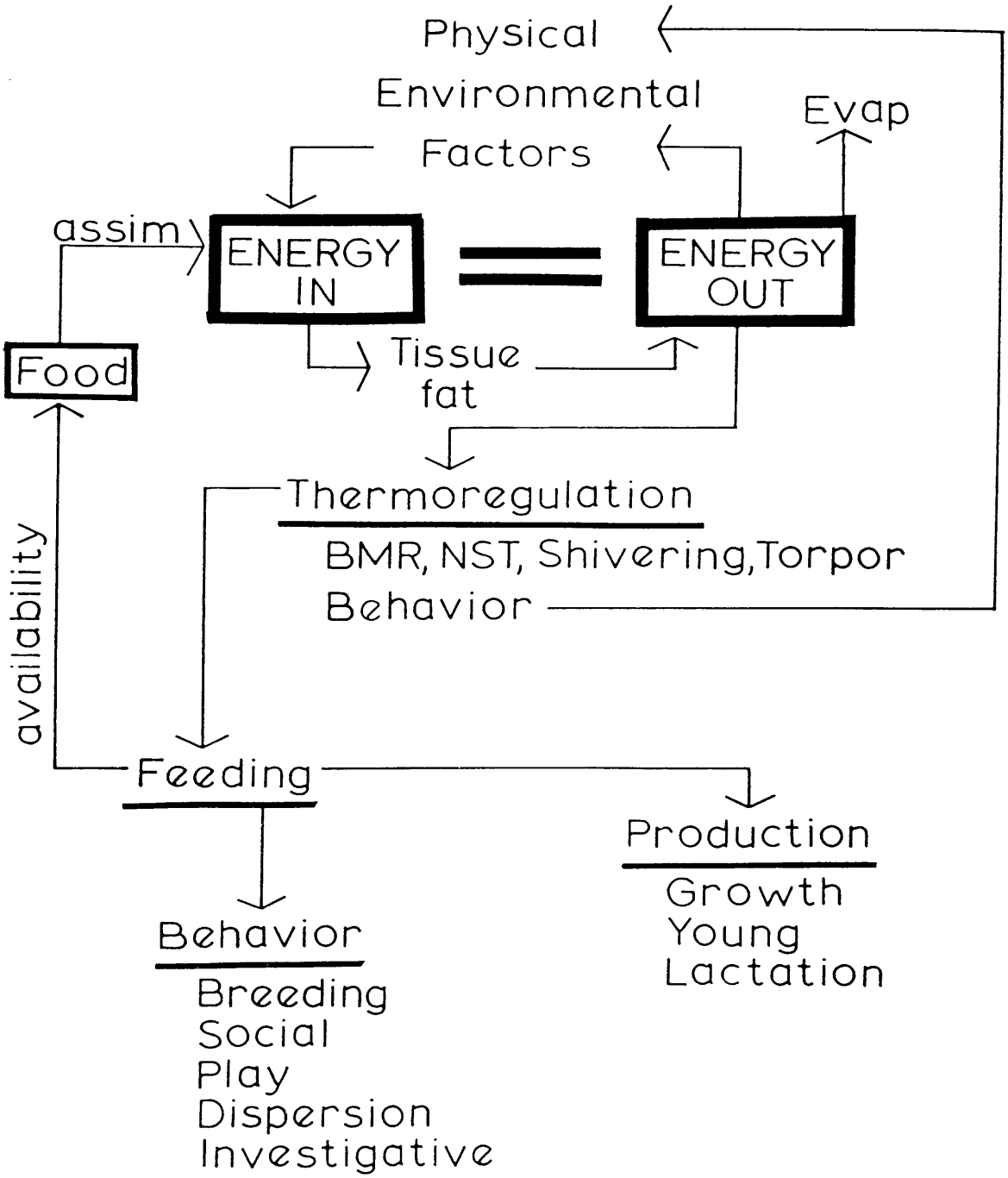


Figure 1 "A conceptual model of energy balance for small mammals, indicating a priority cascade for energy allocation. Lines represent both total energy flow and rate functions." (Taken from Wunder 1978).

Table 1 A selective list of studies which have examined geographical variation in energy metabolism of small rodents, indicating the (sub)species and parameters studied

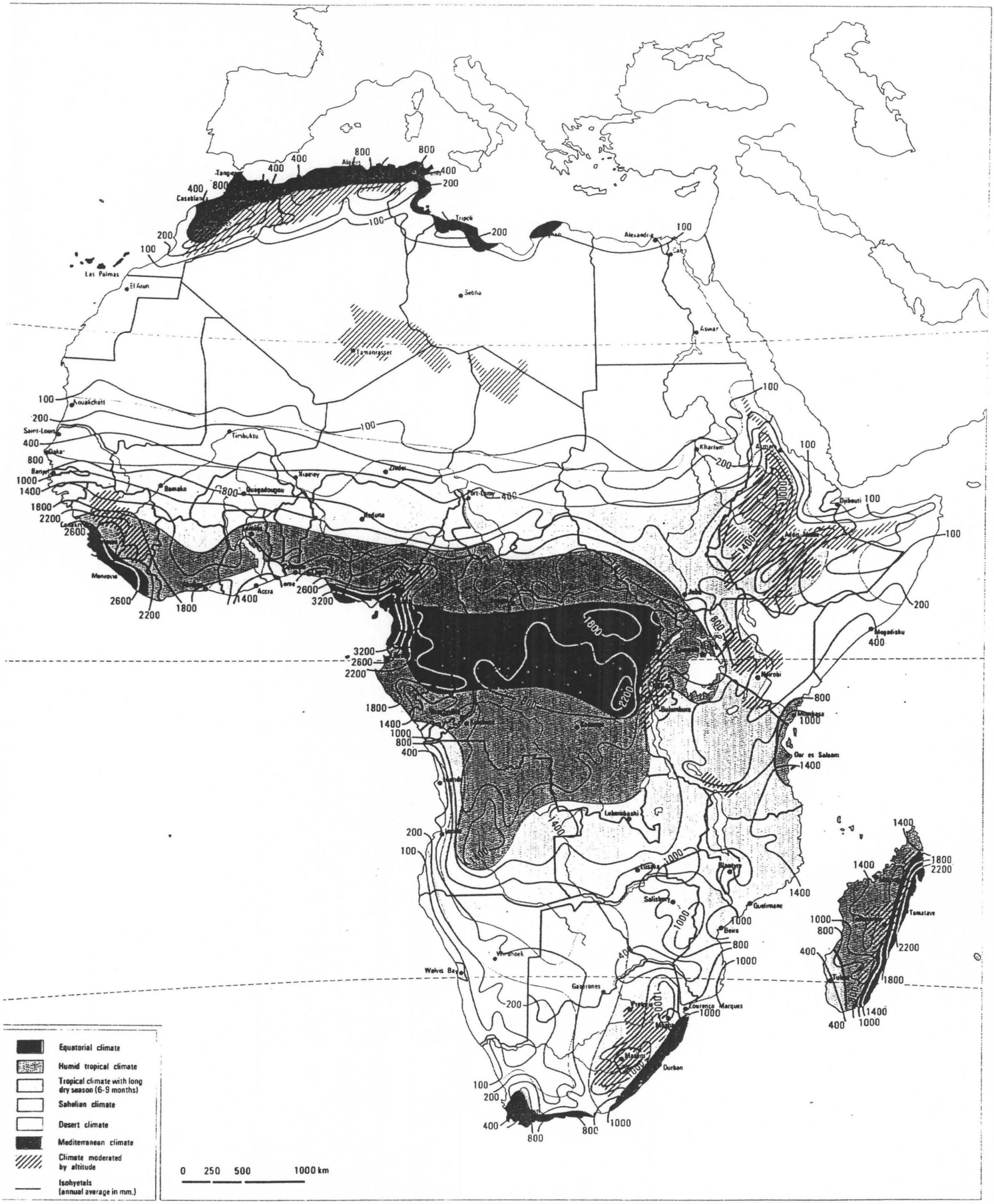
Study	(Sub)Species examined	Field of Study
Armitage et al. 1990	*Marmota flaviventris*	Thermoregulation and body size
Barry 1976	5 *Peromyscus* subspecies	Hoarding activity
Best 1982	*Dipodomys agilis*	Burrow structure
Borut et al. 1978	*Acomys russatus*	Thermoregulation
Bowers 1971	5 *Sigmodon* species	Thermoregulation
Bradley et al. 1974	2 *Thomomys* species	Thermoregulation and body size
Brenner & Lyle 1975	*Tamias striatus*	Torpor
Chappell 1985	*Peromyscus maniculatus*	Thermoregulation
Coetzee 1970	*Rhabdomys pumilio*	Body morphology
Downs & Perrin 1989	4 *Gerbillurus* species	Burrow structure
Downs & Perrin 1990a	4 *Gerbillurus* species	Thermoregulation and body size
Downs & Perrin 1990b	4 *Gerbillurus* species	Water turnover
Ellison & Skinner 1991a	*Graphiurus murinus*	Torpor
Haim 1984	2 *Gerbillus* species	Thermoregulation
Haim & Borut 1976	*Acomys russatus*	Thermoregulation
Haim & Fairall 1986	*Rhabdomys pumilio*	Thermoregulation and body size
Haim et al. 1984	*Spalax ehrenbergi*	Thermoregulation
Haim et al. 1985	*Spalax ehrenbergi*	Thermoregulation
Haim et al. 1987	2 *Xerus* species	Thermoregulation
Hayward 1965a	6 *Peromyscus* subspecies	Body size and composition
Hayward 1965b	6 *Peromyscus* subspecies	Thermoregulation
Hayward 1965c	6 *Peromyscus* subspecies	Burrow temperatures
Hartung & Dewsbury 1979	3 *Peromyscus* and 4 *Microtus* species	Nest-building
Heath & Lynch 1983	*Peromyscus leucopus*	Nest-building and torpor
Horton & Wright 1944	*Neotoma fuscipes macrotis*	Nest-building and food hoarding
Hudson & Deavers 1973	8 *Spermophilus* species	Thermoregulation
King et al. 1964	4 *Peromyscus* species	Nest-building
Lovegrove 1986	2 *Bathyergus* species	Thermoregulation and body size
Lyman et al. 1983	*Mesocricetus brandti*	Torpor
Mason 1974	2 *Peromyscus* species	Thermoregulation
McNab 1979a	2 Heteromyid rodents	Thermoregulation
McNab & Morrison 1963	9 *Peromyscus* subspecies	Thermoregulation
Moreno & Delibes 1981	*Eliomys quercinus*	Torpor
Nevo & Amir 1964	*Dryomys nitedula*	Torpor
Nevo & Shkolnik 1974	*Spalax ehrenbergi*	Thermoregulation
Nevo et al. 1986	*Spalax ehrenbergi*	Body size
Roberts et al. 1969	*Peromyscus maniculatus sonoriensis*	Thermoregulation
Scheck 1982	*Sigmodon hispidus texianus*	Thermoregulation
Shkolnik & Borut 1969	2 *Acomys* species	Thermoregulation
Tannenbaum & Pivorun 1984	3 *Peromyscus* species	Torpor
Tannenbaum & Pivorun 1987a	2 *Peromyscus* species	Food hoarding
Tannenbaum & Pivorun 1987b	2 *Peromyscus* species	Torpor
Tannenbaum & Pivorun 1988	2 *Peromyscus* species	Torpor
Thomas 1974	*Tamias striatus pipilans*	Burrow structure and food-hoarding
Thompson 1985	2 *Reithrodontomys megalotis* subspecies	Thermoregulation and torpor
Thorington 1970	*Peromyscus leucopus noveboracensis*	Morphology
Viljoen 1985	4 Scuirid species	Thermoregulation and body size
Ward & Armitage 1981	*Marmota flaviventris*	Food consumption and body size
Weiner & Heldmaier 1987	2 *Phodopus sungorus* subspecies	Thermoregulation and body size
Wolfe 1970	3 *Peromyscus* species	Nest-building
Yahav et al. 1988	*Spalax ehrenbergi*	Food consumption
Yahav et al. 1989	*Spalax ehrenbergi*	Water turnover
Yousef et al. 1974	8 rodent species	Water turnover

In order to establish the precise nature of ecotypic adaptation one should ideally examine geographical variation in a single species. Unfortunately, few small mammals exhibit extensive distributions and most of the studies mentioned in Table 1 employed comparisons of more than one (sub)species so that their findings are complicated by phylogenetic considerations. The overall aim of the present thesis was therefore to examine geographical variation in thermoregulation and energy metabolism of a single, widely-distributed species: the pouched mouse, *Saccostomus campestris*.

Taxonomy and fundamental biology of the pouched mouse

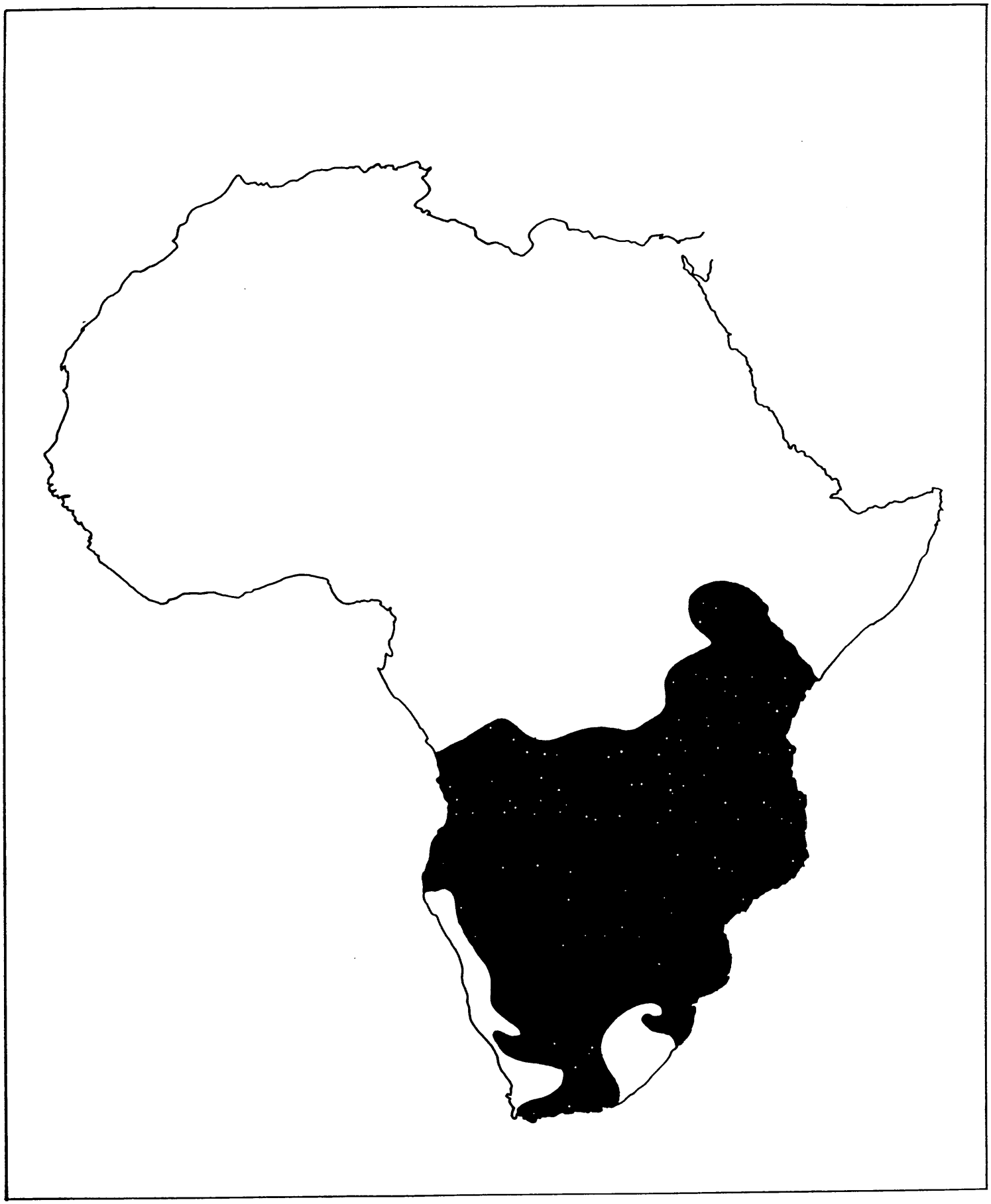
The pouched mouse is a small cricetid rodent, endemic to Africa (see Fig.s 2 & 3), which occurs from southern Uganda to the Cape province of South Africa (Delany 1975; Kingdon 1974; Skinner & Smithers 1990). According to Denys (1988) the genus *Saccostomus* is represented as *S.major* in the Pliocene fossil record at Laetoli (Tanzania) and has therefore been around for over 3.5 million years. By the turn of the century four species and subspecies of *Saccostomus* had been described (Sclater 1901), although current studies only recognise a single species in southern Africa (*S.campestris*, Matthey 1958) and possibly a second in East Africa (*S.mearnsi*, Denys 1988). Karyotypic analysis of *S.campestris* from widely distributed localities in southern Africa has revealed extensive variation in the number of chromosomes between different populations with a total of 16 variants ranging from 2n= 28-50 chromosomes (Fig. 4, Gordon 1986). The karyotypic differences between these populations appear to be caused by Robertsonian translocations and heterochromatic additions (Ferreira 1990, see Fig. 5) neither of which are thought to be effective cytological isolating mechanisms under normal conditions (Sites & Moritz 1987). While monobrachial centric fusions can cause reproductive isolation (Baker & Bickham 1986) this type of rearrangement has not been observed in *S.campestris* (Ferreira 1990).

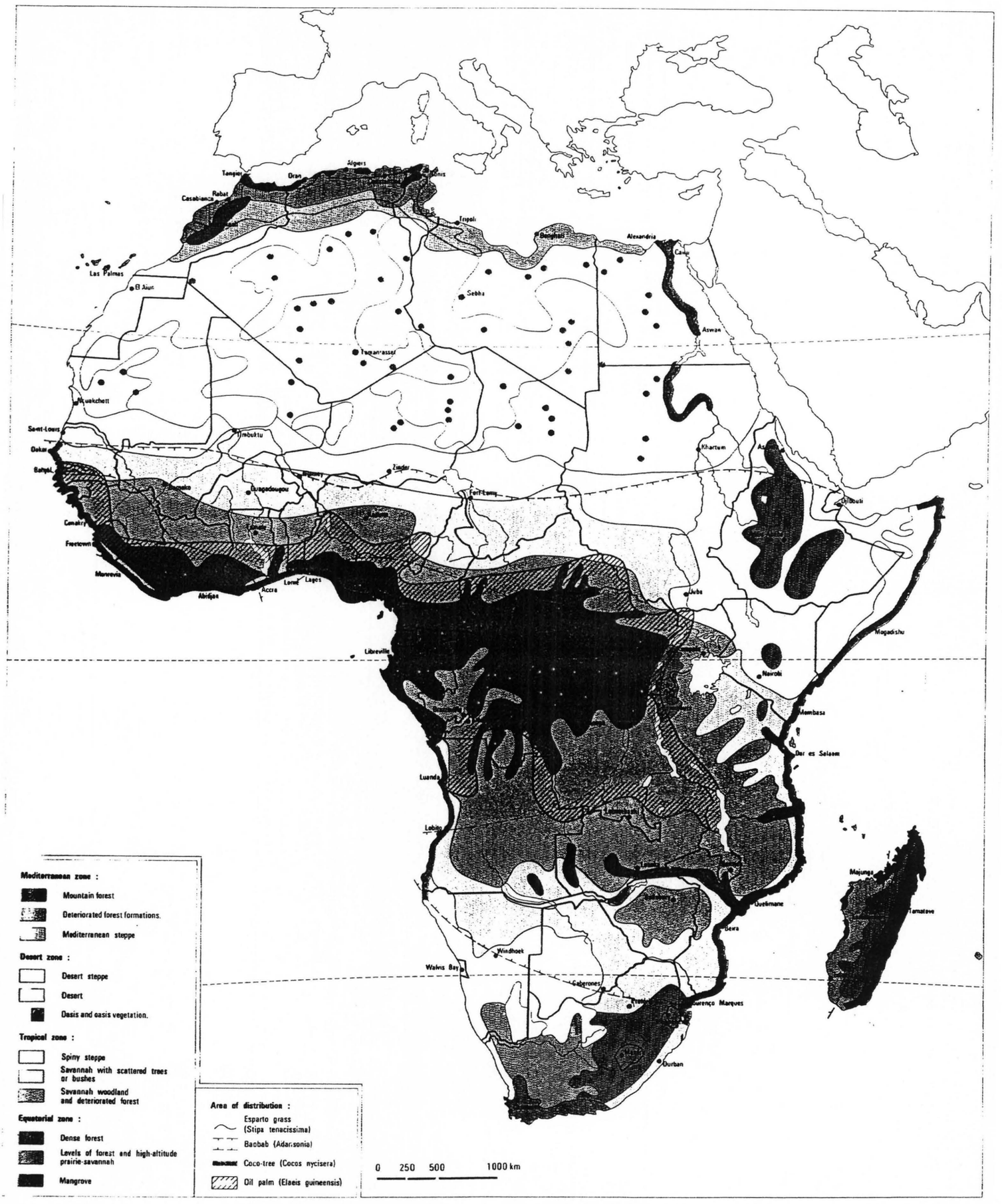
Figure 2 Overleaf: Major climatic zones of Africa (from Van-Chi Bonnardel 1973) with an overlay showing the distribution of *Saccostomus campestris* throughout the continent (from Kingdon 1984; Skinner & Smithers 1990).



MAJOR CLIMATIC ZONES

Figure 3 Overleaf: Major vegetation zones of Africa (from Van-Chi Bonnardel 1973) with an overlay showing the distribution of *Saccostomus campestris* throughout the continent (from Kingdon 1984; Skinner & Smithers 1990).





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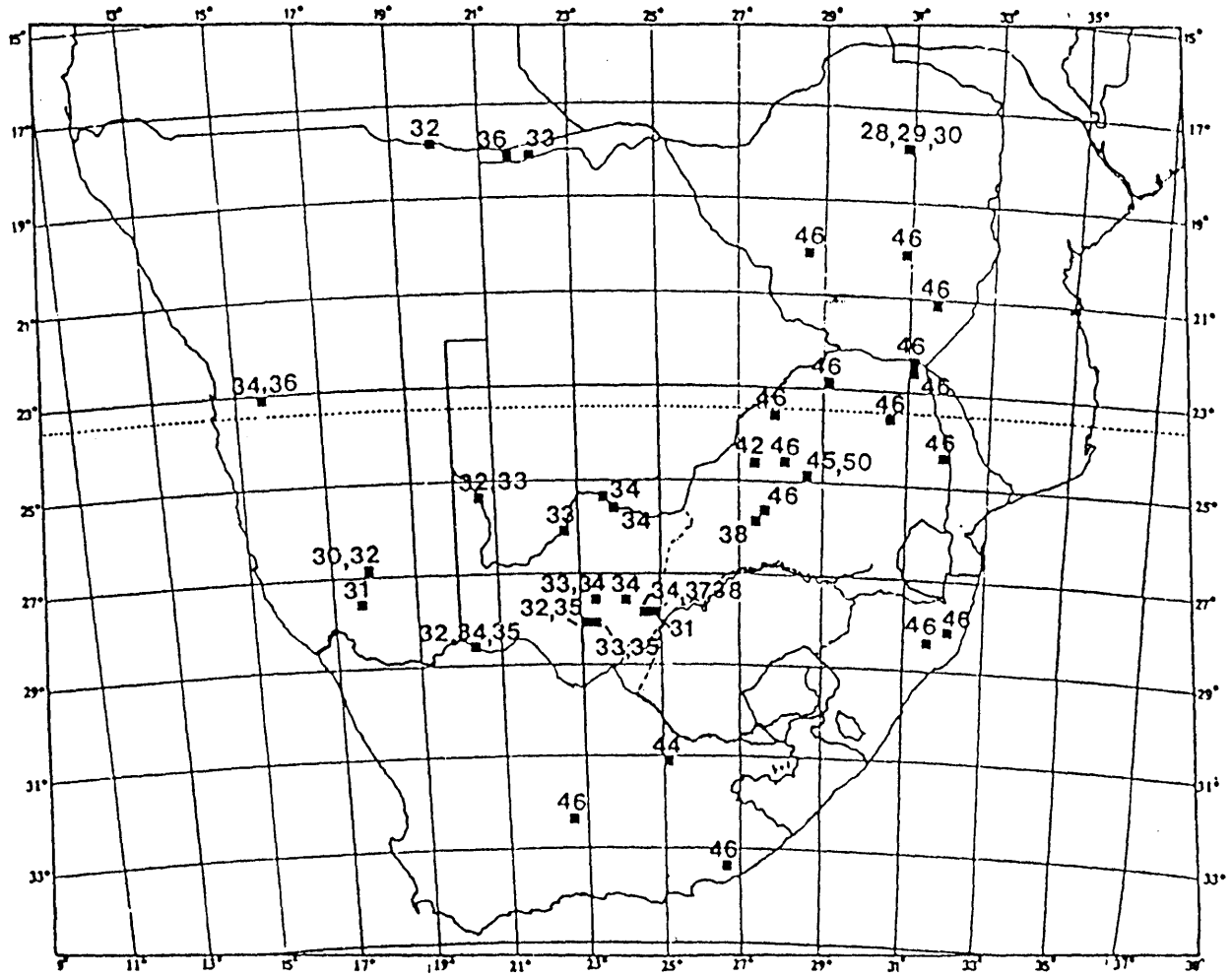

Figure 4 Karyotypic variation in of *Saccostomus campestris* within the southern African subregion (from Gordon 1986).

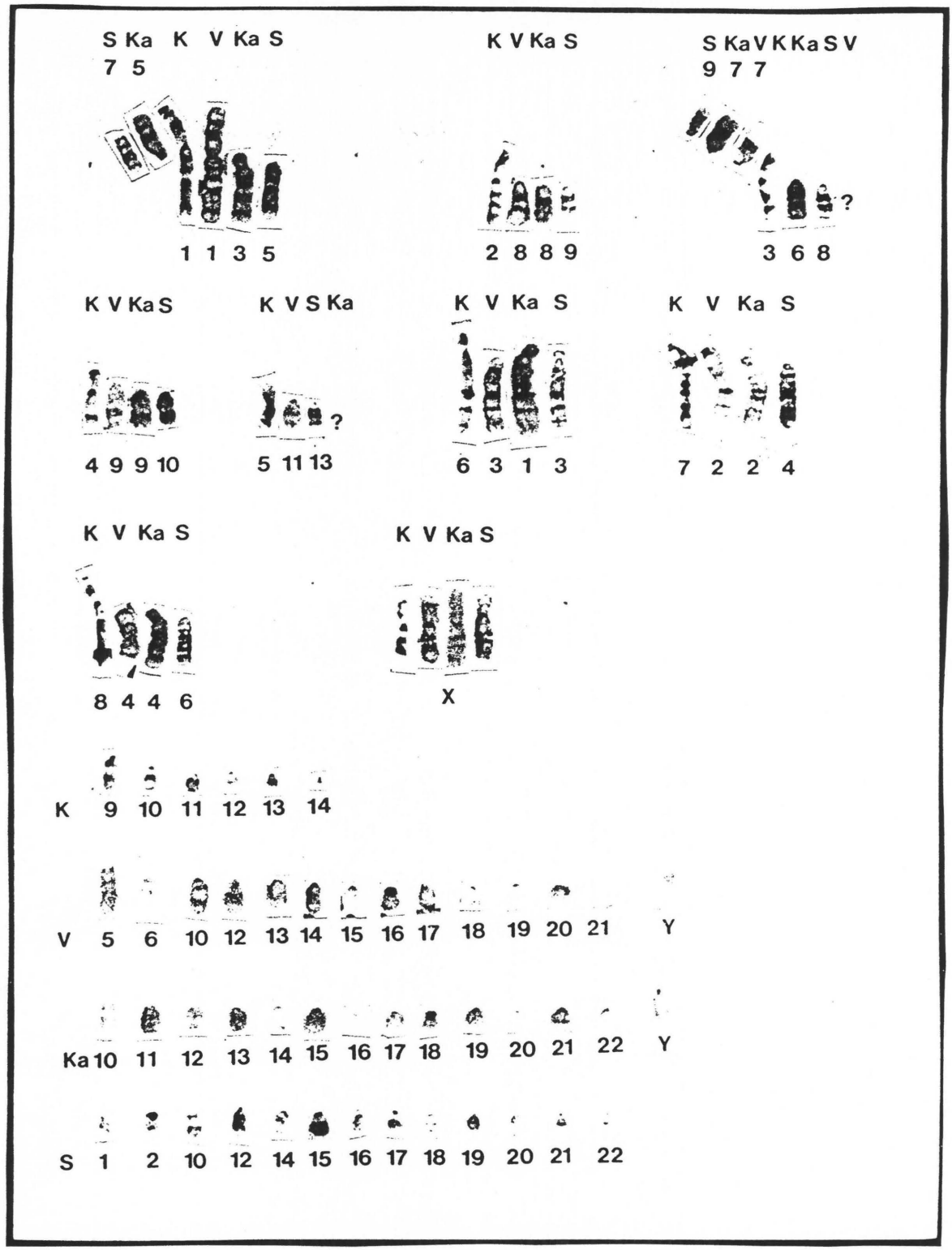

Figure 5 Euchromatic homology between the G-banded karyotypes of pouched mice from four localities in southern Africa (S-Skukuza 24°50'S 31°40'E, Ka-Karoo National Park 32°21'S 22°35'E, V-Vaalkop Dam 25°30'S 27°30'E, and K-Kuruman 27°42'S 23°26'E). The numbers refer to chromosome numbers in the individual karyotypes. A question mark (?) indicates that chromosomes were not identified in the respective karyotype due to poor resolution of banding patterns. (Reproduced from Ferreira 1990).

Within the southern African subregion *S.campestris* occurs in many different habitats: from the arid kalahari desert in Botswana, to the mesic subtropical lowlands of Natal, in areas experiencing mean annual rainfall between 100mm and 1200mm, and in localities from sea level up to altitudes of 1800m (Skinner & Smithers 1990). Although Rautenbach (1978) recorded pouched mice in four of the six biotic zones he assessed, they are primarily associated with open woodland savanna (De Wit 1972; De Graaff 1981; Kern 1981; Watson 1987; Ellison 1990) and are most abundant in the southern savanna woodland and the south west arid zone (De Graaff 1981). In these areas *S.campestris* is not an uncommon member of the rodent community but usually occurs at lower densities than other species (eg. Ellison 1990). For example, Watson (1987) found that *S.campestris* had a maximum, post-drought density of 6.7 individuals/ha in the Kruger National Park compared to 30/ha for multimammate mice (*Mastomys (Praomys) natalensis*). Being poor climbers (Earl & Nel 1976) and poor swimmers (Hickman & Machiné 1986) they are predominantly terrestrial and semi-fossorial whilst Bronner (personal communication*) caught one individual in the canopy of a tree in northern Natal. Burrows are positioned beneath cover or in open ground and may be constructed from the disused burrows of other species (Smithers 1971). Pouched mice are generally omnivorous (Hanney 1965; Kern 1981; Watson 1987) although their diet is dominated by seeds. Indeed, their name is derived from the internal cheek pouches they use to carry food back to their burrow (Skinner & Smithers 1990). There remains some confusion as to their social structure as they have been described as solitary (Copley 1950; Walker 1964) and communal (Ansell 1960) while Smithers (1971), De Graaff (1981) and Rautenbach (1982) all report them occurring singly, in pairs or small family parties.

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* G.N. Bronner, Mammal Department, Transvaal Museum, Pretoria, South Africa.

Field reports of reproductive activity within the subregion suggest that *S.campestris* are seasonal breeders and give birth during the warm summer months (eg. December, Walker 1964; January-April, Smithers 1971). However, breeding occurs all year round under long photoperiod and warm temperatures in captivity (Earl 1978) and a series of studies under laboratory conditions (Westlin-van Aarde 1988; 1989a; 1989b; 1989c; Westlin & Ellison in prep.) has recently increased our understanding of reproduction in this species. *S.campestris* also exhibit seasonal changes in thermoregulation and energy metabolism (Ellison & Skinner 1991; Haim, Racey, Speakman, Ellison & Skinner 1991), while Korn (1989) and Ellison, Bronner & Taylor (in press) have shown that pouched mice from the Transvaal display a winter decline in body weight which might help reduce their energy requirements at this time of year. However, no studies have yet examined geographical variation in energy metabolism of *S.campestris* and this will be the overall aim of the present study.

Can there be a strategy of optimisation for energy metabolism?

According to Krebs & McCleery (1984) "the belief that (biological) design is optimized stems from the axiom that natural selection tends to maximize fitness. Since fitness depends on design features, it follows that they too should be optimized." However, in view of the variety of solutions which different species have developed to cope with similar environmental constraints it might be prudent to apply the principle of optimisation with caution. Schoffeniels (1986) has even gone so far as to suggest that "Nobody has the right... to introduce into their science, into science in general, the idea of a draught (or plan) that must obligatory (sic) take precedence of a realisation." Clearly, there are inherent dangers in the assumption that a human concept of "optimal" can necessarily predict the adaptation crafted by natural selection. Energy metabolism is no exception to this problem as there are a myriad of alternative avenues through which ecotypic adaptation can be expressed (Table 2).

Table 2 Potential avenues for metabolic adaptation of homeotherms which allow them to avoid or resist energetic stresses (either an increase in heat loss or a decrease in food availability) in their natural environment (adapted from Wunder 1984)

Type of stress	Increasing heat loss		Decreasing food availability	
Type of response	Avoid	Resist	Avoid	Resist
Behavioural	Migration Nest-building Microhabitat selection	--	Migration Food hoarding	--
Morphological	Larger body size Smaller appendage size Improved insulation	--	Smaller body size	--
Physiological	Lower thermal conductance Enter hibernation or daily torpor	Higher thermogenesis via basal metabolism shivering or NST	Enter hibernation or daily torpor	--

Therefore, the only expectation in this study will be that individuals inhabiting different localities should display mechanisms that enable them to cope with the energetic consequences of living therein. However, no prediction will be made regarding the avenue(s) through which adaptation(s) should occur, nor the extent to which any adaptation should be developed.

Avenues of energetic adaptation

Behavioural mechanisms are often considered to be the first avenue through which small mammals adapt to their environment because the modification of behaviour enables many species to avoid stressful conditions altogether (Fahri 1987) either by selecting from the conspicuous heterogeneity of their physical environment or by creating more favourable conditions through their own activities (eg. by building a nest or hoarding food, Gordon 1972). Hayward (1965c) and Wunder (1974) have even proposed that behavioural avoidance of environmental stresses can be so effective that additional morphological and physiological adaptations become unnecessary. For this reason it has been suggested that behavioural responses should be preferred (Gordon, Fehler & Long 1986) because they help reduce overall energy expenditure and are therefore more efficient than most autonomic alternatives. Clearly, an investigation of behaviour is the obvious place to begin any study on environmental adaptation of energy metabolism as it determines to what extent a given species is exposed to stressful environmental conditions and to what extent it is able to behaviourally avoid these. For non-migratory pouched mice possible behavioural adaptations include huddling, nest-building and/or food-hoarding (Table 2). To establish the scope for behavioural modification of these parameters under natural conditions (Key Questions 1-3, see below) the group size, burrow structure and hoarding activity of wild pouched mice was examined in Chapter 2. Nest-building and food-hoarding was further investigated under controlled laboratory conditions in Chapter 3 to address Key Questions 4 & 5 (see below).

Although behavioural adaptations probably allow many species to escape the energetic stresses prevailing in their natural environment, it is important not to over-estimate behaviour as a means of dealing with severe conditions such as those which occur seasonally or at the geographical limits of a species' distribution (Hill 1983). Under these circumstances morphological and/or physiological mechanisms assume increasing importance. Morphological adaptations primarily involve changes in body size together with modifications in the size and shape of organs and appendages. Whilst phenotypic changes in body size can occur in response to environmental conditions, such as photoperiod (Steinlechner, Heldmaier & Böckler 1983) temperature (Lyman 1954) and the availability of food (Dauncey & Ingram 1986) and water (Buffenstein & Jarvis 1985), geographical differences in body size have long been viewed as an heritable adaptation to local conditions, as suggested by Bergmann's rule. Because large bodies lose proportionately less heat than small ones, Bergmann's rule was originally formulated to explain the positive correlation between body size and latitude in terms of the thermoregulatory advantages of increasing body size at colder, higher latitudes (Allaby 1985). Recently, the rule has been applied in a more general way by researchers who ignored latitude and simply emphasized the association between body size and temperature (eg. James 1970). However, the rule has been criticised because it describes clinal changes in morphology that may not be important for controlling heat loss (Scholander 1955) whilst thermoregulation may not be the only factor determining body size (Yom-Tov & Nix 1986). Nevertheless, body mass remains one of the most important factors affecting energy metabolism (Nevo, Beiles, Heth & Simson 1986) as it determines the total energy requirements of each animal (Wunder, Dobkin & Gettinger 1977) and also influences basal metabolism (Kleiber 1947; Brody 1945; Elgar & Harvey 1987; Hayssen & Lacy 1985), thermal conductance (Herried & Kessel 1967; Bradley & Deavers 1980), non-shivering thermogenesis (Heldmaier 1971) and maximum metabolic rate (Koteja 1987; Bozinovic & Rosenmann 1989; Weiner 1989). Meanwhile, Allen's rule (which predicts that homeotherms from colder climates have smaller appendages than those from warmer areas in order to prevent

excessive heat loss) has also been criticised because there are many other adaptations for heat conservation, such as increases in the thickness of fur and subcutaneous fat, which are thought to be more important than changes in the size of appendages (Allaby 1985). However, Hart (1956) has shown that small mammals have a limited capacity for improving thermal insulation using changes in the length or density of their fur which suggests that a decrease in appendage size may play a more important role in reducing excessive heat loss from these species. This is supported by the smaller appendages of mice reared at cold temperatures (Sumner 1909) and the positive correlation between ambient temperature and tail length in white footed mice (*Peromyscus leucopus;* Thorington 1970) and striped mice (*Rhabdomys pumilio;* Coetzee 1970). To assess the adaptive significance of morphological variation in pouched mice from the southern African subregion, geographical differences in body length, body mass and appendage size were correlated with climatic variables in a similar way to that conducted for Australian mammals by Yom-Tov & Nix (1986) and for mole rats (*Spalax ehrenbergi*) by Nevo et al. (1986). These analyses are presented in Chapter 4 in answer to Key Questions 6 & 7 (see below).

Over and above the effect of body size on metabolism (McNab 1983), inter- and intra-specific differences in physiological measures of energy metabolism (such as basal metabolic rate and minimum thermal conductance) have been ascribed to dietary (McNab 1980a; 1984; 1986a; 1986b), phylogenetic (Herreid & Kessel 1967; Bradley & Deavers 1980; Hayssen & Lacy 1985; Elgar & Harvey 1987) and behavioural (McNab 1979b; Buffenstein 1984) factors. At the same time, many studies have described geographical variation in physiological characteristics even after prolonged acclimation under standard conditions which suggests that they may be the result of fundamental ecotypic adaptations (see Table 1). For example, lower basal metabolism is common among species from arid areas (eg. Buffenstein & Jarvis 1985b) and is thought to be an adaptive mechanism for reducing food and water requirements in environments where their availability is limited. Likewise, an increase in the capacity for non-shivering thermogenesis (NST) might be

expected among individuals from colder localities (eg. Haim, Heth, Avnon & Nevo 1984) because NST provides an essential contribution to thermoregulatory heat production in small mammals exposed to cold temperatures (Heldmaier, Steinlechner & Raphael 1982). For this reason, the investigation into physiological adaptations of energy metabolism starts in Chapter 5 with an analysis of fundamental geographic variation in thermoregulation and energetics of pouched mice maintained or bred under standard laboratory conditions (Key Questions 8-10, see below).

Whilst fundamental differences between populations can often be observed under standard laboratory conditions (Table 1), these conditions only elicit a small part of the available physiological responses which these populations possess. Indeed, Schmidt-Nielsen (1986) has suggested that responses to stressful conditions are probably a more relevant measure of ecotypic adaptation because they indicate how animals cope with the extremes that they encounter in their natural environment. Seasonal extremes are often used to determine the scope for metabolic adaptation (eg. Heldmaier & Steinlechner 1981) although seasonal differences in environmental conditions vary from place to place so that these can also provide a selective force for localised adaptation. In recent years, a series of studies has shown that most vertebrates which, like *S.campestris*, exhibit seasonal changes in reproduction or energy metabolism use photoperiod to precisely time the annual cycle of these events (see Heldmaier, Steinlechner, Ruf, Wiesinger & Klingenspor 1989 for review). At the same time, there is increasing evidence that populations from areas with milder winters rely on photoperiod to a lesser extent than those from harsher localities whose survival depends to a greater extent on their ability to accurately predict seasonal changes in conditions (Barry 1976; Heath & Lynch 1983). With the positive correlation between temperature, growing season, photoperiod and latitude, this idea may even be viewed as a photoperiodic extension of Bergmann's rule. In Chapter 6 the relative importance of photoperiod and temperature as environmental cues for seasonal acclimation of energy metabolism was assessed in three populations of pouched mice inhabiting localities which

experience different winter conditions. In this way, geographical variation in the cueing of seasonal acclimation was measured as an indication of ecotypic adaptation to localities with different seasonal extremes (see Key Questions 11-13 below).

Ellison & Skinner (in press) have recently demonstrated that pouched mice from southern Africa respond to temperatures of 15°C and below by entering bouts of spontaneous torpor which reduce the cost of thermoregulation by up to 39.0% per day. Although these temperatures are lower than those normally experienced by pouched mice in their burrows (Ellison & Skinner in press) physiological adaptations often exceed the immediate requirements of various small mammals (eg. NST capacity in *Phodopus sungorus*, *Clethrionomys glareolus* and *Apodemus sylvaticus*; Heldmaier, Böckler, Buchberger, Lynch, Puchalski, Steinlechner & Wiesinger 1985). Indeed, Heldmaier, Klaus & Weisinger (1990) have pointed out that small mammals ought to be able to cope with the worst conditions possible in their natural environment as these would only have to happen once to wipe out their entire population if they were unprepared. In a similar way spontaneous torpor might represent an emergency survival mechanism for pouched mice in southern Africa. If this is the case we might expect similar geographical variation to that reported for *Peromyscus* spp. (Tannenbaum and Pivorun 1984; 1987b; 1988) and for *Reithrodontomys* spp. (Thompson 1985) where individuals from colder areas had a greater capacity for torpor than those from warmer localities. This hypothesis is outlined in Key Questions 14 & 15 (below) which are tested in Chapter 7.

Finally, the relative importance of behavioural, morphological and physiological mechanisms for ecotypic compatibility of energy metabolism in pouched mice from southern Africa is summarised in the concluding Chapter (Chapter 8) which also evaluates the extent to which geographical variation in these traits are caused by natural selection (Key Questions 16-18).

Key questions

1. How many adult pouched mice inhabit each burrow?

2. Are there geographical differences in the extent to which temperature extremes are buffered in pouched mouse burrows?

3. Are there any clear sexual and/or geographical differences in the structure of pouched mouse burrows or the quantity of food contained therein?

4. What is the effect of decreasing photoperiod and/or temperature on nest-building and food-hoarding activity of pouched mice in captivity?

5. Are there any sexual and/or geographical differences in nest-building and food-hoarding activity of pouched mice in captivity?

6. Is there any geographical variation in body and/or appendage size of pouched mice within the southern African subregion?

7. If there are geographical differences in body size and/or appendage size of pouched mice, are these correlated with climatic variation within the subregion?

8. Are there geographical differences in fundamental energetic physiology between different populations of pouched mice which persist under standard laboratory conditions?

9. If there are geographical differences in fundamental energetic physiology of pouched mice, are these correlated with variation in climatic conditions at each locality?

10. If geographical differences in fundamental energetic physiology of pouched mice are correlated with local climatic conditions, are these correlations due to heritable factors so that the metabolism of captive bred individuals can be predicted from the climatic conditions of their original locality?

11. To what extent do photoperiod and temperature act as cues for seasonal acclimation of thermoregulation and energy metabolism in pouched mice?

12. Are there any geographical differences in the use of photoperiod and temperature as cues for seasonal acclimation of energy metabolism and thermoregulation in pouched mice?

13. If there are geographical differences in the use of photoperiod and temperature as cues for seasonal acclimation of energy metabolism and thermoregulation, are these differences related to the severity of seasonal changes at each locality?

14. Is there any geographical variation in the incidence, depth and duration of spontaneous torpor among pouched mice from different localities?

15. Are geographical differences in the expression of spontaneous torpor by pouched mice associated with local climatic conditions at each locality?

16. What is the relative importance of behavioural, morphological and physiological mechanisms for ecotypic compatibility of energy metabolism in pouched mice from southern Africa?

17. How do behavioural, morphological and physiological mechanisms interact?

18. To what extent are geographical differences in these traits the result of natural selection?

CHAPTER 2: AN INVESTIGATION INTO GROUP SIZE, BURROW STRUCTURE AND HOARDING ACTIVITY OF WILD POUCHED MICE IN SOUTH AFRICA

Introduction

Pouched mice get their name from the large cheek pouches which they use to transport food back to their burrow (Skinner & Smithers 1990) and a large number of seed varieties and other food items have been recorded from the pouches of trapped individuals (Table 3). However, the contents and structure of few *S.campestris* burrows have been reported, and although Hanney (1965) described two burrows excavated in Malawi he made only passing reference to the seeds they contained. Indeed, the only quantitative measure of seeds found in pouched mouse burrows is given by Fitzsimmons (1920) who reported finding 1 lb (0,45 kg) of "*Acacia* nuts" in a burrow near Port Elizabeth (South Africa). Likewise, the number of burrow occupants remains unclear and *S.campestris* have been described as both solitary (Copley 1950; Walker 1964) and communal (Ansell 1960), while Smithers (1971), De Graaff (1981) and Rautenbach (1982) all report them occurring singly, in pairs or small family parties.

The aim of the present study was to investigate the group size, burrow structure and hoarding activity of pouched mice from South Africa. All three of these characteristics have an important bearing on the behavioural energetics of mammals: first because they determine whether animals can huddle to conserve heat, secondly because they indicate the extent to which they can avoid climatic extremes in an artificial microhabitat, and thirdly because they establish whether they are able to control the temporal availability of their food (Sealander 1952; Wunder 1984; Vander Wall 1990).

Table 3 Previous reports of food items hoarded by pouched mice (unless otherwise stated all items were found in the pouches of trapped individuals)

Item	References
Seeds:	
Acacia spp.	Fitzsimmons (1920 - in burrow)
Acacia giraffae	Smithers (1971)
Acacia karoo	Swanepoel (1972); Kerley (1989); Ferreira (pers. comm.[a])
Acacia nilotica	Swanepoel (1972); Jacobsen (1977)
Acacia tortilis	Swanepoel (1972)
Asparagus spp.	Lynch (1983, pers. comm.[a])
Balanites maughamii	Swanepoel (1972 - in burrow)
Burkea africana	Jacobsen (1977)
Colophospermum mopane	Vesey-Fitzgerald (1966); Smithers (1971);
Combretum spp.	Smithers (1971);
Dichrostachys cinerea	Pienaar et al. (1980)
Diospyros lycioides	Lynch (1983, pers. comm.[a]); Kroese (pers. comm.[a])
Euclea crispa	De Wit (1972); Jacobsen (1977)
Grewia spp.	Smithers (1971);
Grewia bicolor	Pienaar et al. (1980)
Grewia flavescens	Pienaar et al. (1980)
Grewia monticola	Swanepoel (1972)
Peltophorum africanum	Jacobsen (1977)
Psoralea spp.	Kroese (pers. comm.[a])
Rhus ciliata	Lynch (1983, pers. comm.[a])
Rhus erosa	Lynch (1983, pers. comm.[a])
Rhus lancea	Lynch (1983, pers. comm.[a])
Rottboellia escaltata	Hanney (1965 - in burrow)
Xanthocercis zambesiaca	De Graaff (1981)
Insects:	
Isoptera	Transvaal Museum (Catalogue notes for TM1478)
Hemiptera - Pentatomidae	Lynch (1983, pers. comm.[a])
Orthoptera - Acrididae (eggs)	Shortridge (1934)

[a] Personal communications (South Africa):
S.M. Ferreira, Mammal Research Institute, University of Pretoria, Pretoria 0002;
M. Kroese, Port Elizabeth Museum, P.O.Box 13147, Humewood 6013;
C.D. Lynch, Department of Mammals, National Museum, P.O.Box 266, Bloemfontein 9300.

Materials and methods

The present study was conducted at four localities in South Africa which were visited at different times of the year: Skukuza (Kruger National Park 24°59'S 31°36'E) in February, Vaalkop Dam (near Rustenburg 25°23'S 27°28'E) in May and July, and both Pniel Estates (near Kimberley 28°35'S 24°31'E) and Doornhoek (Karoo National Park near Beaufort West 32°21'S 22°35'E) in November. Seasonal extremes in air and soil temperatures for these localities were obtained from the nearest meteorological station and have been summarised in Table 4.

Pouched mice were live-caught at each locality using Sherman traps baited with rolled oats and peanut butter. Individual animals were sexed and weighed, and their pouches and traps examined for hoarded food. They were subsequently toe-clipped for future identification and covered with fluorescent powder before release. Unfortunately, attempts to track pouched mice back to their burrows using the technique of Lemen & Freeman (1985) were unsuccessful because the trail of powder ran out after 5-10 m. However, it was possible to follow animals during daylight if they were marked with brightly coloured powder and although many individuals escaped or went to ground, eleven pouched mice were successfully followed back to a burrow (four at Skukuza, four at Vaalkop, one at Pniel and two at Doornhoek). In order to assess whether this technique was accurately locating *S.campestris* burrows, intensive trapping and hourly observations were conducted over five consecutive nights outside the four Vaalkop burrows and this confirmed that these belonged to pouched mice (see ***Results*** for further details).

Each burrow was subsequently excavated by hand and mapped out using a tape-measure and graph paper. The location of any food stores, latrines and nests were recorded and these were collected for further analysis and identification. Similarly, soil samples were taken from every burrow to evaluate the texture of the substrate. Bedding, faeces, seeds and invertebrates were separated manually using a 2,0 mm wire test-sieve (United, Nigel - RSA) and the quantity of each component was recorded.

Table 4 Seasonal variation in air and soil temperatures at the four pouched mouse localities studied in Chapter 2 (soil temperaures are the mean of measurements taken at 08h00 and 20h00)

Locality:	Skukuza	Vaalkop	Pniel	Doornhoek
Met. Station:	Skukuza (24°59'S 31°36'E)	Rustenburg (25°43'S 27°18'E)	Kimberley (28°48'S 24°46'E)	Beaufort West (32°18'S 22°40'E)
Altitude:	263m	1157m	1198m	893m
Mean annual rainfall:	563mm	685mm	419mm	223mm
	Mean daily temperatures (°C) in summer (November - February)			
Max. air temperature:	31,8	29,4	31,4	30,7
Min. air temperature:	19,6	16,2	16,5	14,6
Soil temperature at 0,1m:	30,3	27,6	26,7	27,0
Soil temperature at 0,2m:	30,8	27,9	28,0	28,4
Soil temperature at 0,3m:	30,4	27,0	27,9	27,4
Soil temperature at 0,6m:	29,5	25,7	26,5	26,0
Soil temperature at 1,2m:	28,2	24,4	24,0	24,3
	Mean daily temperatures (°C) in winter (May - August)			
Max. air temperature:	26,7	21,7	19,9	19,3
Min. air temperature:	7,5	4,3	4,3	5,7
Soil temperature at 0,1m:	19,2	15,5	11,4	12,6
Soil temperature at 0,2m:	20,8	17,6	13,8	14,1
Soil temperature at 0,3m:	21,2	17,8	13,8	14,8
Soil temperature at 0,6m:	22,2	18,7	15,4	16,3
Soil temperature at 1,2m:	23,4	19,9	18,0	18,3

Intact seeds and empty seed cases were identified at the Department of Agricultural Development in Pretoria and the total volume of each seed cache was estimated to the nearest 1,0 ml using an 11mm diameter (0-10ml) and a 21mm diameter (10-50ml) measuring cylinder. The seeds were then examined for signs of germination and evidence of infestation by seed-boring invertebrates (such as bruchid beetles) which leave a tell-tale bore-hole in the seed's outer test.

Daily variation in soil temperature was recorded at representative burrows from each locality using copper-constantan thermocouples connected to a multi-channel temperature recorder (KM1202: Kane May, Welwyn Garden City - UK). Four thermocouples were buried in soil next to the burrow at depths of 0, 200, 400 and 600mm and a fifth was placed at the same depth as the main nesting chamber. The thermocouples were allowed to equilibrate with soil temperatures for 24h before measurements began and thereafter temperature was recorded every 30min for a further 24h.

Results

Field observations

Hoarding by trapped pouched mice was recorded at all four localities, at different times of the year and among individuals of both sexes (Table 5). No insect remains were found in any of the 14 hoards collected, but at least 10 different varieties of seeds were identified and only a few of these had been attacked by seed-boring invertebrates. Seven hoards contained more than one type of seed, while each hoard contained up to 10ml of seeds. Although it wasn't clear how far these animals had travelled to collect their seeds most pouched mice were caught within 25m of their burrows (Table 6). However, some individuals (eg. the female from Vaalkop 2 and the male from Doornhoek 1) were trapped more than 50m away which provides some indication of potential foraging distances in this species.

Table 5 Intact seeds and empty seed cases found in the pouches and/or traps of live-caught pouched mice at four localities in South Africa (the proportion of seeds attacked by seed-boring invertebrates is given in brackets)

Locality	Date (Month)	Sex (M/F)	[a]Contents of pouches and/or trap (intact seeds or empty seed cases)	Volume (ml)	Fresh mass (g)
Skukuza	February	M -	23 cf. *Ziziphus* spp. seeds (0 bored)	4,4	2,46
			& 1 cf. *Ziziphus* spp. case (0 bored)	<1	0,02
			& 1 *Dichrostachys cinerea* seed (0 bored)	<1	0,02
Vaalkop	May	F -	10 *Euclea divinorum* seeds (1 bored)	1	0,52
	May	F -	2 *Euclea divinorum* seeds (0 bored)	<1	0,10
			& 1 *Ziziphus* spp. seed (0 bored)	<1	0,18
	May	M -	1 *Acacia nilotica* seed (0 bored)	<1	0,12
	May	F[b] -	55 *Euclea divinorum* seeds (10 bored)	3,8	2,20
			& 1 *Euclea divinorum* case (0 bored)	<1	0,02
	May	F[b] -	43 *Euclea* spp. seeds (6 bored)	1,4	0,78
	May	M -	1 *Ziziphus* spp. seeds (0 bored)	<1	0,08
			& 3 cf. *Olea* spp. cases (0 bored)	<1	0,21
	May	M[c] -	1 *Euclea divinorum* seed (0 bored)	<1	0,03
	July	M -	1 *Ziziphus* spp. seed (0 bored)	1	0,29
Pniel	November	M -	176 *Acacia tortilis* seeds (0 bored)	9,5	6,44
			& 1 leguminosae seed (0 bored)	<1	<0,01
Doornhoek	November	M -	4 *Acacia karoo* seeds (0 bored)	<1	0,12
			& 4 Unknown no.1 seed cases (0 bored)	<1	0,08
			& 1 *Diospyros lycioides* seeds (0 bored)	<1	0,14
	November	M[d] -	7 *Acacia karoo* seeds (0 bored)	<1	0,18
			& 1 *Acacia karoo* case (0 bored)	<1	0,02
			& 1 cf. *Diospyros lycioides* seed (0 bored)	<1	0,14
	November	M[e] -	3 *Diospyros lycioides* seeds (0 bored)	<1	0,22
	November	M -	1 unknown no.1 seed case (0 bored)	<1	0,02
			& 1 *Acacia karoo* seed (0 bored)	<1	0,02

[a] The prefix cf. ("confer") indicates uncertain identification;
[b] This female was the occupant of Vaalkop 3;
[c] This male was the occupant of Vaalkop 1;
[d] This male was the occupant of Doornhoek 1;
[e] This male was the occupant of Doornhoek 2.

Table 6 The number, sex and body mass of pouched mice occupying 11 burrows excavated at four localities in South Africa

BURROW NUMBER	DATE EXCAVATED (month)	DISTANCE FROM TRAP (m)	NUMBER OF OCCUPANTS (n)	SEX OF OCCUPANTS (M/F)	BODY MASS (g)
Skukuza 1	February	2	1	F	38
Skukuza 2	February	23	1 + 9 pups	1F + 9?	42
Skukuza 3	February	16	1	F	42
Skukuza 4	February	19	1	F	23
Vaalkop 1	July	20	1	M	74
Vaalkop 2	July	70	1	F	46
Vaalkop 3	May	4	1	F	55
Vaalkop 4	July	27	1	F	41
Pniel 1	November	22	1 + 7 pups	1F + 7?	49
Doornhoek 1	November	60	1	M	59
Doornhoek 2	November	25	1	M	44

Without exception, the burrows examined in the present study all contained single adult pouched mice (Table 6) and the only groups found comprised a female and her young (Skukuza 2 and Pniel 1). However, direct observations at Vaalkop Dam revealed that pouched mice visit neighbouring burrows and may move into these if their occupants disappear: During May the male from Vaalkop 1 was once trapped outside the entrance to Vaalkop 3 (24m away) while the female occupant of Vaalkop 2 frequently visited Vaalkop 4 (43m away) and had moved into this burrow in July when the previous occupant had disappeared.

There was also evidence that other rodent species visit or occupy *S.campestris* burrows: Multimammate mice (*Mastomys* spp.) trapped at the entrance to Vaalkop 3 and 4 entered these burrows when released, and a red veld rat (*Aethomys chrysophilus*) entered Vaalkop 1 when it was set free nearby. Meanwhile, three pygmy mice (*Mus minutoides*) were recovered from Vaalkop 2 when this was excavated in July after the previous occupant had moved to Vaalkop 4 (see above).

Several of the burrows examined had additional entrances which appeared to have been blocked with loose soil and small stones by their occupant. However, burrowing activity was only observed at Vaalkop 3 where the female occupant spent five consecutive nights in May excavating soil and filling two of the three entrances to her burrow with debris. Most of the pouched mice also plugged the main entrance to their burrows when they were inside, while at least three individuals (Vaalkop 3, 4 and Skukuza 2) appeared to have done so when they left, and these animals had to dig their way back in when they were tracked back to their burrows. Likewise, the entrance to Vaalkop 2 was blocked with soil in July after the previous occupant had moved to Vaalkop 4. However, it was not clear whether the pouched mouse had plugged the burrow before she left or whether this had been done by the pygmy mice which appeared to be sealed inside.

Burrow structure

Plan maps of each of the eleven burrows excavated are displayed in Figs. 6, 7 and 8, and their dimensions and structure have been summarised in Table 7. Characteristics common to all *S.campestris* burrows included a vertical or steeply sloping entrance and a single nest, 90-190mm across, and more than 200mm below ground. However, there was extensive variation in the location, depth and structure of the burrows examined: approximately half (n=5) were situated beneath vegetation and half (n=6) in open ground, while the mean depth of the tunnels ranged from 124mm to 745mm. There also appeared to be two major types of burrow: simple burrows comprising less than 2000mm of tunnels and one chamber (eg. Skukuza 1, 3 & 4, and Doornhoek 1) and more complex systems containing up to 8500mm of tunnels with 2-4 chambers (eg. Vaalkop 4 and Pniel 1). Although two of the more complex burrows (Skukuza 2 and Pniel 1) contained a mother and her young there was no clear association between burrow complexity and sex or body size (Table 6).

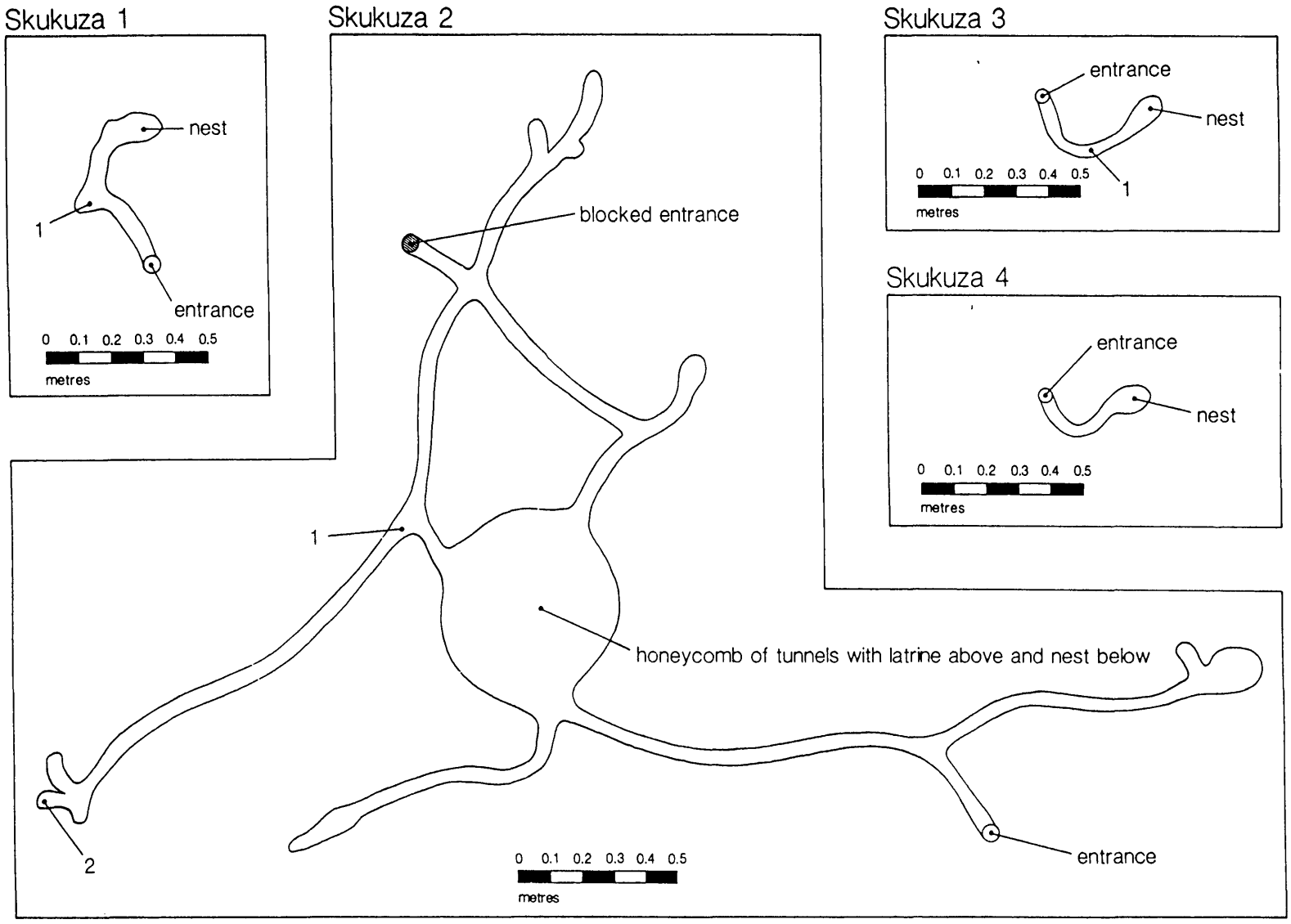


Figure 6 Plan maps of the four burrow systems excavated at Skukuza in the Kruger National Park. Entrances, nests and blocked sections of tunnel are indicated while any storage sites have been numbered and their contents described in Table 8. Skukuza 2 was based within an old termite mound which contained a honeycomb of small tunnels in its core.

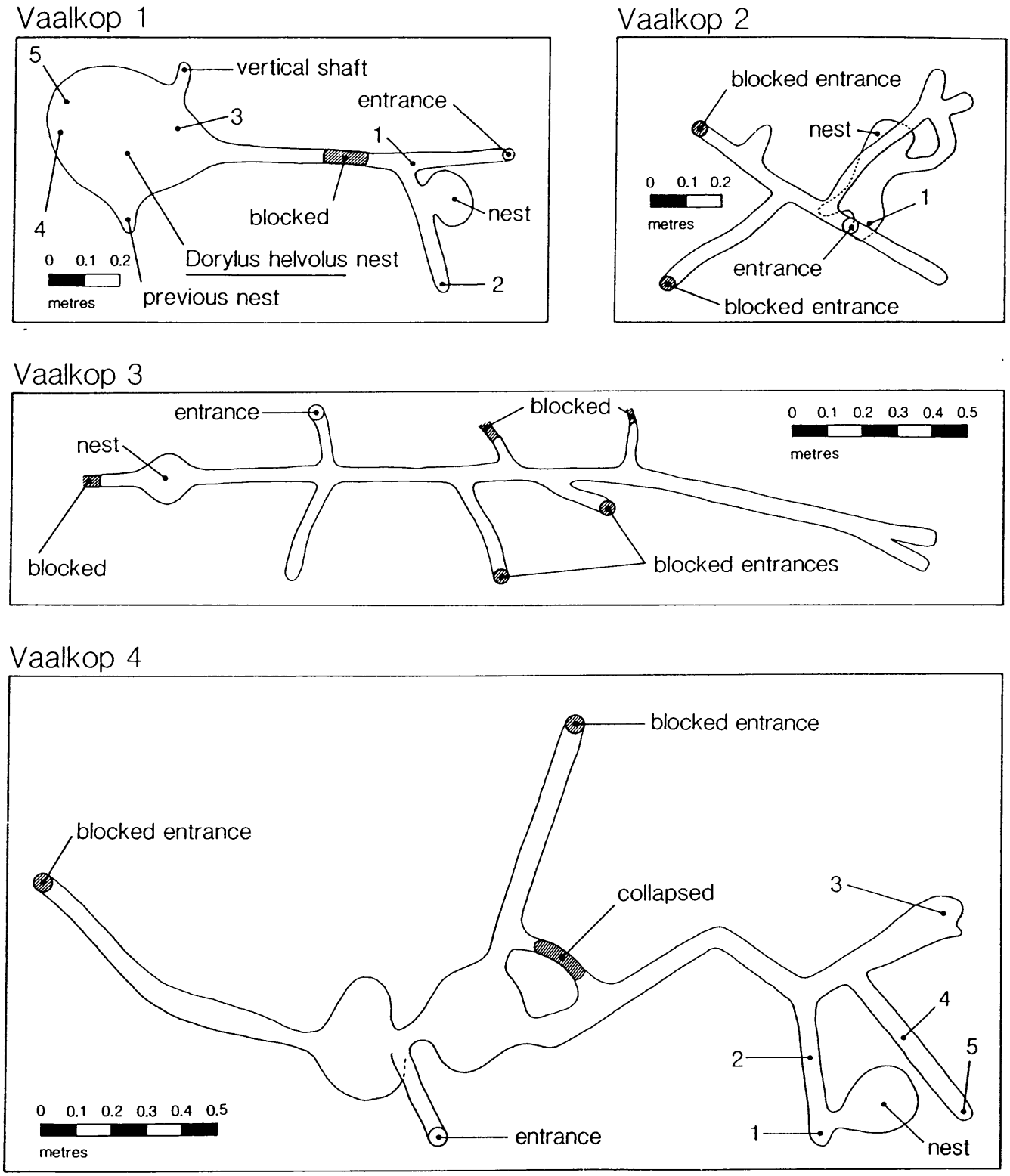


Figure 7 Plan maps of the four burrow systems excavated at Vaalkop Dam near Rustenburg. Entrances, nests and blocked sections of tunnel are indicated while any storage sites have been numbered and their contents described in Table 8. Vaalkop 1 was invaded by driver ants (*Dorylus helvolus*) during May and a connecting tunnel between the nest and the ant colony was found to be blocked with soil when the burrow was excavated in July (Ellison 1988).

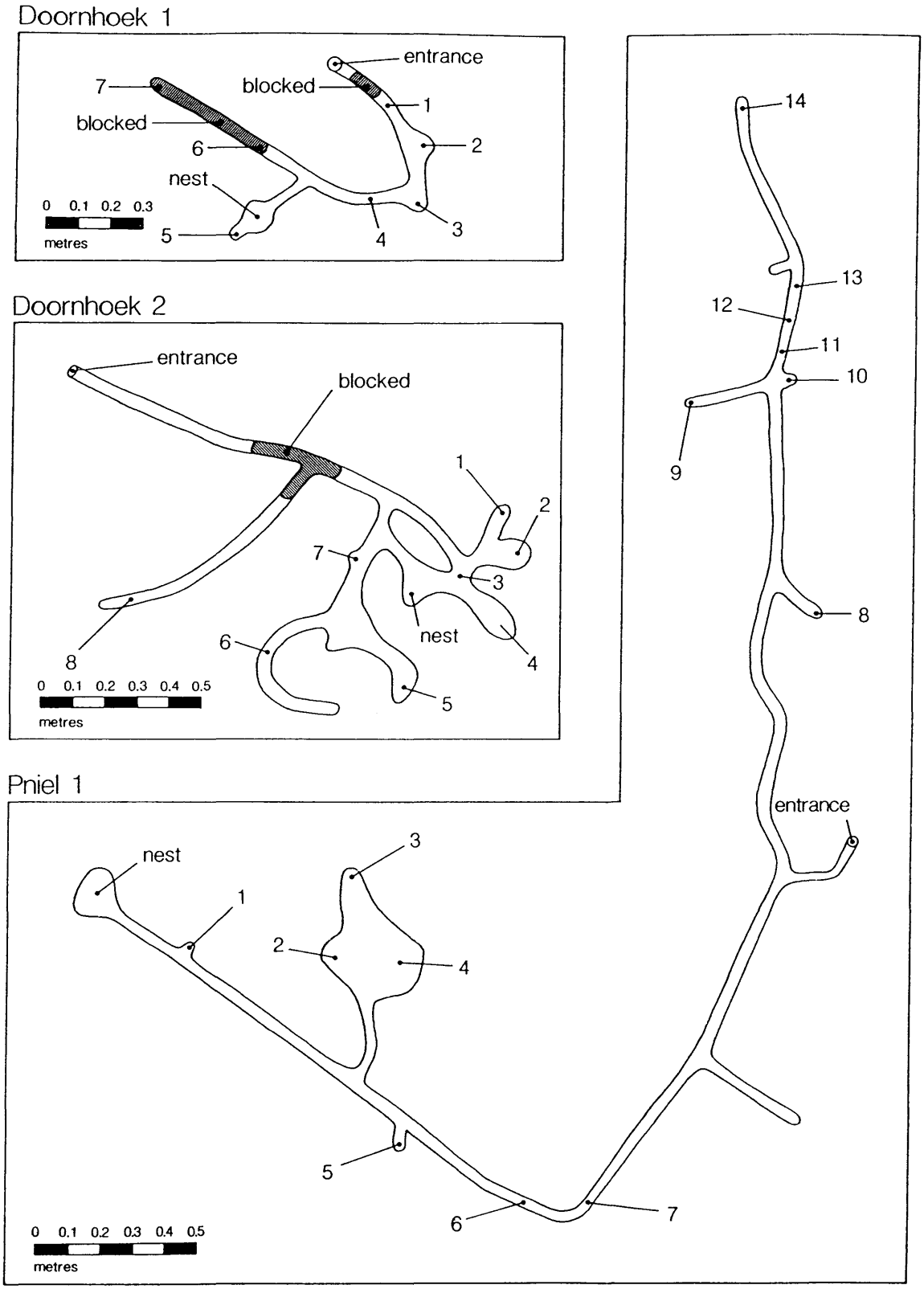


Figure 8 Plan maps of the two burrows excavated at Doornhoek in the Karoo National Park, and the single burrow excavated at Pniel Estates near Kimberley. Entrances, nests and blocked sections of tunnel are indicated while any storage sites have been numbered and their contents described in Table 8. The occupants of Doornhoek 1 & 2 were inside their burrows when these were excavated, and both individuals had blocked the tunnel leading from the entrance with loose soil and debris.

Table 7 Dimensions, structure and substrate texture of 11 pouched mouse burrows excavated at four localities in South Africa

Burrow number	Skukuza 1	Skukuza 2	Skukuza 3	Skukuza 4	Vaalkop 1	Vaalkop 2	Vaalkop 3	Vaalkop 4	Pniel 1	Doornhoek 1	Doornhoek 2
Entrance(s)											
Location	beneath vegetation	open ground	open ground	beneath vegetation	beneath vegetation	open ground	open ground	open ground	open ground	beneath vegetation	beneath vegetation
Number open (n)	1	1	1	1	1	1	1	1	1	1	1
Orientation	steep slope	vertical	steep slope	vertical	vertical	vertical	vertical	vertical	vertical	vertical	steep slope
Mean width (mm)	50	60	60	55	40	50	50	50	35	35	45
Mean depth (mm)	-	200	-	300	730	110	120	200	210	220	-
Total length (mm)	520	8420	320	340	1160	2120	4145	4230	6960	1565	3405
Mean width (mm)	42	55	37	44	39	40	45	57	39	38	44
Mean depth (mm)	172	392	172	340	745	124	168	310	319	297	235
Maximum depth (mm)	220	780	250	400	860	350	330	520	630	350	410
Chamber(s)											
Total number (n)	1	4	1	1	2	2	1	4	2	1	4
Number of storage sites (n)	2	4	2	0	6	2	0	6	15	8	9
Number of nests (n)	1	1	1	1	1	1	1	1	1	1	1
Maximum width of nest (mm)	120	120	100	90	160	100	150	170	140	90	190
Depth of main nest (mm)	200	780	250	400	800	340	250	380	620	350	360
Soil texture											
Stones >2mm diameter (% sand)	3	4	7	34	39	28	44	20	2	8	9
Sand 2-0,05mm diameter (%)	76	59	85	66	66	75	82	82	84	78	84
Silt 0,05-0,002mm diameter (%)	12	12	6	16	16	10	5	7	4	11	7
Clay <0,002mm diameter (%)	12	29	9	18	18	15	13	11	12	11	9
Classification (USDA Soil Conservation Service 1975)	Sandy Loam	Sandy Clay Loam	Loamy Sand	Sandy Loam	Sandy Loam	Sandy Loam	Sandy Loam	Sandy Loam	Loamy Sand	Sandy Loam	Loamy Sand

Many of the burrows appeared to have been modified by filling unused sections with debris, while the tunnel from the entrance was usually blocked by the occupant if they were inside the burrow when it was excavated (eg. Doornhoek 1 & 2, see Fig. 8). All of the burrows were found in predominantly sandy soils, although they were occasionally stony, and often contained a large proportion of sand particles larger than 2,0 mm in diameter (Table 7).

Burrow contents

Accumulated material was found in several of the burrows examined and this was located in nests, unused chambers and/or different sections of tunnel (see Figs. 6, 7 and 8). The number of these storage sites was variable and ranged from 0-2 in the simple burrows to 6-15 in the more complex systems (Table 7). Likewise, there was considerable variation in the amount of bedding, seeds and faeces stored at each of the sites, and this has been summarised in Table 8.

Most of the nests contained small amounts of dried grass and/or leaves, whilst little, if any, bedding material was found in four of the simpler burrows (Vaalkop 3, and Skukuza 1, 3 & 4). Similarly, none of these contained any intact seeds and neither did Vaalkop 2 which only contained a handful of *Ziziphus* spp. shells after its occupant had moved to Vaalkop 4 in July. In contrast, five of the remaining burrows contained extensive stores of intact seeds and empty seed cases and a total of 22 different seed varieties were found therein. None of these showed any signs of germination while the proportion attacked by seed-boring arthropods was variable: in Vaalkop 1 89,5% (1774/1982) of the intact seeds had been bored although only 2,5% (58/2404) of seeds in other burrows were affected. Meanwhile, no potential seed-borers were identified among the invertebrate remains collected and most of these belonged to detritivores (Myriapoda, Tenebrionidae, Nitidulidae and Scarabaeidae) which are common inhabitants of soil and subterranean environments. There was also no evidence that pouched mice in the present study hoarded invertebrate prey, even though one burrow (Doornhoek 2) contained several elytra from chrysomelid beetles which are not usually found in burrows.

Table 8i Contents of each storage site at the 11 pouched mouse burrows excavated at four localities in South Africa

Burrow number	Storage site number	Faeces (n)	Intact seeds (number bored) (n)	Volume (ml)	Mass (g)	Seed Cases (n)	Volume (ml)	Mass (g)	Other material
Skukuza 1	Nest	85	-	-	-	-	-	-	Pieces of leaf
	No.1	13	-	-	-	-	-	-	-
Skukuza 2	Nest	202	3 *Dichrostachys cinerea* (0 bored)	<1	0,03	Ca. 420 *Acacia* spp.	20	4,16	Mollusc shell; bone fragment
		-	-	-	-	Ca. 42 cf. *Grewia* spp.	8	0,83	-
		-	-	-	-	Ca. 385 Unknown no.1	18	5,15	-
		-	-	-	-	1 Unknown no.2	<1	0,01	-
	Nest Latrine	124	-	-	-	-	-	-	-
	No.1	-	-	-	-	Ca. 12 Unknown no.2	<1	0,10	-
	No.2	17	-	-	-	-	-	-	-
Skukuza 3	Nest	1	-	-	-	-	-	-	Pieces of grass and leaves
	No.1	7	-	-	-	-	-	-	Pieces of leaves and twigs
Skukuza 4	Nest	-	-	-	-	-	-	-	-
Vaalkop 1	Nest	17	1731 *Euclea divinorum* (1661 bored)	132	23,14	Ca. 27 *Euclea divinorum*	4	0,38	Pieces of leaves and grass
		-	40 *Ziziphus* spp. (33 bored)	12	4,74	Ca. 41 *Ziziphus* spp.	10	4,21	-
		-	31 *Diospyros lycioides* (30 bored)	4	0,47	-	-	-	-
	Previous nest	58	16 *Euclea divinorum* (3 bored)	2	0,60	Ca. 200 *Euclea divinorum*	22	3,53	A piece of shredded grass
		-	21 *Ziziphus* spp. (4 bored)	4	1,13	Ca. 96 *Ziziphus* spp.	23	7,47	-
		-	1 *Acacia tortilis* (0 bored)	<1	0,04	-	-	-	-
		-	1 Unknown Convolvulaceae (0 bored)	<1	0,05	-	-	-	-
	No.1	-	3 *Euclea divinorum* (1 bored)	<1	0,12	Ca. 2 *Euclea divinorum*	<1	0,03	Pieces of leaves and grass
		-	-	-	-	Ca. 6 *Ziziphus* spp.	1	0,33	-
	No.2	-	1 *Ziziphus* spp. (0 bored)	<1	0,01	1 *Ziziphus* spp.	<1	0,03	-
		-	2 *Euclea divinorum* (0 bored)	<1	0,01	-	-	-	-
	No.3	56	64 *Euclea divinorum* (23 bored)	8	1,84	Ca.1198 *Euclea divinorum*	177	33,30	Scaraboidea thorax
		-	19 *Ziziphus* spp. (5 bored)	5	1,51	Ca. 219 *Ziziphus* spp.	56	17,42	Cetoninae pronotum
		-	2 *Diospyros lycioides* (1 bored)	<1	0,08	Ca. 2 Unknown no.7	<1	0,12	-
	No.4	101	34 *Euclea divinorum* (7 bored)	4	1,16	Ca. 444 *Euclea divinorum*	63	11,52	-
		-	14 *Ziziphus* spp. (6 bored)	4	1,17	Ca. 109 *Ziziphus* spp.	33	10,76	-
		-	1 *Diospyros lyciodes* (0 bored)	<1	0,10	1 Unknown no.8	<1	<0,01	-
		-	1 Unknown no.7 (0 bored)	<1	0,01	-	-	-	-
	No.5	139	-	-	-	Ca. 560 *Euclea divinorum*	110	19,69	-
		-	-	-	-	Ca. 140 *Ziziphus* spp.	35	10,45	-
		-	-	-	-	1 *Acacia* spp.	<1	0,01	-
Vaalkop 2	Nest	-	-	-	-	Ca. 16 *Ziziphus* spp.	3	0,65	-
		-	-	-	-	1 Unknown no.3	<1	0,03	-
	No.1	24	-	-	-	-	-	-	-

Notes: 1. The prefix cf. ("confer") indicates uncertain identification.
2. 10 of the less common seed varieties (no.1 - no.10) could not be identified.
3. The prefix Ca. ("circa") indicates the approximate number of complete seed cases found.

Table 8ii Contents of each storage site at the 11 pouched mouse burrows excavated at four localities in South Africa

Burrow number	Storage site number	Faeces (n)	Intact seeds (number bored) (n)	Volume (ml)	Mass (g)	Seed Cases (n)	Volume (ml)	Mass (g)	Other material
Vaalkop 3	Nest	-	-	-	-	-	-	-	-
Vaalkop 4	Nest	44	30 *Ziziphus* spp. (0 bored)	9	4,38	Ca. 29 *Ziziphus* spp.	7	2,88	Pieces of leaves and grass
		-	224 *Euclea divinorum* (17 bored)	15	8,57	-	-	-	-
		-	6 *Diospyros lycioides* (1 bored)	<1	0,37	-	-	-	-
		-	1 *Acacia tortilis* (0 bored)	<1	0,02	-	-	-	-
	No.1 & 2	289	242 *Euclea divinorum* (7 bored)	17	10,88	Ca. 55 *Euclea divinorum*	6	0,78	Lower jaw of *Otomys* spp. (Rodentia)
		-	105 *Ziziphus* spp. (0 bored)	30	14,11	Ca. 39 *Ziziphus* spp.	10	3,66	-
	No.3	32	-	-	-	-	-	-	-
	No.4 & 5	189	3 *Euclea divinorum* (1 bored)	<1	0,06	Ca. 199 *Euclea divinorum*	18	2,74	-
		-	6 *Ziziphus* spp. (0 bored)	<1	0,03	Ca. 94 *Ziziphus* spp.	27	10,59	-
		-	1 *Acacia tortilis* (0 bored)	<1	0,03	-	-	-	-
		-	-	"	-	1 Unknown no.3	<1	<0,01	-
Doornhoek 1	Nest	28	17 *Acacia karoo* (3 bored)	1	0,65	Ca. 419 *Acacia karoo*	30	6,02	Pieces of leaves, grass and twigs
		-	-	-	-	1 cf. *Ziziphus* spp.	1	0,12	1 Nitidulidae; 1 Cucujoidea
		-	-	-	-	1 Unknown no.2	<1	0,01	-
		-	-	-	-	Ca. 4 Unknown no.4	<1	0,03	-
	No.1	2	3 *Acacia karoo* (0 bored)	<1	0,08	Ca. 15 *Acacia karoo*	1	0,13	Pieces of leaves and bark
		-	-	-	-	1 Unknown no.3	<1	0,01	-
	No.2 & 3	5	-	-	-	-	-	-	-
	No.4	20	5 *Acacia karoo* (0 bored)	<1	0,10	Ca. 23 *Acacia karoo*	2	0,36	3 Nitidulidae
		-	-	-	-	1 Unkown no.4	<1	0,01	-
	No.5	8	-	-	-	Ca. 4 *Acacia karoo*	<1	0,05	Pieces of twig
	No.6	27	4 *Acacia karoo* (0 bored)	<1	0,17	Ca. 33 *Acacia karoo*	2	0,60	2 Nitidulidae
		-	1 Unknown no.4	<1	0,02	1 Unknown no.4	<1	0,01	-
	No.7	16	-	-	-	Ca. 63 *Acacia karoo*	4	0,63	-
		-	-	-	-	1 cf. *Ziziphus* spp.	1	0,08	-
		-	-	-	-	1 Unknown no.2	<1	0,03	-
		-	-	-	-	Ca. 17 Unknown no.4	<1	0,15	-
Doornhoek 2	Nest & No.3	19	3 *Acacia karoo* (0 bored)	<1	0,11	Ca. 176 *Acacia karoo*	10	1,64	*Acacia* pods
		-	-	-	-	Ca. 3 Unknown no.4	<1	0,01	-
	No.1	3	1 *Acacia karoo* (0 bored)	<1	0,04	Ca. 3 *Acacia karoo*	<1	0,04	Pieces of stick and bark; 2 Millipedes;
		-	-	-	-	-	-	-	4 Chrysomelidae elytra; 1 Scarabaeidae leg
	No.2	-	-	-	-	Ca. 9 *Acacia karoo*	<1	0,10	1 Chrysomelidae elytra
	No.4	30	13 *Acacia karoo* (4 bored)	<1	0,21	Ca. 243 *Acacia karoo*	13	2,68	10 Chrysomelidae elytra; 1 Millipede;
		-	2 *Ziziphus* spp. (1 bored)	<1	0,05	Ca. 4 Unknown no.4	<1	0,01	1 Tenebrionidae larva
	No.5	-	1 *Acacia karoo* (0 bored)	<1	0,04	Ca. 26 *Acacia karoo*	2	0,33	-
		-	-	-	-	1 cf. *Ziziphus* spp.	<1	0,01	-
		-	-	-	-	1 Unknown no.4	<1	0,01	-
	No.6	3	-	-	-	Ca. 43 *Acacia karoo*	3	0,51	Pieces of stick and *Acacia* pods
		-	-	-	-	Ca. 2 Unknown no.4	1	0,01	-
	No.7	7	-	-	-	Ca. 63 *Acacia karoo*	3	0,64	Pieces of stick and bark
		-	-	-	-	-	-	-	2 Chrysomelidae elytra; 1 Nitidulidae
	No.8	1	161 *Acacia karoo* (11 bored)	12	5,52	Ca. 70 *Acacia karoo*	6	1,24	1 Millipede; 4 Chrysomelidae elytra
		-	3 *Ziziphus* spp. (0 bored)	2	0,35	1 cf. *Ziziphus* spp.	<1	0,02	1 Chrysomeldae; 1 Tenebrionidae thorax
		-	15 *Diospyros lycioides* (10 bored)	6	1,05	Ca. 3 Unknown no.5	<1	0,02	-
		-	8 Unknown no.6 (0 bored)	2	0,15	1 Unknown no.6	<1	0,01	-

Table 8iii Contents of each storage site at the 11 pouched mouse burrows excavated at four localities in South Africa

Burrow number	Storage site number	Faeces (n)	Intact seeds (number bored) (n)	Volume (ml)	Mass (g)	Seed Cases (n)	Volume (ml)	Mass (g)	Other material
Pniel 1	Nest	10	61 *Acacia tortilis* (0 bored)	3	2,28	Ca. 479 *Acacia* spp.	54	17,01	Pieces of grass, sticks and leaves
		-	-	-	-	Ca. 26 *Acacia erioloba*	8	2,69	-
		-	-	-	-	Ca. 11 *Acacia karoo*	<1	0,17	-
		-	-	-	-	Ca. 5 *Ziziphus* spp.	<1	0,27	-
		-	1 cf. *Ziziphus* spp. (0 bored)	<1	0,02	Ca. 3 cf. *Ziziphus* spp.	<1	0,16	-
	No.1	9	95 *Acacia* cf. *tortilis* (0 bored)	5	3,79	Ca. 5 *Acacia* cf. *tortilis*	<1	0,10	-
		-	-	-	-	1 *Acacia* cf. *erioloba*	<1	0,07	-
		-	-	-	-	Ca. 155 *Acacia* spp.	10	3,73	-
		-	4 *Ziziphus* spp. (0 bored)	<1	0,27	Ca. 8 *Ziziphus* spp.	1	0,34	-
	No.2	18	20 *Acacia tortilis* (0 bored)	1	0,70	Ca. 30 *Acacia* cf. *tortilis*	3	0,84	-
		-	-	-	-	1 *Acacia* cf. *erioloba*	<1	0,07	-
		-	-	-	-	Ca. 161 *Acacia* spp.	17	5,34	-
		-	-	-	-	Ca. 3 *Ziziphus* spp.	<1	0,12	-
	No.4	88	221 *Acacia tortilis* (0 bored)	13	8,66	Ca. 805 *Acacia* spp.	68	22,86	-
		-	2 *Acacia erioloba* (2 bored)	1	0,37	Ca. 4 *Acacia erioloba*	2	0,36	-
		-	-	-	-	Ca. 9 *Acacia* cf. *erioloba*	3	1,01	-
		-	1 *Ziziphus* spp. (0 bored)	<1	0,04	Ca. 6 *Ziziphus* spp.	1	0,24	-
	No.3	11	436 *Acacia tortilis* (0 bored)	23	17,22	Ca. 612 *Acacia* spp.	45	14,41	-
		-	14 *Acacia erioloba* (0 bored)	6	3,32	Ca. 4 *Acacia erioloba*	1	0,28	-
		-	7 *Ziziphus* spp. (0 bored)	1	0,49	Ca. 33 *Ziziphus* spp.	4	1,44	-
	No.5	-	9 *Acacia tortilis* (0 bored)	<1	0,35	Ca. 35 *Acacia* spp.	3	0,92	-
		-	-	-	-	Ca. 2 *Ziziphus* spp.	<1	0,08	-
	No.6	4	158 *Acacia tortilis* (0 bored)	8	6,32	Ca. 80 *Acacia* spp.	5	1,82	-
		-	-	-	-	Ca. 2 *Acacia erioloba*	1	0,18	-
		-	-	-	-	1 *Ziziphus* spp.	<1	0,03	-
	No.7	-	4 *Acacia tortilis* (0 bored)	<1	0,18	Ca. 2 *Acacia* spp.	<1	0,03	-
	No.8	1	36 *Acacia tortilis* (0 bored)	2	1,58	Ca. 32 *Acacia* spp.	4	0,95	Pieces of grass; 1 Lagomorph pellet
		-	2 *Acacia erioloba* (0 bored)	1	0,47	Ca. 2 *Acacia erioloba*	<1	0,17	-
		-	-	-	-	1 *Ziziphus* spp.	<1	0,06	-
	No.9	-	4 *Acacia tortilis* (0 bored)	<1	0,14	-	-	-	-
		-	1 *Acacia erioloba* (0 bored)	<1	0,21	-	-	-	-
	No.10	13	24 *Acacia tortilis* (0 bored)	1	0,85	Ca. 8 *Acacia* spp.	<1	0,23	-
		-	7 *Acacia erioloba* (0 bored)	3	1,84	1 *Acacia* cf. *erioloba*	<1	0,08	-
		-	2 *Acacia* cf. *erioloba* (0 bored)	1	0,25	1 *Ziziphus* spp.	<1	0,04	-
	No.11	2	226 *Acacia tortilis* (0 bored)	18	12,91	Ca. 22 *Acacia* spp.	2	0,53	-
		-	8 *Acacia erioloba* (0 bored)	3	1,64	Ca. 2 *Acacia erioloba*	<1	0,17	-
		-	43 *Ziziphus* spp. (1 bored)	7	3,22	Ca. 17 *Ziziphus* spp.	2	0,85	-
	No.12	12	88 *Acacia tortilis* (0 bored)	6	3,76	Ca. 13 *Acacia* spp.	1	0,31	1 Lagomorph pellet
		-	5 *Acacia erioloba* (0 bored)	2	1,16	1 *Acacia erioloba*	<1	0,09	-
		-	3 *Ziziphus* spp. (0 bored)	<1	0,23	1 *Ziziphus* spp.	<1	0,06	-
	No.13	162	21 *Acacia tortilis* (0 bored)	1	0,92	1 *Acacia erioloba*	<1	0,13	-
		-	19 *Acacia* spp. (0 bored)	1	0,93	Ca. 20 *Acacia* spp.	2	0,47	-
		-	1 *Ziziphus* spp. (0 bored)	<1	0,10	-	-	-	-
		-	12 Unknown no.10 (0 bored)	3	0,13	-	-	-	-
	No.14	303	5 *Acacia tortilis* (0 bored)	<1	0,22	Ca. 10 *Acacia* spp.	1	0,22	-
		-	-	-	-	1 *Ziziphus* spp.	<1	0,01	-
		-	6 Unknown no.10 (0 bored)	5	0,43	1 Unknown no.10	<1	0,01	-

All but two of the burrows (Skukuza 4 and Vaalkop 3) contained faecal remains, although faeces were rarely segregated in specific latrines. Instead, many of the storage sites contained faeces, intact seeds and empty seed cases all together. Finally, several unusual items were found in some of the *S.campestris* burrows, and these included: a hibernating olive toad (*Bufo garmani*) in the nest of Vaalkop 3, the lower jaw of a vlei rat (*Otomys* spp.) in Vaalkop 4 and an unidentified mollusc shell and vertebrate bone fragment from the nest of Skukuza 2.

Soil temperatures

Daily extremes in soil temperatures from representative burrows at each locality are displayed in Fig. 9. Despite differences in the time of year when these measurements were taken the temperature below ground (600mm) was remarkably stable and ranged from 17,5°C at Vaalkop 1 in July, to 30,5°C at Skukuza 2 in February. Daily and seasonal variation in soil temperature declined as depth increased while most of the reduction in variability ocurred within the first 200mm (see Fig. 9 and Table 4). Likewise, geographical variation in temperature was less pronounced below ground and at a depth of 300mm ranged from 21,2-30,4°C at Skukuza, 17,8-27,0°C at Vaalkop, 13,8-27,9°C at Pniel and from 14,8-27,4°C at Doornhoek (Table 4).

Discussion

The results of the present study confirm that *S.campestris* is a solitary species (Copley 1950; Walker 1964) and although individuals were observed visiting neighbouring burrows there was no evidence of the loose social structure proposed by Ansell (1960) and implied by Smithers (1971), DeGraaff (1981) and Rautenbach (1982).

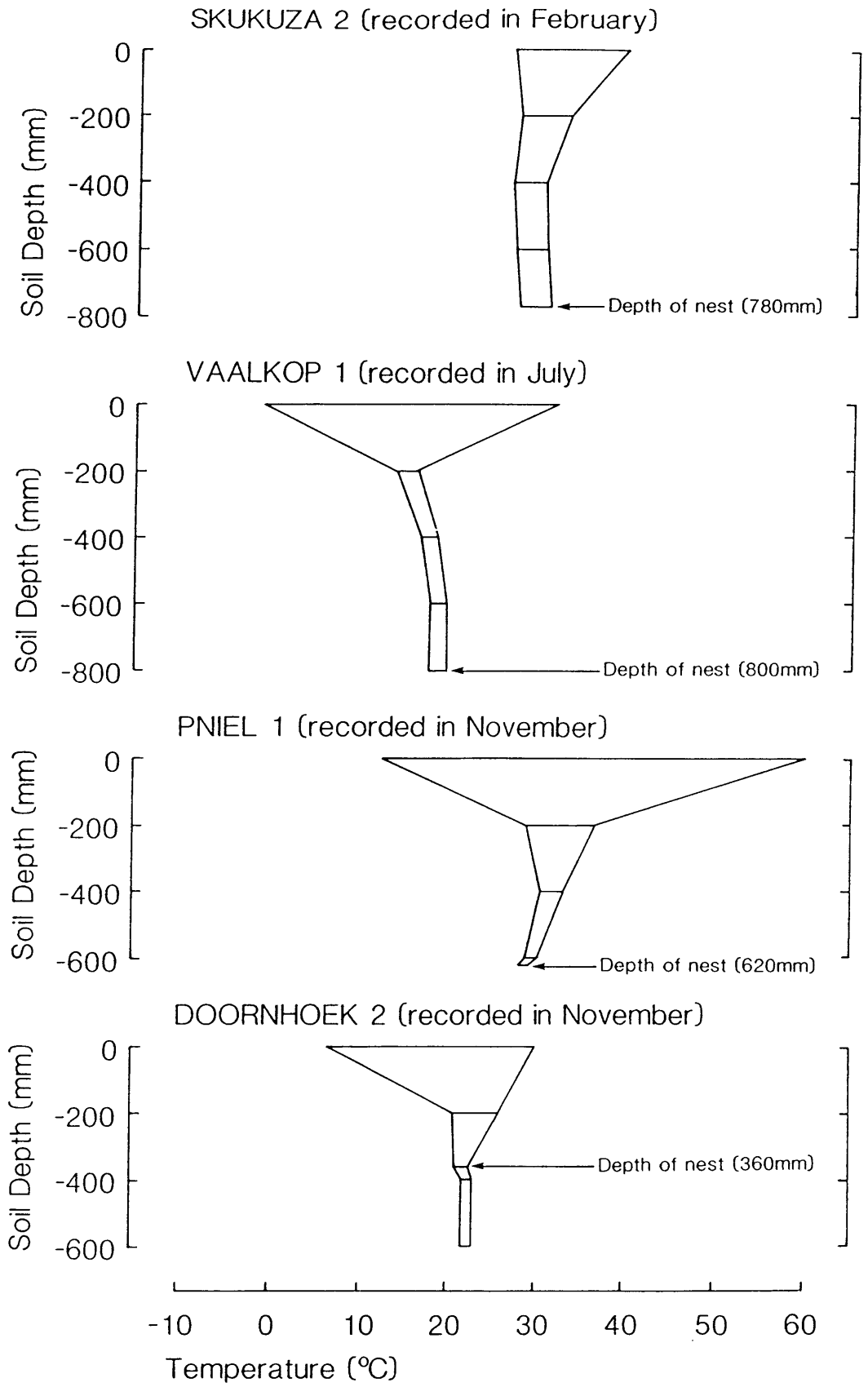


Figure 9 Daily variation in soil temperature recorded at representative burrow systems in each of the four localities in South Africa.

It remains unclear whether any of the burrows examined had actually been constructed by their occupants because previous studies have observed *S.campestris* making use of existing holes in termite mounds, under piles of rock and beneath logs, while they also nest in unoccupied burrows of gerbils (*Tatera* spp.), springhaas (*Pedetes capensis*) and aardvark (*Orycteropus afer*) (Vesey-Fitzgerald 1966; Smithers 1971). Indeed, Kingdon (1984) pointed out that the paws of pouched mice are poorly adapted for digging, while Vesey-Fitzgerald (1966) was doubtful that they ever burrow at all. Nevertheless, *S.campestris* are adept diggers in captivity (Earl 1978, pers. obs.) and Smithers (1971) maintained that they are capable of making their own burrows in sandy soils such as those recorded at localities in the present study. In view of the common features displayed by each of the burrows examined it appears that pouched mice excavated these themselves or, where they took over existing holes (such as the termite mound at Skukuza 2) modified them in a characteristic way, as observed at Vaalkop 3. At the same time, there remained considerable variation in the size and structure of *S.campestris* burrows yet there was no indication that sex, size or breeding activity could account for these differences in burrow complexity.

Although many studies indicate that *S.campestris* are omnivorous (eg. Hanney 1965; Kern 1981; Watson 1987) seeds were the only food item stored in any of the burrows examined. Indeed, Kerley (1989) caught one individual which had vegetation and insect remains in its stomach and *Acacia karoo* seeds in its pouches. It therefore appears that pouched mice preferentially hoard seeds, possibly because these are the least perishable component of their diet (Vander Wall 1990). At the same time, the large number of seed varieties collected by pouched mice in this (Tables 5 and 8) and previous (Table 3) studies implies that they are opportunistic and cache whatever is available. However, few of the seeds found in the traps or pouches of live-caught individuals had been attacked by seed-boring invertebrates (Table 5) which suggests that they may have avoided damaged seeds as reported for gray squirrels (*Scuirus carolinensis*) by Dennis (1930). Likewise, many of these hoards comprised more than one type of seed which indicates that pouched mice

may have selected a variety of seeds rather than concentrating on the most abundant type available (Reichman & Fay 1983).

Assuming that 10ml represents the maximum pouch capacity of *S.campestris* (Table 5) then the intact seeds and seed remains recovered from the five burrows with appreciable hoards (Vaalkop 1, 4; Pniel 1; and Doornhoek 1 & 2) represent the result of at least 70 foraging trips (see Table 9). Likewise, if each of the pouched mice travelled as far from their burrows to collect seeds as they did when they were caught (ie. a mean distance of 26,2m; Table 4) then they would have travelled between 100m and 1,86km to gather these and bring them back to their burrow (Table 9). However, these distances are undoubtedly underestimates because the mice usually transported less than 10ml of food per trip (Table 5) while the volume of seed cases left in each burrow would be less than the original volume of intact seeds collected.

Table 9 An estimation of the distance travelled by pouched mice to collect the seeds and seed cases found in each of the 11 burrows excavated.

Burrow number	Total volume of intact seeds and seed cases (ml)	Equivalent number of foraging trips (@10ml per trip)	Total distance covered (@26,2m per trip)
Skukuza 1	-	-	-
Skukuza 2	46	4,6	120,5
Skukuza 3	-	-	-
Skukuza 4	-	-	-
Vaalkop 1	709	70,9	1857,6
Vaalkop 2	3	0,3	7,9
Vaalkop 3	-	-	-
Vaalkop 4	139	13,9	264,2
Pniel 1	353	35,3	924,9
Doornhoek 1	42	4,2	110,0
Doornhoek 2	60	6,0	157,2

Nevertheless, it is clear that burrow hoards represent a substantial investment in time and energy for resident pouched mice and for this reason one might expect burrow occupants to protect their larders from potential thieves. These would include the conspecifics and other rodent species which were observed entering *S.campestris* burrows as well as the seed-boring invertebrates whose activity was recorded in the present study. Eastern chipmunks (*Tamias striatus*) and bannertail kangaroo rats (*Dipodomys spectabilis*), both of which collect large quantities of seeds in their burrows, defend these by establishing territories and chasing away conspecifics and other rodent species (Elliott 1978, Randall 1984). Unfortunately, no comparable field observations have been made for pouched mice. However, captive individuals are often intolerant of other cage occupants (Choate 1972) and this might indicate that they are equally aggressive towards conspecifics in the wild. At the same time it is possible that pouched mice protect their larders from rodent thieves by blocking the entrance to their burrow, a behaviour which has long been identified as an anti-predator mechanism in semi-fossorial rodents (eg. Burns, Flath & Clark 1989). Sealing the entrance would be particularly useful when the burrow is unoccupied because it might prevent other animals gaining access to unprotected seed stores. However, this was only observed on three occasions in the present study, and two of these occurred at burrows (Vaalkop 3 and Skukuza 1) which contained little or no food. Meanwhile, it appears that *S.campestris* are not capable of protecting their larders from infestation by seed-boring invertebrates because all of the burrows with seed stores contained some seeds which had been bored (Table 8). However, no potential seed-borers were identified among the invertebrate remains collected which suggests that ovipositing adults and/or emerging larvae were consumed by resident pouched mice.

According to Vander Wall (1990) food hoarding has three possible functions: 1. to minimise the time spent foraging, 2. to ensure a constant supply of food during periods of food scarcity, and 3. to provide food for developing offspring. Reducing the time spent foraging allows animals to devote more time to other activities and can be beneficial when foraging involves an increase in energy expenditure and/or

higher risks of predation (Vander Wall 1990). Pettifer & Nel (1975) found that pouched mice hoarded freely in captivity but rarely ate outside their nests and they suggested that this was an adaptation to avoid predation. Likewise, Smithers (1971) found large accumulations of seed remains outside the entrance to *S.campestris* burrows and ascribed this to their habit of eating near the security of their burrow. This would explain why hoarding activity was observed at different times of the year in the present study, and also by one individual (the female from Vaalkop 3; see Table 5) whose burrow contained no stores of seeds (Table 8). However, some food items (vegetation and insects) are rarely, if ever, found in the pouches of live-caught pouched mice (Table 3 and 5) which implies that these are consumed wherever they are found and are not transported back to the burrow to be eaten in safety. Alternatively, these food items may be observed less often than seeds simply because *S.campestris* collect more seeds than other components of their diet and use these not only as an immediate source of food but also to stock their burrows with a supply for periods when food is scarce.

Indeed, food availability can vary dramatically between wet and dry seasons at each of the localities examined and Walker (1964) suggested that *S.campestris* hoard seeds in summer for use during winter. Unfortunately, the present study could not confirm whether seasonal variation in food hoarding occurs in this species and although seeds were found in burrows excavated throughout the year, some of these may have been left over from winter or even from previous years as described for red squirrels (*Tamiasciurus hudsonicus*) by Smith (1968). In this context, summer stores of food might play an important role in the successful development of young pouched mice by permitting the female to spend more time with her offspring, by introducing them to hard foods within the security of the nest, and/or by ensuring that they reach an optimal size before they disperse from the maternal burrow (Earl 1978; Vander Wall 1990; Willan 1990; Ellison et al. in press). Meanwhile, it is interesting to speculate whether the large stores of seeds found in several of the more complex burrow systems are the result of the activities of more than one occupant as suggested for chipmunks (Elliott 1978) and kangaroo rats

(Randall 1984). If similar "traditional" burrows are used by successive generations of pouched mice it would be advantageous if subsequent occupants (such as the female which moved to Vaalkop 4) were closely related so that any unused food which they inherited benefitted the previous occupant through kin selection (Andersson & Krebs 1978). This aspect of pouched mouse biology is worthy of future study.

All of the burrows in the present study provided a degree of thermal protection to their inhabitants because seasonal, daily and geographical variation in temperature was greatly reduced even at the depth of the shallowest nesting chambers (Fig. 9 and Table 4). Similar reductions in temperature variability have been reported for other small mammal burrows including those of *Gerbillurus* spp. from southern Africa (Downs & Perrin 1989). Meanwhile, Bennett, Jarvis & Davies (1988) pointed out that annual soil temperatures in rodent mole burrows only stabilise at depths greater than 600mm. As only three of the burrows examined had nests below this level, most of the occupants would experience some seasonal variation in nest temperature ranging from around 30°C in summer to 15°C in winter (Table 2). Although the former falls within the thermoneutral zone of summer acclimated pouched mice the latter is well below the lower critical temperature of 28°C recorded in winter acclimated individuals (Ellison & Skinner 1991b; Haim et al. 1991). However, several studies have indicated that occupied nests of small mammals are anything between 8°C (Stark 1963) and 21°C (Sealander 1952) warmer than ambient which suggests that the temperature of occupied *S.campestris* nests may rise above the lower critical temperature in winter. At the same time, these nests might also become too warm in summer and exceed the upper critical temperature of ca. 32°C (Ellison & Skinner 1991b; Haim et al 1991), particularly in warmer localities such as Skukuza. This might explain why little bedding material was found in many of the shallower burrows excavated during summertime.

In conclusion, the results of the present study confirm that *S.campestris* are solitary and would not normally be able to huddle in the wild. However, pouched

mice are capable of excavating or modifying their own burrows which provide a more stable thermal environment than that prevailing above-ground. Although several of the burrows contained considerable stores of food, the specific function(s) of food hoarding remains unclear because hoarding activity and burrow stores were observed at different times of the year, while some burrows contained no seeds at all.

CHAPTER 3: ENERGETIC BEHAVIOUR OF POUCHED MICE: NEST-BUILDING AND FOOD-HOARDING UNDER LABORATORY CONDITIONS

Introduction

Behavioural adaptations are generally considered to be the principal mechanisms which enable small endotherms to cope with seasonal changes in their environment not only because behaviour usually changes faster than physiological or morphological alternatives (Gordon et al. 1986) but also because behavioural modifications often help animals to avoid environmental stresses altogether (Wunder 1984). In this way, small mammals could escape a seasonal decline in food availability or temperature simply by migrating, hoarding food or constructing a more favourable microhabitat (Wunder 1984; Vander Wall 1990; Table 2). However, migration is impracticable for terrestrial species (Wunder 1984) although food hoarding enables many small mammals to control the availability of their food in space and time (Vander Wall 1990). Likewise, the creation of a nest, or protected microhabitat, allows an animal to evade the conditions prevailing elsewhere in the environment and thus reduces its exposure to the energetic stresses therein (Fahri 1987).

Although pouched mice inhabit tropical and subtropical environments which generally experience less variation in temperature and food availability than temperate areas, in some parts of their range they occur in strongly seasonal habitats such as the subtropical woodland savannas of southern Africa (Korn 1989). Under these circumstances pouched mice might be expected to display seasonal and/or geographical variation in nest-building and food-hoarding activity. However, the field studies, conducted in Chapter 2, found no clear differences in these behaviours at different times of the year or at different localities. The aim of the present study was therefore to assess the extent of energetic behaviour among pouched mice from four localities in southern Africa by studying the effect of changes in photoperiod and temperature on their nest-building and food-hoarding activity under controlled conditions.

Materials and methods

Animals

Nest-building and food-hoarding activity was investigated in 40 pouched mice which originated from four localities in southern Africa and displayed different karyotypes (2n): *Gam* in S.E. Bushmanland, Namibia (20°27'S 20°44'E: 2n=35); *Letaba* in the Kruger National Park, South Africa (23°58'S 31°35'E: 2n=46); *Ga-Tlhose* near Sishen in the Northern Cape, South Africa (27°49'S 23°30'E: 2n=30); and *Pniel* near Barkly West, also in the Northern Cape, South Africa (28°35'S 24°31'E: 2n=32). Annual rainfall and air temperature data for each locality were obtained from the nearest meteorological station and these are summarised in Table 10.

Five adult males and five nonpregnant females from each locality were studied. Approximately equal numbers of these were derived from wild-caught (n=19) and first generation laboratory-bred (n=21) stocks. All laboratory-bred mice were reared under long photoperiod (14L:10D) at 25°C and were at least five months old at the start of the present study. Wild-caught animals had been kept under similar conditions for at least five months before the present study began.

The pouched mice were housed separately in standard laboratory rat cages (405mm x 240mm) with a 7,0 mm hardboard false floor supported by a wooden frame which created a 580mm long, L-shaped tunnel, 65mm wide and 45mm high that was accessible through a 38mm diameter hole at one corner of the cage (see Fig. 10).

This false floor provided the occupant of each cage with an artificial burrow which could be used to construct a nest and hoard food, and had a similar vertical entrance to that found in the wild (see Chapter 2). To ensure that the pouched mice were fully accustomed to these cages they were transferred to them at least six weeks before the experiments began. Each individual spent the entire experiment with the same false floor and artificial burrow so that they were not exposed to different burrows containing the scent of previous occupants, which can influence hoarding behaviour (Miller & Viek 1944).

Table 10 Climatic conditions at the four pouched mouse localities studied in Chapter 3

Locality	(Meteorological Station)	Altitude (m)	Mean Annual Rainfall (mm)	Annual Mean Daily Air Temperatures Maximum (°C)	Minimum (°C)	Range (Max.-Min.) (°C)
1. *Gam*	(Grootfontein 19°36'S 18°08'E)	1398	611	28,3	12,6	15,8
2. *Letaba*	(Letaba 23°51'S 31°35'E)	215	387	30,4	15,8	14,7
3. *Ga-Tlhose*	(Sishen 27°42'S 22°59'E)	1204	386	26,6	11,5	15,4
4. *Pniel*	(Barkly West 28°32'S 24°32'E)	1204	391	26,6	9,7	16,9

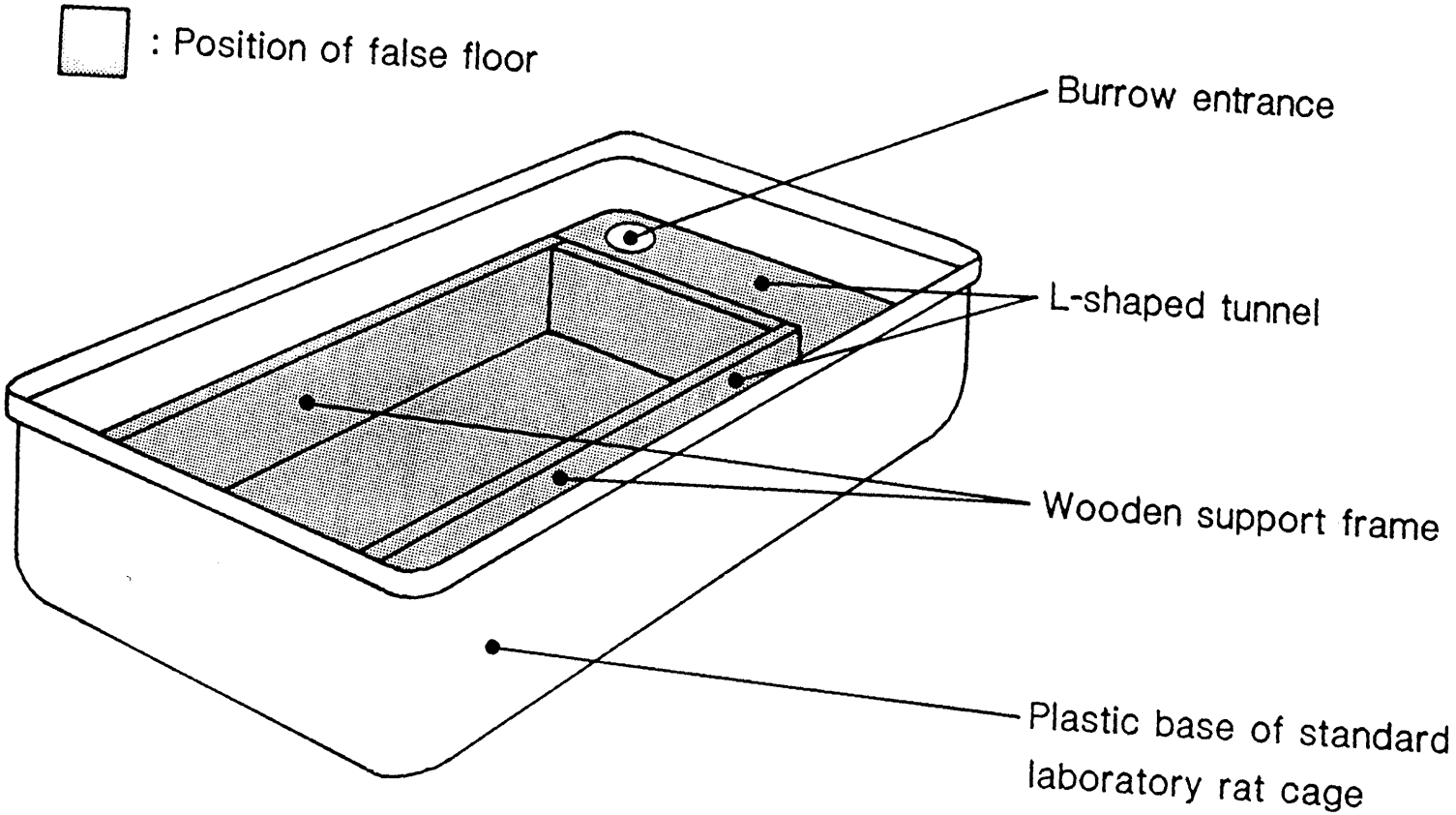


Figure 10 The base of a modified rat cage, showing the position of the false floor, wooden support frame and artificial burrow.

The pouched mice were fed a standard diet of rat cubes, rabbit pellets (Epol, Vereeniging) and water *ad libitum* and this was supplemented with sunflower seeds, apples and carrots during weeks when no measurements were made. Shredded paper, sawdust and sand were provided for bedding and the cages were cleaned weekly, and dusted with insecticide to eradicate ectoparasites (Karbadust: Sentrachem, Silverton. Containing 50g/kg Carbamate). Relative humidity was maintained above 45% to prevent ringtail (Ellison & Westlin-Van Aarde 1990).

Acclimation

Throughout the study the pouched mice were housed in a windowless climate chamber in which photoperiod and temperature could be accurately controlled. Within this chamber they were subjected to a nine week protocol comprising three weeks acclimation under long photoperiod (14L:10D) at 25 ± 1°C followed by six weeks under short photoperiod (10L:14D) in which the first three weeks were spent at 25 ± 1°C and the last three weeks at 15 ± 1°C. During the third (test) week of acclimation under each photoperiodic/temperature regime the following measurements were made:

Body mass

Body mass (M_b) was recorded using a PB3000 Mettler Scale (Mettler, Zurich). Animals were restrained within a cotton sack, and M_b was measured to the nearest 0,1g.

Nest-building activity

At the beginning of each test week the cages were thoroughly cleaned and fresh bedding was supplied as follows: 400ml of dry, sifted river sand (Mixcrete, Silvertondale) was distributed equally along the artificial burrow and two sheets of shredded tissue paper (Kleenex perforated medical towels: Kimberly-Clark, Johannesburg) weighing 5,2g, was placed in the cage, one above and one below the false floor. An additional sheet of shredded tissue paper was added to the cage after

two and/or four days if the pouched mouse had taken all the tissue down to its burrow. The total amount of tissue used for nesting at the end of the week was then calculated as the difference between the mass of tissue provided and that remaining above the false floor. However, if there was more tissue above the floor than below it, the pouched mouse was assumed to have nested outside the burrow. Under these circumstances the total amount of paper used for nesting was determined as the mass of tissue left above the false floor. On occasion, some individuals also blocked the entrance to the artificial burrow with sand, paper and/or food and this was recorded whenever it occurred.

At the end of the test week each pouched mouse was removed from its cage so that the cooling coefficient of their nest could be estimated using the temperature change constant, ***K***, of Chew, Lindberg & Hayden (1967) where:

$$K = \frac{\ln(\text{Temperature in °C at time 1}) - \ln(\text{Temperature in °C at time 2})}{\text{The difference between time 1 and time 2 in minutes}}$$

The cages were first heated to 42.0 $\pm$5.1°C in an oven and then placed on a bench at room temperature (22.2 $\pm$1.3°C). A J-type thermocouple was then inserted into the nest (whether this was above or below the false floor of the cage) and allowed to equilibrate with nest temperature for at least 10min. The temperature of the nest was subsequently recorded for about half an hour using a data logger (1200 Series Squirrel: Grant instruments, Cambridge).

Food consumption

At the start of each test week approximately 30g (range: 29,3-33,9g) of rat cubes and 30g (range: 29,8-33,3g) of rabbit pellets were placed on the false floor of each cage. These two varieties of food were of different diameter (rat: 13-14mm, rabbit: 5-6mm) and had different minimum protein (rat: 20%, rabbit: 16%) and maximum fibre contents (rat: 6%, rabbit: 17%). Daily consumption of both food types was estimated by subtracting the amount of food left at the end of the week

from that provided at the beginning and dividing this by seven (days). To compensate for any differences in body mass food consumption was expressed per $[M_b]^{0,75}$ as suggested by Hart (1971).

Food hoarding

In the present study hoarded food was defined as any food which was removed from the false floor of the cage and cached in the artificial burrow. Food hoarding was thus determined by measuring the mass of any rat and/or rabbit pellets remaining on the false floor of the cage after the first two days of each test week. This was subtracted from the total amount of food given at the beginning of the week and divided by two (days) to provide an estimate of the amount of food hoarded per day. To correct for the amount of unstored food which had been eaten by the pouched mice the daily food consumption (uncorrected for body mass) of each individual was subtracted from their estimate of daily food hoarding.

Statistical analyses

All results are presented as mean $\pm$ one standard deviation. Spearman's rank correlation coefficient (SRC), the Chi-squared test (X^2) and analyses of variance with no repeated measures (ANOVA) were used for the statistical analysis of results (Sokal & Rohlf 1981)

Results

Body mass

The mean M_b of male and female pouched mice from all four localities are given in Table 11. There was a significant interactive effect of sex and locality such that males from *Gam* and *Pniel* were heavier than females while the reverse was true for animals from *Letaba* and *Ga-Tlhose* (ANOVA: F=3,879, d.f.= 3 & 96, $p<0,05$). However, there was no independent effect of sex (F=0,004, d.f.= 1 & 3, $p>0,05$), locality (F=0,408, d.f.= 3 & 3, $p>0,05$) or acclimation regime (F=0,683, d.f.= 2 & 96, $p>0,05$) on M_b. Likewise there was no significant difference in the mean M_b

of wild-caught (73,5 ±12,0g) and laboratory-bred (71,7 ±15,5g) individuals (F=0,020, d.f.= 1 & 94, p>0,05). There was also no significant difference between the mean body mass of mice nesting inside and outside their burrow (F=2,771, d.f.= 1 & 119, p>0,05) nor between those animals which blocked the entrance to their burrows and those that didn't (F=3,785, d.f.= 1 & 94, p>0,05)

Table 11 Mean body mass (M_b) of male and female pouched mice from each locality examined in Chapter 3

Locality	Male M_b (g) n = 5	Female M_b (g) n = 5
Gam	80,0 ± 11,8	71,8 ± 11,1
Letaba	69,7 ± 22,0	71,2 ± 10,3
Ga-Tlhose	61,6 ± 10,7	75,9 ± 15,9
Pniel	78,7 ± 08,2	71,5 ± 16,4

Nest-building activity

25 of the 120 nests analysed in the present study were constructed outside the artificial burrow. The incidence of this behaviour appeared to be random as there was no significant difference in the nesting habits of pouched mice from different localities (X^2=0,874, d.f.= 3, p>0,05) nor was there any significant effect of photoperiod (X^2=0,328, d.f.= 1, p>0,05) or temperature (X^2=1,195, d.f.= 1, p>0,05) on the location of nests. Likewise, a similar number of wild-caught and laboratory-bred individuals nested above the false floor of the cage (X^2=0,010, d.f.= 1, p>0,05) as did a similar number of males and females (X^2=0,110, d.f.= 1, p>0,05). However,

because additional sheets of tissue were only supplied to those mice which transferred all this bedding to their burrow, these individuals used significantly more tissues than those nesting outside the burrow (ANOVA: F=12,175, d.f.= 1 & 119, p<0,001). Similarly, because hoarded food was defined as that cached inside the artificial burrow, pouched mice which nested there "hoarded" significantly more rat cubes than those which nested outside (F=31,069, d.f.= 1 & 119, p<0,001). It was therefore decided to exclude data from individuals nesting above the false floor of their cage in order to restrict the analyses to animals which displayed the same type of nesting behaviour (ie. nesting within the artificial burrow).

Among these pouched mice there was a significant interactive effect of locality and sex on the mass of tissue carried into their artificial burrow such that males from *Ga-Tlhose* and *Pniel* collected more bedding than females whilst the reverse was true for animals from *Gam* and *Letaba* (ANOVA: F>6,183, d.f.= 3 & 62, p<0,01). However, there was no independent effect of sex (F=0,031, d.f.= 1 & 3, p>0,05), locality (F=0,364, d.f.= 3 & 3, p>0,05), photoperiod (F=1,237, d.f.= 1 & 64, p>0,05) or temperature (F=0,070, d.f.= 1 & 62, p>0,05) on the mass of tissue used. At the same time, there was no significant correlation between the amount of tissue used and the cooling coefficient of the nest, ***K*** (SRC: r_s=0,084, n=25, p>0,05) and there was also no significant effect of sex (F<0,852, d.f.= 1 & 64, p>0,05), locality (F<1,297, d.f.= 3 & 64, p>0,05), photoperiod (F=2,914, d.f.= 1 & 64, p>0,05) or temperature (F=0,015, d.f.= 1 & 62, p>0,05) on ***K***.

The blocking of burrow entrances was observed 45 times in the present study and there was significant variation in the incidence of blocking among pouched mice from different localities (X^2=8,34, d.f.= 3, p<0,05) with animals from *Ga-Tlhose* blocking their burrows 19/30 times, those from *Letaba* 13/30 times, those from *Gam* 12/30 times and those from *Pniel* only 2/30 times. However, there was no significant difference in the number of male and female pouched mice which blocked the entrance to their burrows (X^2=0,409, d.f.= 1, p>0,05) nor in the number of wild-caught or laboratory-bred animals that did so (X^2=0,074, d.f.= 1, p>0,05). There was also no significant effect of photoperiod (X^2=0,825, d.f.= 1, p>0,05) or temperature

(X^2=1,333, d.f.= 1, p>0,05) on the number of pouched mice which blocked their burrows. At the same time, nests within blocked burrows had a significantly lower mean cooling coefficient (***K*** =0,011 ±0,003) than unblocked burrows (***K*** =0,013 ±0.003; ANOVA: F=13,372, d.f.= 1 & 94, p<0,001) whilst animals which blocked the entrance to their burrows also ate and hoarded significantly more rat cubes than those which did not (rat cubes eaten: F=7,688, rat cubes hoarded: F=6,782, d.f.= 1 & 94, p<0,01).

Food consumption

The mean daily consumption of rat cubes and rabbit pellets by animals from all four localities during each acclimation period is displayed in Fig. 11 (above). There was a pronounced preference for rat cubes and pouched mice consumed significantly more of these than rabbit pellets during all three acclimatory regimes (ANOVA: F>26,087, d.f.= 1 & 65, p<0,001). Males consumed significantly more rat cubes than females (F>4,766, d.f.= 1 & 62, p<0,05) while there was a significant effect of both photoperiod (F=8,026, d.f.= 1 & 64, p<0,01) and temperature (F=17,935, d.f.= 1 & 62, p<0,001) on the intake of rat cubes which decreased when photoperiod declined from 14L:10D to 10L:14D and increased again when temperature fell from 25°C to 15°C. There was also a significant effect of locality on the consumption of rat cubes (F>2,935, d.f.= 3 & 62, p<0,05) and individuals from *Ga-Tlhose* consumed the most, those from *Pniel* the least, while animals from *Gam* and *Letaba* ate an intermediate amount. However, there was no significant effect of either locality (F<1,892, d.f.= 3 & 64, p>0,05) or photoperiod (F=0,001, d.f.= 1 & 64, p>0,05) on the daily intake of rabbit pellets, although there was a significant increase in the consumption of these when temperature fell from 25°C to 15°C (F=14,481, d.f.= 1 & 62, p<0,001) at which time males ate significantly more than females (F=8,315, d.f.= 1 & 62, p<0,01).

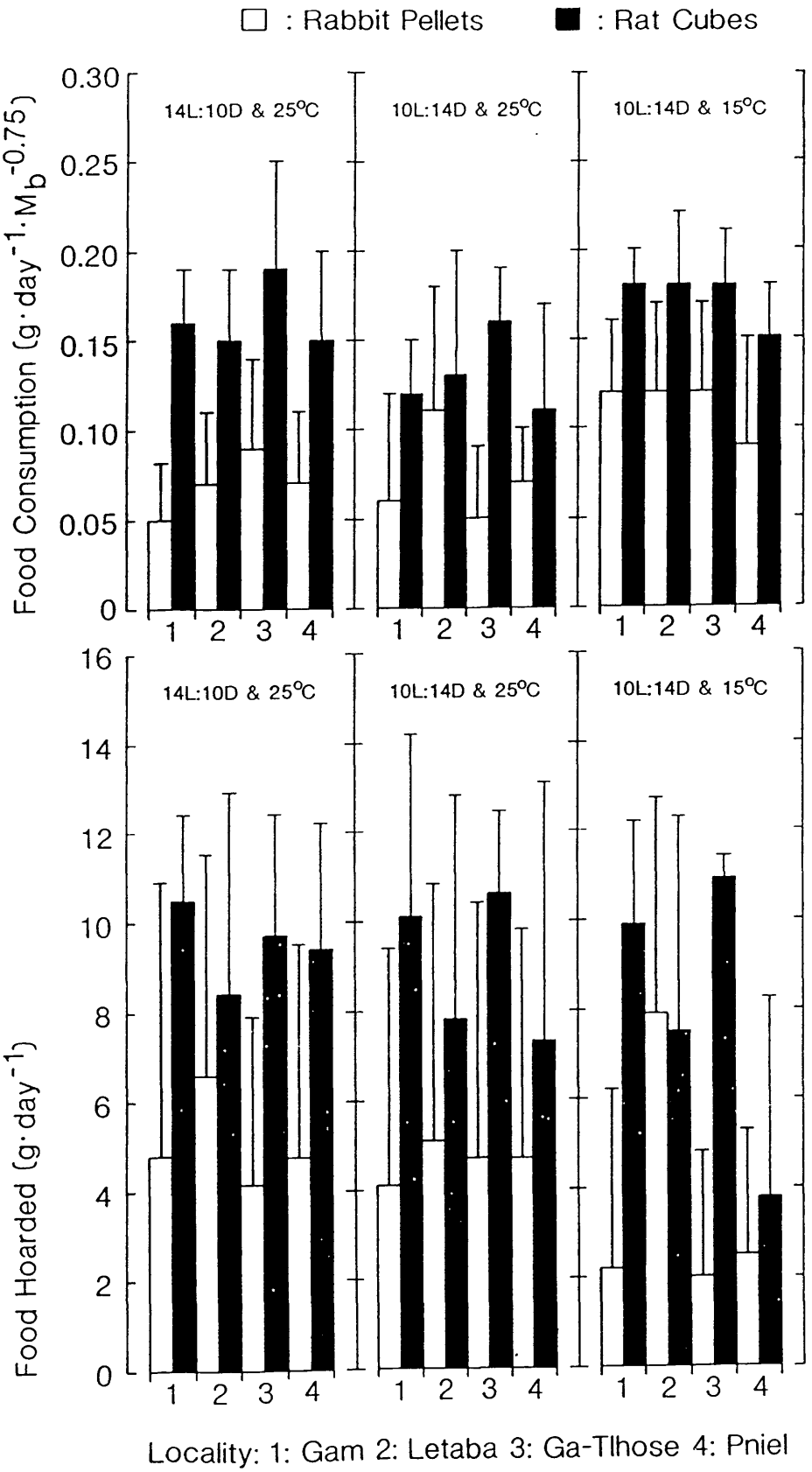


Figure 11 The mean daily mass of rat cubes and rabbit pellets consumed (above) and hoarded (below) by pouched mice from each locality during all three acclimation periods.

Food hoarding

The mean amount of food hoarded by pouched mice from each locality throughout the present study is also displayed in Fig. 11 (below). As for food consumption, there was a clear preference for rat cubes and significantly more of these were hoarded than rabbit pellets during each acclimatory regime (ANOVA: $F>17.05$, d.f.= 1 & 59, $p<0,001$). At the same time there was a significant positive correlation between the mass of rat cubes and rabbit pellets hoarded (SRC: $r_s=0,426$, n=25, $p<0,001$). However, for both types of food there was no significant effect of photoperiod (rat cubes: $F=0,179$, rabbit pellets: $F=0,147$, d.f.= 1 & 64, $p>0,05$), temperature (rat cubes: $F=0,544$, rabbit pellets: $F= 0,588$, d.f.= 1 & 62, $p>0,05$) or sex (rat cubes: $F<1,774$, rabbit pellets: $F<2,786$, d.f.= 1 & 64, $p>0,05$) on the amount hoarded. Meanwhile, under short photoperiod (10L:14D) there was a significant effect of locality on the mass of rat cubes hoarded ($F=4,730$, d.f.= 3 & 62, $p<0,01$) and individuals from *Ga-Tlhose* hoarded the most, those from *Pniel* the least, while animals from *Gam* and *Letaba* hoarded an intermediate amount.

Discussion

The absence of a correlation between the mass of tissues collected by pouched mice and the cooling constants of their nests clearly indicates that nest size is not necessarily a reliable measure of nest quality. This would occur if only some of the tissue removed by each individual actually contributed to the insulation of their nest. Moreover, Lynch & Gendler (1980) have reported extensive intraspecific variation in the quantity of nesting material (cotton wool) removed from food hoppers by *Peromyscus leucopus*, while Rajendram, Brain, Parmigiani & Mainardi (1987) even found that gerbils (*Meriones unguiculatus*) constructed warmer nests with less paper at 15°C than at 22°C. For this reason the significant interactive effect of sex and locality on the quantity of tissues used by pouched mice in the present study may have been due to factors unrelated to nest insulation, particularly as there was no similar variation in the cooling constant of their nests.

Meanwhile, there was no significant increase in either the size or insulation of *S.campestris* nests when temperature fell from 25°C to 15°C. This suggests that pouched mice don't display an increase in nesting activity in response to a drop in temperature or that they constructed nests with minimal cooling constants at both of the temperatures used in the present study. Either of these possibilities would explain why there was no geographical variation in nesting behaviour as reported for *Peromyscus* spp. by King, Maas & Weisman (1963). However, Wolfe (1970) and Hartung & Dewsbury (1979) were unable to replicate King et al.'s (1963) results and suggested that the microclimate experienced by different species might be a more important determinant of laboratory nesting behaviour than the macroclimate of their original locality. In particular, they suggested that burrow-dwelling species probably experience minimal variation in temperature conditions and thus do not display extensive nesting ability. While the subterranean nests of *S. campestris* contain little bedding material they probably provide their occupants with adequate thermal protection because they are usually more than 200mm below ground at which depth they experience limited variation in temperature (see Table 4 and Fig. 9).

The significant reduction in heat loss from nests within plugged burrows implies that these may have been blocked to reduce heat exchange. However, if the purpose of burrow plugging was solely to improve the thermal insulation of a nest we might expect an increase in the number of burrows blocked at lower temperatures while this was not the case. We might also expect individuals from colder localities to block their burrows more often than those from warmer areas yet those from the coolest locality (*Pniel*) blocked their burrows the least. It therefore appears that pouched mice block the entrance to their burrows for some other reason, such as protecting themselves from predators (Thomas 1974; Burns et al. 1989; Arnold, Heldmaier, Ortmann, Pohl, Ruf & Steinlechner 1991). Alternatively, they may plug their burrows to prevent the theft of their food cache (Thomas 1974; Chapter 2) because individuals with blocked burrows hoarded significantly more rat cubes than those without.

Although *S.campestris* are omnivorous their diet is dominated by seeds and has a high protein content (Chapter 2). Indeed, Perrin & Kokkin (1986) found relatively low densities of bacteria in the forestomach of this species and suggested that pouched mice may not harvest symbiotic microbes for protein. This might explain why animals in the present study displayed a significant preference for rat cubes because these contained more protein and less fibre than the rabbit pellets. Meanwhile, the higher food consumption of male pouched mice is similar to that described for male gerbils (*M.unguiculatus*) by Wong & Jones (1985) and supports the suggestion by De Wit (1972) that they are more active/mobile than females. Alternatively, these sexual differences may be due to contrasting thermoregulatory abilities such as those found in Wistar rats where females produce less heat and thermoregulate more efficiently than males (Doi & Kuroshima 1982).

The decline in food intake which occurred when photoperiod fell from 14L:10D to 10L:14D is similar to that reported for the Djungarian hamster (*Phodopus sungorus*) by Wade & Bartness (1984) and Masuda & Oishi (1988). However, in these studies the reduction in food consumption resulted from a decline in body weight and no such decline was observed among *S.campestris* in the present study. For this reason the reduction in food consumption by pouched mice remains equivocal unless it was caused by a decline in activity, increased thermolability or undetected bouts of torpor (Ellison & Skinner in press). In contrast, the increase in the intake of rat cubes and rabbit pellets which occurred following the decline in temperature is easier to explain in terms of the increased energy required for thermoregulation at lower temperatures (Masuda & Oishi 1982). At the same time, the significant variation in the food intake of individuals from different localities appears to be unrelated to climatic factors because pouched mice which ate the most (*Ga-Tlhose*) and the least (*Pniel*) food came from localities less than 120km apart with similar climatic conditions (see Table 10). For this reason, the geographical differences in food intake of *S.campestris* are probably the result of intraspecific variation in energy metabolism which will be investigated further in Chapter 5.

Not only did pouched mice preferentially consume rat cubes (see above) but they also hoarded significantly more of these than rabbit pellets in the present study. This indicates that hoarding activity was selective, and specifically aimed at the collection of preferred food items rather than the random accumulation of any available food. However, the positive correlation between the quantity of rat cubes and rabbit pellets hoarded suggests that good hoarders collected more of both food types than those individuals which hoarded less. While it is generally acknowledged that food hoarding is an adaptive response to shortages caused by fluctuations in food availability and/or energy demand (Vander Wall 1990) there are three ways in which hoarding can be adaptive: 1. it helps provide food for developing offspring, 2. it ensures a constant supply of food during periods of food scarcity, or 3. it minimises the time spent foraging (Vander Wall 1990). According to Smith & Reichman (1984) those rodents that use cached food as part of their parental care display sexual differences in hoarding activity, with females hoarding more than males. Certainly, the females of a large variety of small mammals including laboratory rats (Herberg, Pye & Blundell 1972), gerbils (*Gerbillurus paeba*, Stutterheim & Skinner 1973; *M.unguiculatus*, Nyby, Wallace, Owen & Thiessen 1973), hamsters (*Mesocricetus auratus*, Wong & Jones 1985) chipmunks (*Tamias striatus*, Brenner & Lyle 1975) and shrews (*Cryptotis parva*, Formanowicz, Bradley & Brodie 1989) consistently hoard more than males. Nevertheless, despite the fact that female pouched mice bring soft food back to the nest for their pups (Earl 1980) there were no significant sexual differences in the quantity of food hoarded in the present study which suggests that *S.campestris* do not hoard primarily to feed their young.

Several studies have shown that a decline in photoperiod and/or temperature can effect a substantial increase in food hoarding among those species that hoard food in summer or autumn to see them through a period of food shortage during winter (Lyman 1954; Brenner & Lyle 1975; Barry 1976; Dufour 1978; Masuda & Oishi 1988). For similar reasons there are often geographical differences in hoarding activity with populations from colder environments or areas where food

becomes seasonally scarce exhibiting higher levels of hoarding activity (Horton & Wright 1944; Thomas 1974; Goertz, Dawson & Mowbray 1975) which may even be detectable under artificial laboratory conditions (Barry 1976). However, there was no significant effect of photoperiod or temperature on food hoarding by pouched mice in the present study which suggests that food hoarding doesn't primarily serve to prepare for a winter decline in food availability. At the same time, the significant geographical variation in hoarding ability was identical to that observed for food intake (see above) and also appeared to be unrelated to climatic factors. However, a similar disparity was reported by Tannenbaum & Pivorun (1987a) for two *Peromyscus* spp. from contrasting altitudes which led him to conclude that climatological factors "may not be the only determinants of... (a population's) proclivity towards hoarding".

Finally, some authors have suggested that *S.campestris* hoard food primarily to reduce the amount of time spent outside the relative security of the burrow (Smithers 1971; Pettifer & Nel 1975). If this were the case we might expect hoarding activity to occur as a fundamental component of feeding activity which would explain why it was displayed by individuals of both sex under a wide range of environmental conditions. This would also explain why the geographical variation in food intake and hoarding activity was identical, because those individuals from localities with the highest food intake would also hoard the most food. Further support is provided by the fact that animals which blocked the entrance to their burrows not only hoarded more, but also ate more food than those which didn't. However, if pouched mice hoard food in direct proportion to the amount they ate we would also expect males, which ate the most food, to hoard more than females, which was not the case. Likewise, it is not clear why there was no significant change in hoarding activity when food intake decreased with a decline in photoperiod and increased again when temperature fell to 15°C. In part, this might have been due to the high degree of intraspecific variation in hoarding activity (see Fig. 11) while variation in the intake of rabbit pellets, particularly at 15°C, might have masked changes in the quantity of rat cubes hoarded.

In conclusion, the results of the present study indicate that although *S.campestris* display energetic behaviour in the form of nest-building and food-hoarding these do not improve in response to a decline in temperature when higher thermoregulatory costs necessitate an increase in food consumption. Instead, pouched mice maintain a similar degree of nesting activity regardless of photoperiod, temperature or locality, while hoarding activity appears to be associated with returning food to the nest to be eaten rather than preparing for periods when the availability and/or cost of food changes.

CHAPTER 4: MORPHOLOGICAL VARIATION OF POUCHED MICE WITHIN THE SOUTHERN AFRICAN SUBREGION

Introduction

Wunder (1978) proposed a hierarchy of energy use for small mammals in which thermoregulation assumed priority, because an individual that is not thermoregulating "is either hypo- or hyper-thermic or is becoming so, and ultimately cannot perform its normal functions; therefore, channeling (sic) energy to other functions would not be possible" (Wunder 1984:167). Although homeothermy does not necessarily entail the strict maintenance of a constant body temperature (McArthur & Clark 1988) the fact that many homeotherms expend an extensive amount of energy on thermoregulation has been used to explain the selective pressure behind two of the oldest biogeographical relationships: Bergmann's and Allen's rules. These rules suggest that homeotherms from cold environments should be larger (Bergmann's rule) and have smaller appendages (Allen's rule) than those from warmer areas, in order to reduce the relative surface area from which heat is lost to the environment (Allaby 1985).

Recently, Searcy (1980) developed a model which has given additional support to Bergmann's rule because it predicts that optimum body size should increase with decreasing temperature. Searcy (1980) argued that a larger body size not only reduces the relative amount of energy spent on thermoregulation but also increases the amount available for other activities including that which can be stored for future use. As large animals are capable of storing more fat than smaller individuals (Morrison 1960) some studies have even suggested that selection might operate for large body size in seasonal environments where periods of food scarcity occur more regularly: what Boyce (1978:15) called the "seasonal hypothesis".

The aim of the present study was to investigate morphological variation in pouched mice from throughout the southern African subregion.

Materials and Methods

Morphological variation of pouched mice from the southern African subregion was studied among specimens housed at the Kaffrarian, McGregor and Transvaal museums in South Africa. Each individual was allocated to one of five age classes according to the toothwear criteria described by Ellison et al. (in press). To eliminate the effect of age and season on body size only specimens in age class 4 were used as these do not show significant seasonal differences in body mass (Ellison et al. in press.). Likewise, if the catalogue indicated that females were pregnant when caught they were excluded from the analysis in order to minimise any morphological variability caused by pregnancy. This left a total of 209 specimens (105 males, 101 females and 3 of unknown sex) from 104 verified localities comprising one in Zimbabwe, two in Mozambique, three in Botswana, 19 in Namibia, and 79 in South Africa (see Fig. 12). For each of these specimens three external body measurements (body, tail and ear length) and body mass were recorded from museum labels. However, because measurement techniques often vary between different collectors (Levenson 1990) the condylo-basal length of intact skulls was also recorded to provide a more reliable indication of body size (Delany 1970)

Climatic data (mean monthly rainfall, maximum and minimum temperatures) for each locality were generated using a climatic surface model developed for Africa by the Centre for Resource and Environmental Studies at the Australian National University in Canberra. The basic technique involves fitting mathematical surfaces to existing climate station records using Laplacian smoothing splines with generalised cross-validation (Hutchinson, Booth, McMahon & Nix 1984). This provided the best available estimates of climatic conditions particularly at localities far away from meteorological stations.

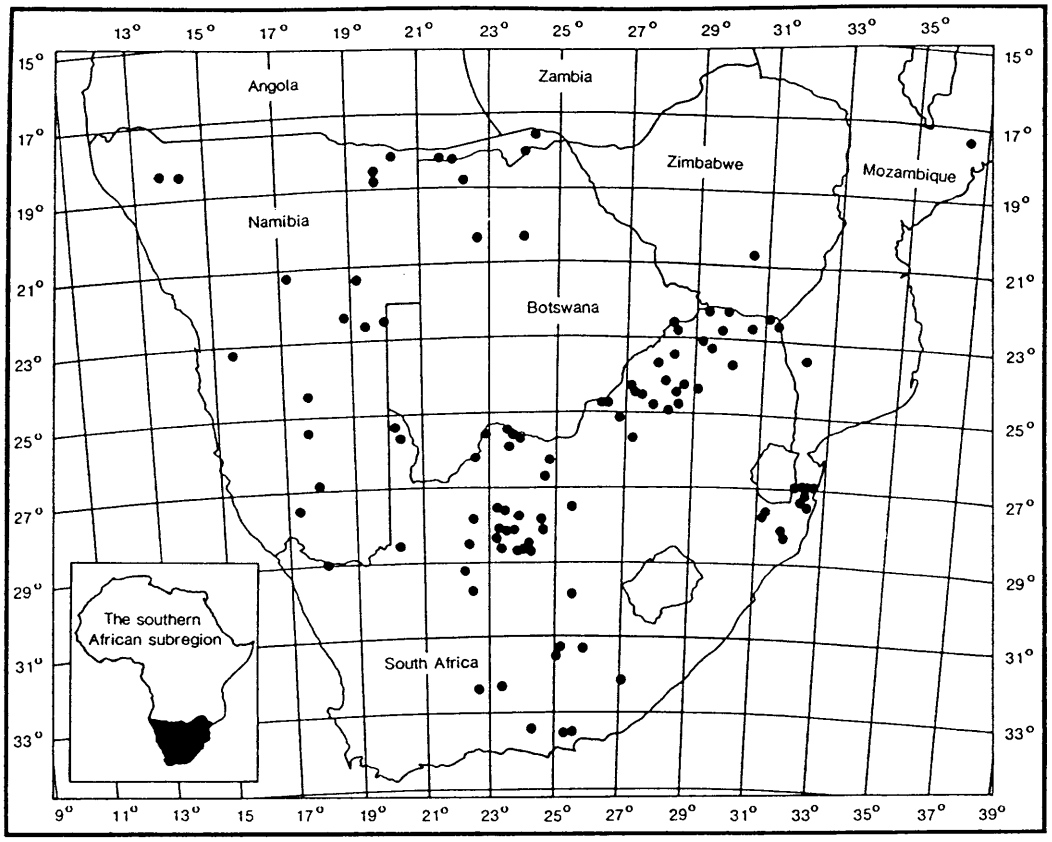


Figure 12 Morphological variation was examined among 209 pouched mice from 104 localities in the southern African subregion which are indicated by filled circles on this map of the subregion.

Although biologically significant climatic factors are difficult to define (Yom-Tov & Nix 1986), an attempt was made to restrict those used in the present study to a few relevant characteristics rather than include a large number of arbitrary measures. For this reason mean annual rainfall (MAR) was used because it provides an index of primary productivity and food availability (Boyce 1978). Likewise, mean monthly minimum temperature (T_{min}) was taken as the most relevant indication of the thermal environment experienced by pouched mice because T_{min} occurs at night when these animals are active outside their burrows (see Chapter 2). Finally, the coefficient of variation (Sokal & Rohlf 1981) of mean monthly rainfall and T_{min} was calculated in order to provide an estimate of seasonal variability of rainfall (S_r) and temperature (S_t) at each locality.

All results are presented as mean ± one standard deviation. The Mann-Witney U-test and Spearman's rank correlation were used for statistically analysing the results, and associations between morphological and climatic measurements were assessed using forward step-wise regressions (Sokal & Rohlf 1981).

Results

A preliminary analysis of morphological variation among males and females from a restricted area (the Transvaal Province of South Africa) indicated that *S.campestris* does not display significant sexual dimorphism, even though male pouched mice were generally larger and heavier than females (see Table 12). For this reason, data from both sexes were combined during the subsequent analyses and arithmetic means of the five morphological measures recorded for all the specimens examined are given in Table 13 together with the range of values recorded for each. There were highly significant correlations between condylo-basal length, body mass and the three external measurements (r_s>0,216, p<0,01, Table 14) which confirmed that skull size provided an accurate indication of all aspects of body size. There were also significant cross-correlations between most of the external measurements, including body mass, although tail length was not correlated with either body or ear length (r_s<0,116, p>0,05, Table 14).

Table 12 Sexual dimorphism of pouched mice from the Transvaal (South Africa)

	Character means				Mann-Witney	
Character	Male	(n)	Female	(n)	U	p
Condylo-basal length (mm)	32,3 ± 1,3	(16)	32,2 ± 1,6	(21)	165,5	>0,05
Head and body length (mm)	131 ± 15	(17)	123 ± 14	(24)	162,5	>0,05
Tail length (mm)	42 ± 5	(17)	39 ± 6	(24)	147,0	>0,05
Ear length (mm)	17 ± 1	(18)	17 ± 2	(24)	202,5	>0,05
Body mass (g)	63 ± 18	(13)	51 ± 16	(19)	86,0	>0,05

Table 13 Morphological measurements of pouched mice within the southern African subregion

Character	Mean ± S.D.	(n)	Range
Condylo-basal length (mm)	31,4 ± 1,7	(184)	26,4 - 35,7
Head and body length (mm)	120 ± 15	(200)	67 - 163
Tail length (mm)	42 ± 7	(200)	20 - 60
Ear length (mm)	17 ± 2	(201)	9 - 21
Body mass (g)	50 ± 14	(156)	15 - 100

Table 14 Cross-correlations between external morphological measurements of pouched mice within the southern African subregion

	Spearman's correlation coefficient (r_s)[a] Character		
Character	Tail length	Ear length	Body mass
Head and body length	$-0,020^{n.s.}$	$+0,302^{***}$	$+0,815^{***}$
Tail length	------	$+0,115^{n.s.}$	$+0,127^{*}$
Ear length	------	------	$+0,374^{***}$
Body mass	------	------	------

[a] Levels of significance: n.s.= p>0,05; *= p<0,05; ***= p<0,001.

All four climatic factors generated for localities examined in the present study displayed significant latitudinal variation: MAR ranged from 150 mm to 1396 mm and decreased with increasing latitude (r_s=-0,319, p<0,001) as did T_{min} (r_s=-0,646, p<0,001) which varied between 5,9 °C and 19,2 °C. Seasonal variability of rainfall (S_r= 16,9-120,9 %) also declined at higher latitudes (r_s=-0,579, p<0,001) while seasonality of T_{min} (S_t= 14,6-88,6 %) increased (r_s=0,519, p<0,001). As a consequence of the strong associations between latitude and climate there were also several cross-correlations between different climatic factors. For example, MAR was higher at localities which experienced warmer T_{min}s (see Table 15).

Table 15 Cross-correlations between climatic factors generated for 104 localities in the southern African subregion

	Spearman's correlation coefficient (r_s)[a] Climatic factor		
Climatic factor	T_{min}	S_r	S_t
Mean annual rainfall (MAR)	$+0,571^{***}$	$-0,375^{***}$	$-0,597^{***}$
Mean monthly minimum temperature (T_{min})	------	$-0,139^{n.s.}$	$-0,913^{***}$
Seasonal variation of rainfall (S_r)	------	------	$+0,071^{n.s.}$
Seasonal variation of temperature (S_t)	------	------	------

[a] Levels of significance: n.s.= p>0,05; ***= p<0,001.

Forward step-wise regressions between the four climatic factors, latitude and body size (condylo-basal length) or appendage size (tail and ear length) are given in Table 16. There was a significant correlation between climate and condylo-basal length which was largely caused by a strong positive correlation with MAR, although there were also significant associations with latitude and the remaining 3 climatic variables. Tail length was weakly correlated with seasonality of rainfall and latitude, while ear length was only correlated with a combination of seasonal variation in rainfall and temperature.

Table 16 Forward step-wise regressions between the four climatic factors, latitude and body size (condylo-basal length) or appendage size (tail and ear length) of pouched mice from the southern African subregion

	Multiple regression coefficient (r^2)[a] with 4 climatic factors and latitude[b,c]				
Measurement	Step 1	Step 2	Step 3	Step 4	Step 5
Condylo-basal length	+MAR 0,14***	+MAR+S_t 0,14***	+MAR-S_t+Lat 0,15***	+MAR-S_t-Lat+T_{min} 0,15***	+MAR-S_t+Lat-T_{min}+S_r 0,15***
Tail length	+S_r 0,03*	+S_r-Lat 0,03*	+S_r-Lat-T_{min} 0,03n.s.	+S_r-Lat-T_{min}-MAR 0,03n.s.	+S_r-Lat-T_{min}-MAR-S_t 0,03n.s.
Ear length	+S_r 0,01n.s.	+S_r+S_t 0,03*	+S_r-S_t+Lat 0,03n.s.	+S_r-S_t-Lat+MAR 0,03n.s.	+S_r-S_t-Lat+MAR+T_{min} 0,03n.s.

[a] Levels of significance: n.s.= $p>0,05$, *= $p<0,05$, ***= $p<0,001$.
[b] Abbreviations for climatic factors and latitude as follows: mean annual rainfall= MAR, mean monthly minimum temperature= T_{min}, seasonal variation of rainfall= S_r, seasonal variation of temperature= S_t, latitude= Lat.
[c] Positive and negative correlations are indicated by a plus (+) or minus (-) respectively.

Discussion

Both Searcy's (1980) model and Bergmann's rule assume that animals can actually reach an "optimal" body size and that sufficient food resources are available to support an increase in body size (because larger animals inevitably have higher total energy requirements despite their *relatively* lower thermoregulatory costs; McNab 1971). Indeed, Gould & Lewontin (1979) have criticised this type of approach because it ignores architectural constraints as well as the non-energetic ecological and social factors which influence body size. Certainly, smaller species are usually unable to achieve their "optimal" energetic size and therefore cannot escape the relatively high thermoregulatory costs associated with being small (Hart 1971). These costs would not necessarily disadvantage small mammals (assuming that it is not detrimental to forage and eat more often) as long as there is plenty of food available. However, when food availability declines these animals may be faced with a shortfall in their energy requirements which would be even greater at lower temperatures as a result of higher thermoregulatory costs. Even if these conditions only occurred for limited periods of time small mammals might not be able to survive as they are usually incapable of storing sufficient fat reserves for more than a few hours (Bronson 1987; Bronson, Heideman & Kerbeshian 1991).

Under these circumstances a reduction in body size might represent an alternative mechanism for coping with energetic deficits because it would help reduce the total energy requirements (McNab, 1971). Indeed, several small mammals, including *S.campestris* (Korn 1989; Ellison et al. in press), display a lower mean body mass in winter which causes a reduction in total energy requirements at a time when energy availability declines and thermoregulatory costs increase (Wunder et al. 1977; Heldmaier & Steinlechner 1981; Ellison and Skinner 1991b). In a similar way, the smaller body size of pouched mice in localities experiencing lower rainfall and colder temperatures might represent an adaptation to conserve energy in areas where food supplies are limited and thermoregulatory costs are high.

However, it is often impossible to determine the relative importance of temperature and rainfall on body size because geographical variation in climatic factors are usually closely related (Yom-Tov & Nix 1986; Nevo et al. 1986). Despite the strong correlation between rainfall and temperature in the present study it appears that rainfall was predominantly responsible for variation in the size of pouched mice: first there was a stronger correlation between MAR and body size than between body size and T_{min}, and secondly there were no clear correlations between T_{min} and the size of appendages (tail and ear length) which might be expected (according to Allen's rule) if temperature was influencing morphology. Likewise, Nevo et al. (1986) found that the higher body mass of mole rats (*Spalax ehrenbergi*) from cooler, more productive environments was primarily the result of geographical differences in rainfall and primary productivity, because the influence of food availability overrode that of temperature within a single species (2n=54) living in the Golan heights. Although, they also found a contradictory increase in the mass of animals living in colder, less productive environments these morphological differences may have been the result of phylogenetic factors as the two contrasting populations belonged to different chromosomal species (2n=52 & 2n=54; Nevo et al. 1986).

The present study and that of Nevo et al. (1986) were both conducted on species which spend most, if not all, of their lives underground. Although subterranean temperatures do vary geographically (e.g. Bennett et al. 1988) daily and seasonal temperature fluctuations are much lower within burrows and these may even be heated to some extent by their occupant(s) (see Chapter 2). It is therefore possible that these microhabitats are warm enough to eliminate the effect of geographical variation in temperature as a selection pressure for morphological adaptation (Hayward 1965c) which would explain why temperature was not an important determinant of body size in *S.campestris* or *S.ehrenbergi*. Whether this is generally true for fossorial or semi-fossorial mammals remains to be seen, because McNab (1979b) has argued that species from warmer areas should be small in order to limit the build up of excess heat within the confines of their burrows.

It is not clear whether the significant correlations between morphology and climate observed in the present study are the result of heritable adaptations or simply the response of a flexible phenotype to environmental induction. In this respect, several studies have demonstrated that a decline in ambient temperature (Ogle 1934), food (Dauncey & Ingram 1986), and even water availability (Buffenstein & Jarvis 1985a) can induce a reduction in body size similar to that observed among pouched mice from cooler, drier localities. Although selection operates on phenotype (which in turn is under genetic control) it does not necessarily follow that phenotypic responses enable us to understand patterns of adaptation simply because these are expected to be in the same direction as adaptive genetic variation (Boyce 1978). Indeed, phenotypic differences may not imply that selection is operating at all (Gould & Johnston 1972) while it is also possible that variation in body size is the result of other selection pressures, such as character displacement (McNab 1971; Alcantara 1991) or sexual effects (Levenson 1990), which may obscure or even reverse the effects of metabolic constraints (Searcy 1980).

Ultimately there is only one satisfactory way to differentiate between phenotypic and genotypic causes and that involves examining the size of individuals from different localities under uniform conditions (Gould & Johnston 1972). This question will be addressed in the next Chapter (Chapter 5).

CHAPTER 5: FUNDAMENTAL INTRASPECIFIC VARIATION IN THERMOREGULATION AND ENERGETIC PHYSIOLOGY OF POUCHED MICE

Introduction

Scholander, Hock, Walters & Irving (1950b) proposed three possible avenues for thermal adaptation in homeotherms, namely: (i) the body to air gradient (ie. body temperature), (ii) basal metabolism, and (iii) thermal insulation. However, they could find no evidence of adaptation in body temperature, while variations in basal metabolism were fully explained by differences in body size irrespective of climate. They therefore concluded that phylogenetic temperature adaptation "rests entirely upon the plasticity of the factors which determine the heat loss, mainly the fur insulation".

Hart (1956) has since indicated that there is limited potential for improved insulation in smaller mammals, while a series of studies have illustrated that heritable differentiation in energy metabolism can also be attributed to taxonomic differences, diet, behaviour, and local climatic factors (see Table 1). For this reason, investigations on related populations occurring in different habitats not only demonstrate how selective environmental pressures induce phenotypic modifications, but also how the adaptations acquired by a species allow it to live in diverse climatic zones (Bowers 1971).

The aim of the present study was to examine fundamental intraspecific variation in thermoregulation and metabolism of *S.campestris* from throughout southern Africa, to assess whether this species displays any geographical differences in energetic physiology which represent ecotypic adaptations to their local environment.

Materials and methods

Animals

Intraspecific variation in energy metabolism was investigated in pouched mice which originated from nine localities in southern Africa and displayed variable karyotypes (see Fig. 13). Annual rainfall and air temperature data for each locality were obtained from their nearest meteorological station and these are summarised in Table 17.

A total of 54 adult males and nonpregnant females, comprising six individuals from each locality, was studied. Approximately twice as many mice were derived from wild-caught (n=35) as opposed to first generation laboratory-bred (n=19) stocks. However, those from *Komatipoort* had been in captivity since 1959 at the Medical Research Council in Nelspruit and these were studied to determine whether genetic differences in energy metabolism were still evident after prolonged periods in captivity. A similar analysis was employed by McNab & Morrison (1963) who included a laboratory population of *Peromyscus* spp. when analysing intraspecific variation in metabolism among 10 populations from arid and mesic environments.

All laboratory-bred mice were reared under long photoperiod (14L:10D) at 25°C and were at least four months old at the start of the present study. In order to eliminate the effects of local acclimatization, wild-caught animals were exposed to similar conditions for at least four months before the present study began. The pouched mice were housed separately in standard laboratory rat cages and provided with rabbit pellets, rat cubes (Epol, Vereeniging) and water *ad libitum*, a diet occasionally supplemented with sunflower seeds, apple and carrots. Shredded paper and sawdust were provided for bedding and their cages were cleaned weekly, and dusted with insecticide to eradicate ectoparasites (Karbadust: Sentrachem, Silverton. Containing 50g/kg Carbamate). Relative humidity was maintained above 45% to prevent ringtail (Ellison & Westlin-Van Aarde 1990).

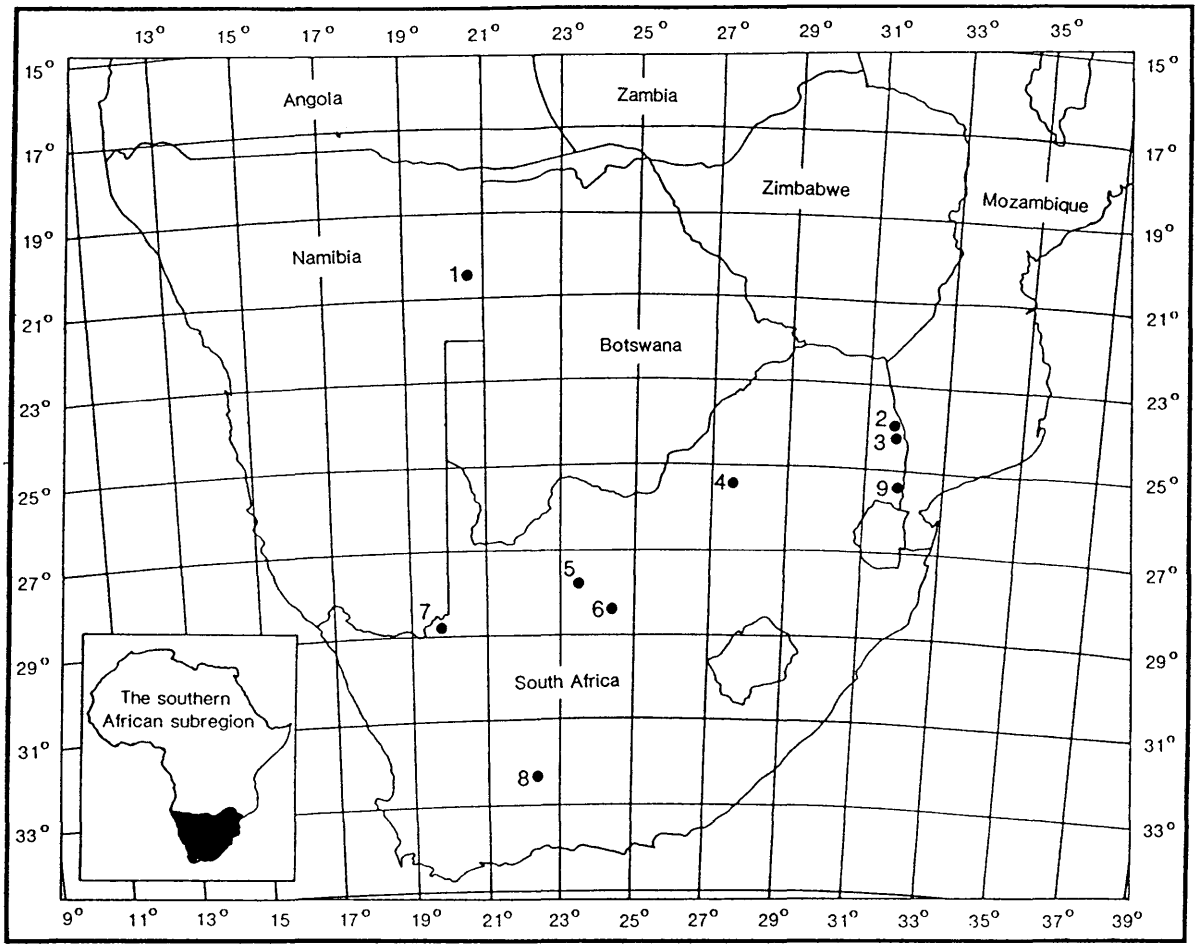


Figure 13 The localities and karyotypes of nine pouched mouse populations examined:

Locality	(Grid Reference)	Karyotype (2n)
1. *Gam*	(S.E.Bushmanland, Namibia: 20°27'S 20°44'E)	35
2. *Letaba*	(Kruger National Park, E.Transvaal, RSA: 23°56'S 31°35'E)	46
3. *Nwanedzi*	(Kruger National Park, E.Transvaal, RSA: 24°23'S 31°40'E)	46
4. *Vaalkop*	(Brits, Central Transvaal, RSA: 25°23'S 27°28'E)	44
5. *Ga-Tlhose*	(Sishen, N.Cape, RSA) 27°49'S 23°30'E)	30
6. *Pniel*	(Barkly West, Kimberley, N.E.Cape, RSA: 28°35'S 24°31'E)	32
7. *Schuinklip*	(N.E. Pofadder, N.Cape, RSA: 28°51'S 19°42'E)	35
8. *Doornhoek*	(Karoo National Park, S.Cape, RSA: 32°21'S 22°35'E)	46
9. *Komatipoort*[a]	(Komatipoort, E.Transvaal, RSA: 25°24'S 31°58'E)	46

[a]The population from Komatipoort had been maintained in captivity at the Medical Research Council in Nelspruit since 1959

Table 17 Climatic conditions at the nine pouched mouse localities studied in Chapter 5[a]

Locality (Meteorological Station)	Altitude (m)	Mean Annual Rainfall (mm)	Annual Mean Daily Air Temperatures Maximum (°C)	Minimum (°C)	Range (Max.-Min.) (°C)
1. *Gam* (Grootfontein 19°36'S 18°08'E)	1398	611	28,3	12,6	15,8
2. *Letaba* (Letaba 23°51'S 31°35'E)	215	387	30,4	15,8	14,7
3. *Nwanedzi* (Satara 24°24'S 31°47'E)	437	437	29,6	15,0	14,6
4. *Vaalkop* (Brits 25°35'S 27°49'E)	1158	621	26,6	10,4	16,2
5. *Ga-Tlhose* (Sishen 27°42'S 22°59'E)	1204	386	26,6	11,5	15,4
6. *Pniel* (Barkly West 28°32'S 24°32'E)	1204	391	26,6	9,7	16,9
7. *Schuinklip* (Pofadder 29°14'S 16°52'E)	989	105	25,8	11,1	14,7
8. *Doornhoek* (Beaufort West 32°18'S 22°35'E)	837	260	25,2	10,3	14,9
9. *Komatipoort* (Komatipoort 25°24'S 31°58'E)	189	599	28,9	15,7	13,3

[a]There were significant correlations between minimum and maximum air temperatures (SRC: r_s=0,848, n=9, p<0,01), and between minimum air temperature and temperature range (r_s=-0,695, n=9, p<0,05). However, there was no significant correlation between maximum air temperature and temperature range (r_s=-0,315, n=9, p>0,05) nor were there significant correlations between rainfall and maximum temperature (r_s=0,509), minimum temperature (r_s=0,267) or temperature range (r_s=-0,008, n=9, p>0,05).

Acclimation

Throughout the study the pouched mice were housed in a windowless climate chamber in which photoperiod and temperature could be accurately controlled. Within this chamber they were acclimated to a standard photoperiod (12L:12D) at 25°C for a further eight weeks before the following metabolic measurements were taken:

Oxygen consumption

Oxygen consumption ($VO_{2)}$ was measured at ambient temperatures (T_a) between 5°C and 34°C, using an open circuit system (as described by Depocas & Hart 1957; Hill 1972; Withers 1977, Equation 3a) and assuming an RQ of 0,85. Usually, the VO_2 of any one pouched mouse was only recorded at a single T_a per day, but occasionally, at T_as around the thermoneutral zone (24°C - 32°C), VO_2 was measured at two consecutive T_as daily. Measurements were made during daylight, the inactive phase of *S.campestris*, and whilst each animal was at rest. The pouched mice were placed in a clear perspex respiration chamber (volume 700ml) through which a flow of dried air (Silica gel: Holpro, Johannesburg) was passed at a rate of $600ml \cdot min^{-1}$. The respiration chamber was immersed in a constant temperature water bath (Labotec, Isando) and a chromel-alumel thermocouple (52K/J: Fluke, Everett) placed within the 'chamber was used for monitoring T_a. After a two-three hour stabilising period and following full equilibration, VO_2 was recorded using an Ametek S-3A/I (Applied Electrochemistry, Pittsburgh) oxygen analyser, and readings were taken every three minutes for half an hour. For Ta's between 15°C and 34°C, five consecutive readings which did not differ by more than 0,03% were used for calculating resting metabolic rate (RMR). At 5°C and 10°C VO_2 was variable, and the mean of ten consecutive three minute readings, recorded whilst each animal was at rest, was used for computing RMR. All VO_2 measurements were corrected to standard temperature and pressure, dry (STPD) and the oxygen analyser was calibrated before and after each measurement. Minimum observed metabolic rate (MOMR) was taken as an estimate of basal metabolism (BMR: Simon

1987) and compared to that predicted by Hayssen & Lacy (1985) for cricetid rodents with the same body mass. MOMR was subsequently expressed per (mass in grams)^{0,774}, as indicated by Hayssen & Lacy (1985), in order to compensate for any variation in body mass.

Body mass

Body mass (M_b) was measured after each VO_2 measurement using a PB3000 Mettler Scale (Mettler, Zurich). Animals were restrained within a cotton bag, and M_b was recorded to the nearest 0,1g.

Body temperature

Rectal temperature (T_b) was recorded following each VO_2 measurement using a chromel-alumel thermocouple connected to a 52K/J Fluke digital thermometer. The thermocouple was inserted approximately 30mm into the rectum for a period not exceeding 15 sec.

Minimal thermal conductance

Minimal thermal conductance (C_m) was calculated at ambient temperatures below thermoneutrality from the slope of the regression line relating VO_2 and T_a (McNab 1980b). According to Newton's Law of cooling, this slope is equivalent to C_m if the regression achieved intercepts the X-axis at a T_a corresponding to T_b (Scholander, Walters, Hock & Irving 1950a; McNab 1980b). Measured C_m was compared to that predicted according to Bradley & Deavers' (1980) equation relating C_m to M_b in cricetids. C_m was then expressed per (mass in grams)^{0,46}, as their equation suggests, in order to exclude the effect of body mass on C_m (Bradley & Deavers 1980).

Lower critical temperature

As suggested by Hayward (1965b), the intercept between MOMR and the regression of VO_2 on T_a was used to calculate the lower critical temperature, at the bottom of the thermoneutral zone, below which increases in metabolism are required to maintain T_b.

Non-shivering thermogenesis

Non-shivering thermogenesis (NST) was measured in unanaesthetised pouched mice at 30°C, as described by Ellison & Skinner (1990), using a dose of noradrenaline (NA: 1,5mg/Kg body mass, L-noradrenaline bitartrate: Sigma, St.Louis) above that required to elicit a maximal thermogenic response in mammals weighing more than 25g (Heldmaier 1971). The NA was dissolved in sterile saline (0,9% NaCl) and injected into the mice subcutaneously. The maximum VO_2 (VO_{2NA}) of inactive individuals, recorded over the next 45 min, was taken as an indication of the maximum capacity for NST. This was compared to that predicted using Heldmaier's (1971) allometric equation relating VO_{2NA} to M_b in cold-acclimated mammals. VO_{2NA} was subsequently expressed per (mass in grams)^{0,546}, as indicated by Heldmaier's (1971) equation, in order to eliminate the effect of any intraspecific variation in M_b.

Correlations between energy metabolism and climate

Geographical variation in thermoregulatory parameters and local climatic conditions was compared using forward step-wise regressions (FSR; Sokal & Rohlf 1981) with mean annual rainfall and air temperature data (minimum, maximum and range) recorded at the nearest meteorological station to each locality (see Table 17). M_b was also included in the regressions of MOMR (per $M_b^{0,774}$), C_m (per $M_b^{0,46}$), and VO_{2NA} (per $M_b^{0,546}$) to assess whether there were any residual effects of body mass on these parameters. Metabolic data recorded for the laboratory colony originating from *Komatipoort* were excluded from these calculations so that their data could be compared to those predicted by the regression equations. In this way

it was hoped to establish whether genetic differences in energy metabolism could be accurately determined by climatic factors even after several generations in captivity.

Statistical analyses

All results are presented as mean ± one standard deviation. In addition to the forward step-wise regressions, Spearman's rank correlation coefficient (SRC), and one-way analyses of variance (ANOVA) with multiple comparisons using the studentised range (Q) were used for the statistical analysis of results (Sokal & Rohlf 1981).

Results

Body mass

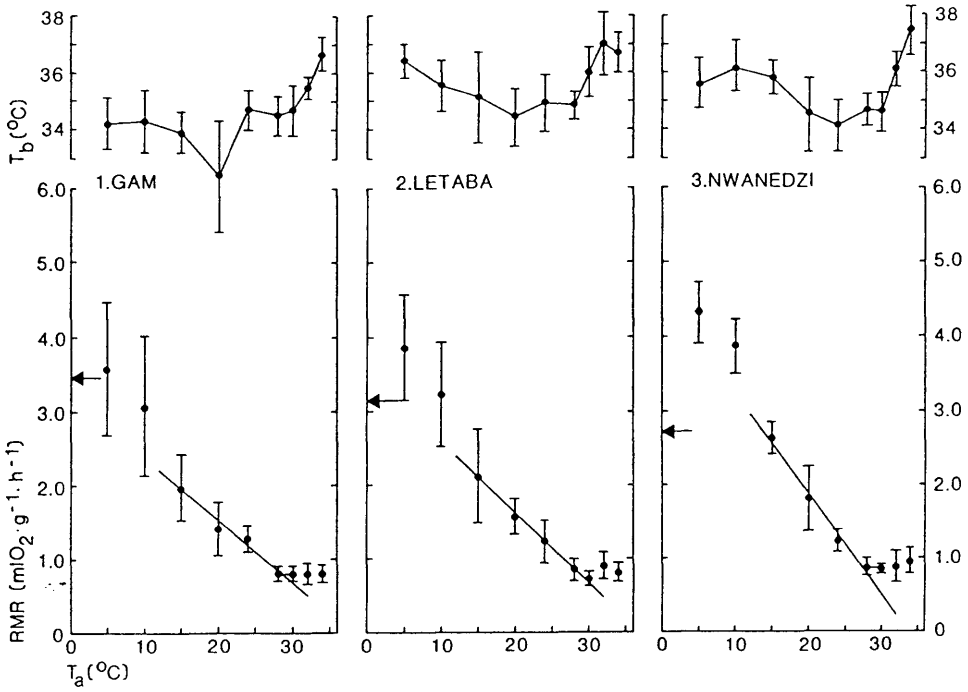
The mean M_b of adult pouched mice ranged from 40,9 ±5,7g (*Schuinklip*) to 92,3 ±13,0g (*Nwanedzi*) and there were significant geographical differences in M_b (ANOVA: F=10,09, d.f.= 8 & 45, p<0,001).

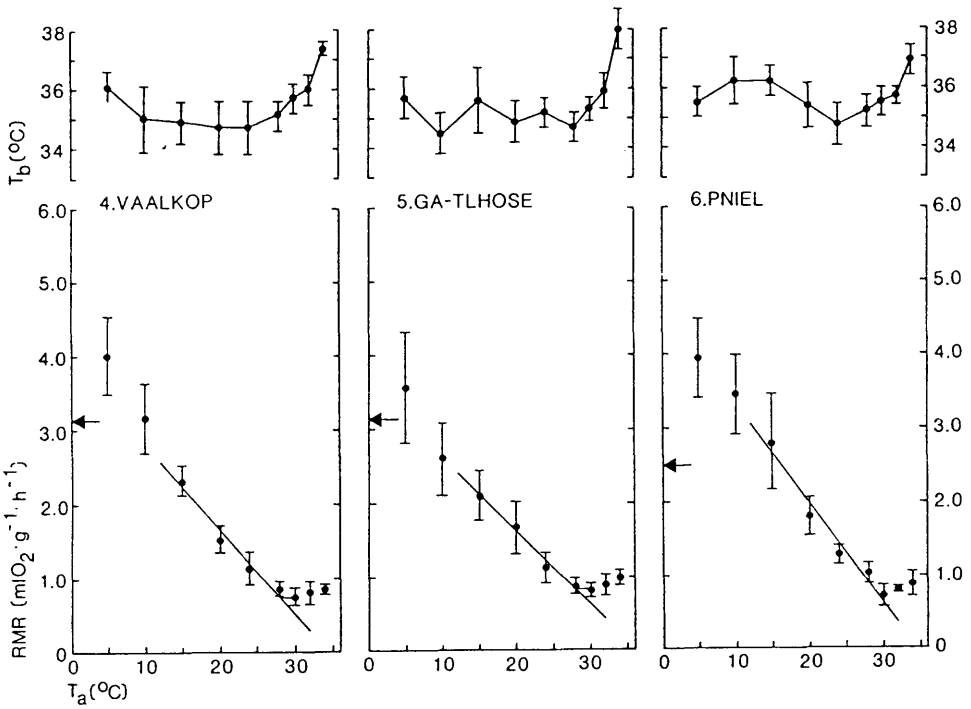
Resting metabolic rate

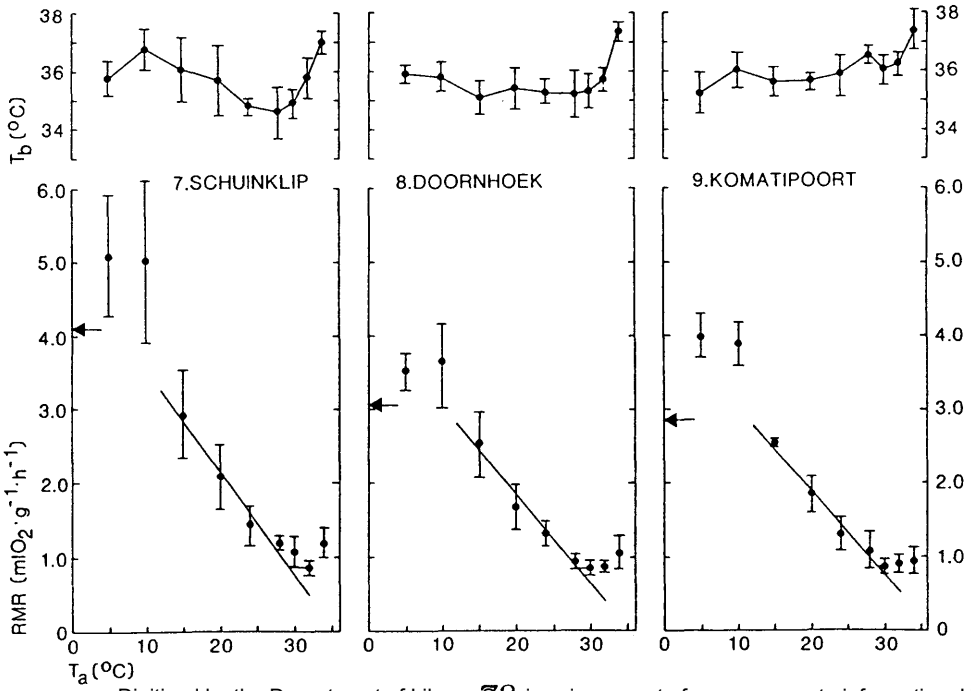
Fig. 14 displays the mean RMR and T_b of pouched mice recorded at T_as between 5°C and 34°C. All nine populations displayed narrow thermoneutral zones (TNZ) between 28°C and 32°C above and below which RMR increased with increasing and decreasing T_a. MOMR ranged between 58,3% and 72,2% of that predicted allometrically for cricetid rodents (see Fig. 15). When expressed per (mass in grams)^{0,774} to compensate for geographical differences in M_b there was still a significant effect of locality on MOMR (ANOVA: F=3,11, d.f.= 8 & 45, p<0,01) and individuals from *Nwanedzi* displayed the highest levels of MOMR (2,371 ±0,160 $mlO_2 \cdot g^{-0,774} \cdot h^{-1}$) which were significantly higher than the lowest levels exhibited by pouched mice from *Pniel* (1,879 ±0,355 $mlO_2 \cdot g^{-0,774} \cdot h^{-1}$: Q=4,88, k=9, v=45, p<0,05).

Figure 14 Overleaf: Resting metabolic rate (RMR) and body temperature (T_b) of pouched mice from nine localities recorded at ambient temperatures (T_a) between 5°C and 34°C. The arrows indicate the mean VO_{2NA} measured following an injection of noradrenaline. Vertical lines indicate one standard deviation of the mean.
Regression lines describing the relationship between RMR on T_a between 15°C and 28°C were fitted using the least-squares technique, and have the following equations:

Gam:-	$RMR = 3,207 - 0,085 \cdot T_a$ (r= -0,817)
Letaba:-	$RMR = 3,526 - 0,095 \cdot T_a$ (r= -0,801)
Nwanedzi:-	$RMR = 4,606 - 0,137 \cdot T_a$ (r= -0,932)
Vaalkop:-	$RMR = 3,952 - 0,114 \cdot T_a$ (r= -0,942)
Ga-Tlhose:-	$RMR = 3,601 - 0,099 \cdot T_a$ (r= -0,885)
Pniel:-	$RMR = 4,695 - 0,136 \cdot T_a$ (r= -0,879)
Schuinklip:-	$RMR = 4,966 - 0,140 \cdot T_a$ (r= -0,871)
Doornhoek:-	$RMR = 4,234 - 0,120 \cdot T_a$ (r= -0,902)
Komatipoort:-	$RMR = 4,132 - 0,112 \cdot T_a$ (r= -0,928)







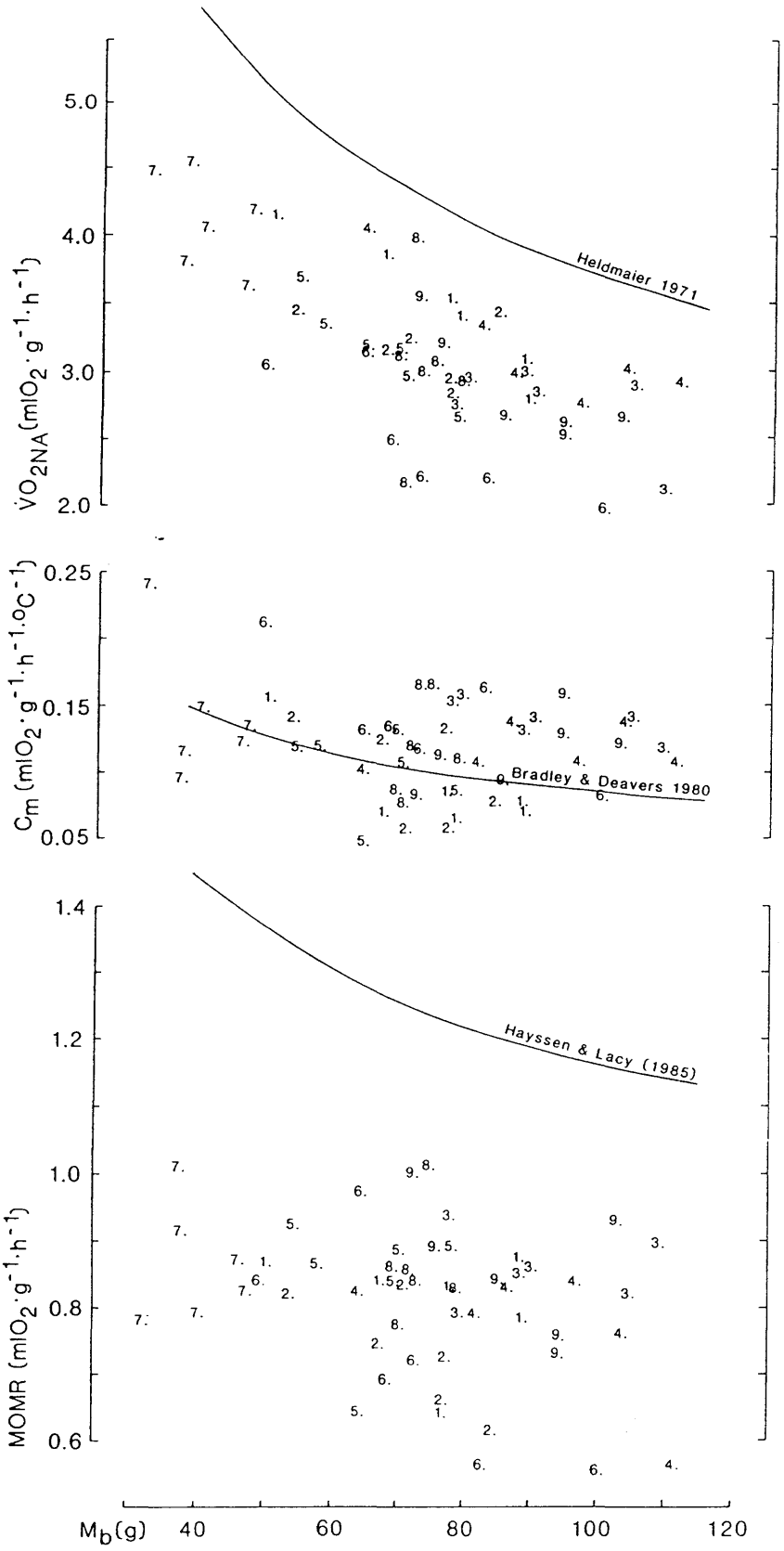


Figure 15 The relationships between body mass (M_b) and minimum observed metabolic rate (MOMR - bottom), minimum thermal conductance (C_m - middle), and noradrenaline-induced VO_2 (VO_{2NA} - top). Measurements recorded for pouched mice from each locality are indicated with the same locality numbers used in **Figure 13**. Curves describing predicted relationships from Hayssen & Lacy (1985: BMR/MOMR), Bradley & Deavers (1980: C_m), and Heldmaier (1971: VO_{2NA}) have also been drawn in each case.

Body temperature

Mean Tb was highly variable between T_a= 5°C and T_a= 34°C (Fig. 14) and although there was a uniform increase in T_b at T_as above 30°C there was a variable response to T_as below thermoneutrality. For example, pouched mice from *Vaalkop* displayed an increase in T_b as T_a declined while the T_b of animals from *Gam* fell at lower T_as (see Fig. 14). Clearly, *S.campestris* are extremely thermolabile which makes it difficult to determine a "normal" value of T_b for this species. Indeed, a comparison of T_b measured at T_a=30°C revealed that there was significant geographical variation in T_b (ANOVA: F=2,931, d.f.=8 & 45, p<0,05) ranging from a mean of 36,0°C (for pouched mice from *Letaba* and *Komatipoort*) to 34,7°C (for animals from *Gam*).

Minimal thermal conductance

Assuming that T_b at T_a=30°C represents a realistic estimate of normal T_b in *S.campestris* then the regression of VO_2 on T_a should intercept the X-axis between 34,7-36,0°C if the slope is a true reflection of minimal thermal conductance. However, for VO_2 between T_a= 5°C and 28°C the regression predicted a mean T_b of 33,5 ±1,0°C which was below the range of T_b measured at T_a= 30°C. This may have been the result of an increase in thermal conductance at lower temperatures possibly caused by the onset of shivering. Similar increases in thermal conductance at low temperatures have been reported for other rodent species (*Phodopus sungorus*: Heldmaier 1975, *Gerbillurus paeba*: Buffenstein 1984) while the VO_2 of pouched mice in the present study became variable at 5°C and 10°C and was disproportionately high at these T_as which might indicate that shivering was taking place (see Fig. 14). If these values are excluded, the regressions of VO_2 on T_a between 15°C and 28°C predict a mean T_b of 35,7 ±1,4°C which lies within the range observed. These regressions produced estimates of C_m which varied between 85,4% and 152,4% of that predicted allometrically for cricetids (Fig. 15). Even when expressed per (mass in grams)^{0,46}, there were still significant differences between the C_m of animals from different localities (ANOVA: F=3,64, d.f.= 8 & 45, p<0,01).

Lower critical temperature

In contrast, the lower critical temperatures (T_{lc}) calculated from the intercept between these regressions and MOMR were relatively constant and ranged between 27,5°C for pouched mice from *Nwanedzi* and 29,6°C for animals from *Pniel*, *Schuinklip* and *Komatipoort*. Indeed, there was no significant geographical variation in T_{lc} (ANOVA: F=0,72, d.f.= 8 & 45, p>0,05).

Nonshivering thermogenesis

The VO_{2NA} of pouched mice acclimated to 25°C was only 58,6% to 82,3% of that predicted for cold-acclimated mammals of similar body size (Fig. 15). However, there were fundamental geographical differences in NST capacity (ANOVA: F=5,50, d.f.= 8 & 45, p<0,001) which were still present after VO_{2NA} had been expressed per (mass in grams)^{0,546} to eliminate the effect of geographical variation in body mass.

Correlations between energy metabolism and climate

The metabolic measurements discussed above have been summarised in Table 18. There were no significant correlations between climatic factors and MOMR (F=2,18), C_m (F=2,10), T_b at T_a=30°C (F=2,25) or T_{lc} (F=0,87, d.f.= 5 & 47, p>0,05). However, there were significant associations between climatic conditions and $\dot{M}_b$ (FSR: F=10,31, d.f.= 4 & 43, p<0,001, see Table 19) and between these factors and VO_{2NA} (F=6,90, d.f.= 5 & 42, p<0,01, see Table 20). The combined correlation of r^2=0,489 between climate and $\dot{M}_b$ was largely the result of a strong association with rainfall (r^2=0,417) such that mean annual rainfall at *Komatipoort* successfully predicted the $\dot{M}_b$ of pouched mice from that locality (Table 19). Rainfall was also capable of predicting the VO_{2NA} of individuals from *Komatipoort* although the combined correlation of r^2=0,451 between VO_{2NA}, $\dot{M}_b$ and climatic conditions appeared to be more equally distributed between (four of) the five factors tested and together these did not provide an accurate estimate of VO_{2NA} (Table 20).

Table 18 A summary of metabolic measurements recorded for pouched mice from the nine localities examined in Chapter 5

Locality	M_b (g)	BMR ($ml O_2 \cdot g^{-0,774} \cdot h^{-1}$)	% BMR Predicted[a]	T_b at T_a = 30°C (°C)	T_{lc} (°C)	C_m ($ml O_2 \cdot g^{-0,46} \cdot h^{-1,o}C^{-1}$)	% C_m Predicted[b]	VO_{2NA} ($ml O_2 \cdot g^{-0,546} \cdot h^{-1}$)	% VO_{2NA} Predicted[c]	F-Ratio %BMR/%C_m[d]
1. *Gam*	75,8 ±14,4	2,122 ±0,233	64,8	34,7 ±0,9	28,5 ±2,0	0,847 ±0,228	85,4	24,334 ±1,677	82,3	0,76
2. *Letaba*	72,5 ±10,5	1,908 ±0,187	58,3	36,0 ±0,9	29,5 ±2,0	0,948 ±0,350	94,2	22,061 ±1,919	73,9	0,62
3. *Nwanedzi*	92,3 ±13,0	2,371 ±0,160	72,2	35,6 ±0,7	27,5 ±1,1	1,562 ±0,107	127,4	21,200 ±2,153	71,1	0,57
4. *Vaalkop*	91,5 ±16,8	2,106 ±0,255	64,4	35,7 ±0,5	28,1 ±1,6	1,311 ±0,258	152,4	24,263 ±1,744	82,0	0,42
5. *Ga-Tlhose*	66,5 ±08,9	2,156 ±0,265	65,6	35,3 ±0,4	29,0 ±4,4	0,945 ±0,278	92,7	21,062 ±1,178	70,7	0,71
6. *Pniel*	73,7 ±17,3	1,879 ±0,355	57,8	35,5 ±0,5	29,6 ±1,7	1,348 ±0,321	135,6	17,217 ±1,985	58,6	0,43
7. *Schuinklip*	40,9 ±05,7	1,995 ±0,219	60,8	34,9 ±0,5	29,6 ±1,2	1,022 ±0,313	100,8	22,092 ±1,655	74,0	0,60
8. *Doornhoek*	73,7 ±03,4	2,270 ±0,238	69,0	35,3 ±0,6	28,4 ±1,6	1,230 ±0,400	118,8	21,341 ±4,088	71,2	0,58
9. *Komatipoort*	88,1 ±11,9	2,337 ±0,258	71,3	36,0 ±0,5	29,6 ±2,4	1,273 ±0,368	123,2	21,587 ±1,894	72,5	0,58

Footnotes:

[a] % BMR predicted according to Hayssen and Lacy's (1985) equation relating BMR to M_b for Cricetid rodents.

[b] % C_m predicted according to Bradley and Deavers' (1980) equation relating C_m to M_b for Cricetid rodents.

[c] % VO_{2NA} predicted according to Heldmaier's (1971) equation relating VO_{2NA} to M_b for mammals.

[d] F-Ratio calculated from % BMR predicted / % C_m predicted as suggested by Buffenstein (1984).

Table 19 Forward step-wise regression of M_b on local climatic factors for pouched mice from localities 1 to 8 together with the values predicted for individuals originating from *Komatipoort* (Observed M_b= 88.1 ±11.9g)

Step	Rainfall	T_a Range	T_a Minimum	T_a Maximum	Intercept	r^2	F-Test	d.f.	*p*	Predicted M_b
	b	*b*	*b*	*b*	*a*		F			for *Komatipoort*
1	0,079	-	-	-	42,216	0,417	32,893	1 & 47	<0,01	89,5
2	0,089	-4,000	-	-	99,670	0,438	17,523	2 & 47	<0,01	99,8
3	0,093	-0,440	-5,082	-	120,223	0,439	11,464	3 & 47	<0,01	90,3
4	0,096	43,171	-43,367	-48,452	121,707	0,489	10,308	4 & 47	<0,01	-1327,7

Table 20 Forward step-wise regression of VO_{2NA} on local climatic factors and M_b for *S.campestris* from localities 1 to 8 and the values predicted for individuals originating from *Komatipoort* (Observed VO_{2NA}= 21.587 ±1.894ml$O_2 \cdot g^{-0,546} \cdot h^{-1}$)

Step	Rainfall	T_a Range	M_b	T_a Minimum	T_a Maximum	Intercept	r^2	F-Test	d.f.	*p*	Predicted VO_{2NA}
	b	*b*	*b*	*b*	*b*	*a*		F			for *Komatipoort*
1	0,005	-	-	-	-	19,618	0,077	3,847	1 & 47	>0,05	22,613
2	0,010	-1,685	-	-	-	43,823	0,231	6,767	2 & 47	<0,01	27,401
3	0,016	-1,958	-0,068	-	-	50,643	0,341	7,575	3 & 47	<0,01	28,195
4	0,022	-0,707	-3,709	-7,098	-	83,922	0,436	8,318	4 & 47	<0,01	-350,505
5	0,023	3,800	-4,490	-7,568	-0,079	85,010	0,451	6,903	5 & 47	<0,01	-367,343

Discussion

Reductions in energy metabolism represent an important adaptation to arid environments where food and water resources are limited (Louw & Seely 1982). For this reason, many arid-adapted rodents exhibit low resting metabolic rates (particularly BMR) which reduce their energy expenditure and, concomitantly, their food and water requirements (eg. McNab & Morrison 1963; Nevo & Shkolnik 1974; Bradley, Miller & Yousef 1974; McNab 1979b; Hill 1983; Buffenstein 1984; Haim, Skinner & Robinson 1987; Downs & Perrin 1990; Armitage, Melcher & Ward 1990). Similar reductions in MOMR were recorded in the present study among *S.campestris* from arid and semi-arid localities in southern Africa (see Table 17).

Likewise, adaptive increases in thermal conductance are common among fossorial mammals because a high C_m helps dissipate heat within the confines of a burrow (Ar 1987). In fact, elevated C_ms are also found among many semi-fossorial species which experience similar conditions to those living permanently underground (Maclean 1981). However, there was considerable variation in the C_m of semi-fossorial pouched mice examined in the present study, even though most populations displayed values of C_m which were close to or higher than those predicted by Bradley & Deavers (1980). Indeed, the low C_m exhibited by individuals from *Gam* was unusual and was probably the result of their significantly lower body temperature (see Table 18).

According to Buffenstein (1984) the "F-ratio" between the % predicted BMR (MOMR) and % predicted C_m represents an indication of the ability to maintain a constant body temperature where a ratio close to 1 is indicative of well-developed homeothermy. Therefore, the low F-ratios displayed by *S.campestris* (F=0,42 to 0,76, see Table 18) suggest that pouched mice cannot precisely maintain homeothermy and this is supported by the high degree of thermolability which was evident in the present study (Fig. 18). Indeed, it has been shown that pouched mice from throughout southern Africa abandon homeothermy and enter spontaneous torpor when subjected to cold-stress (Ellison & Skinner in press).

Pouched mice display extensive intraspecific variation in body mass, energy metabolism and thermoregulation (Table 18), and presumably these differences are innate for they occurred under controlled laboratory conditions. At the same time, the variable karyotype exhibited by *S.campestris* in the present study suggest that different populations may be genetically distinct. For this reason it is possible that the geographical differences in energy metabolism of pouched mice represent inherent subspecific variation similar to those reported for chromosomal species of *Spalax ehrenbergi* in Israel (Nevo & Shkolnik 1974; Haim et al. 1984; Nevo et al. 1986). However, in view of Ferreira's (1990) study (see Chapter 1 and Fig. 5) it is probable that the geographical differences in energy metabolism recorded in the present study simply reflect intraspecific physiological polymorphisms among different populations of *S.campestris*.

Body mass and NST capacity were significantly correlated with local climatic conditions, implying that geographical differences in these parameters might also represent ecotypic adaptation (Bowers 1971):

Body mass had a combined correlation of r^2=0,489 with four climatic factors although this was largely the result of a strong association with mean annual rainfall which produced a correlation of r^2=0,417 on its own (see Table 19). Mean annual rainfall represents an index of primary productivity and thus provides an indication of both food and water availability (Nevo et al. 1986). Reductions in the availability of either food or water can result in slower growth and smaller adult body size (Buffenstein & Jarvis 1985a; Dauncey & Ingram 1986). It is not surprising then that several studies have reported similar positive correlations between rainfall and body mass (eg. Bradley et al. 1974; Scheck 1982; Haim & Fairall 1986; Yom Tov & Nix 1986; Nevo et al. 1986; Armitage et al. 1990). The relationship between rainfall and Mb in the present study was still evident among captive bred individuals and those which had been subjected to prolonged laboratory acclimation with access to food and water *ad libitum*. Indeed, mean annual rainfall recorded at Komatipoort was capable of accurately predicting the M_b of a population from that locality which had been in captivity for several generations (see Table 19). This

suggests that body mass is heritable and might have been subjected to selection by rainfall prior to captivity. Smaller individuals are at an energetic advantage when food shortages occur because they require less food to maintain their smaller body size than do larger animals (Heldmaier & Steinlechner 1981; Wunder 1984, 1985; Ellison & Skinner 1991b). This would explain why pouched mice from localities with lower rainfall, where food supplies are limited, exhibit a lower body mass than those from areas with higher rainfall.

Unlike body mass, the combined correlation between NST capacity and local climatic conditions appeared to be more evenly distributed among the four climatic factors tested (see Table 20). In addition, there was a residual, negative correlation between NST and body mass despite transforming VO_{2NA} per (mass in grams)^{0,546} as indicated by Heldmaier (1971). The correlations with minimum and maximum temperatures were also negative, whilst those with temperature range and rainfall were positive. NST is an important mechanism for heat production in small mammals (Wunder 1984) and represents an essential component of cold tolerance in these species (Heldmaier et al. 1982). For this reason it is clearly advantageous for individuals from colder localities, experiencing larger temperature ranges, to exhibit a higher capacity for NST than those from warmer areas. Similar geographical differences in NST capacity have also been reported by Borut, Haim & Castel (1978) and Haim et al. (1984). Usually, cold exposure is required to elicit the full development of NST (Feist & Morrison 1981; Heldmaier et al. 1982) yet in the present study, and those of Borut et al. (1978) and Haim et al. (1984), fundamental geographical differences in NST were still apparent following acclimation to temperatures close to thermoneutrality. This indicates that even under thermoneutral conditions pouched mice from cooler localities retain a greater capacity for NST and this would enhance their survival if unpredictable periods of cold stress are more common in their natural environment. However, with the exception of rainfall, climatic measurements from Komatipoort did not successfully predict the VO_{2NA} of the colony from that locality (see Table 20) which implies that the capacity for NST might be modified by captive breeding. The positive

association between NST and rainfall suggests that individuals from wetter localities require higher levels of heat production even though there was a trend (not significant) for localities with higher rainfall to be warmer than drier areas (see Table 17). Alternatively, the relationship between NST and rainfall might indicate that individuals from wetter environments, with access to more abundant food supplies, can afford to maintain a higher capacity for NST than those from drier areas where food availability is low. In fact, no increase in energy expenditure is required to maintain a higher capacity for NST because this potential is only expressed when increases in heat production are required to maintain T_b at temperatures below thermoneutrality (Jansky 1973; Wunder 1985) and, as such, the association between NST and rainfall appears to be anomalous.

In view of the many studies relating geographical variation in BMR, C_m and T_{lc} to local climatic conditions (eg. Scholander et al. 1950b; McNab & Morrison 1963; Shkolnik & Borut 1969; Scheck 1982; Hill 1983; Bowers 1971) it seems surprising that similar correlations weren't found in the present study. However, it is possible that the climatic variables analysed reflect macroclimatic trends and only provided a rather rough estimate of the microclimatic conditions experienced by nocturnal, semi-fossorial pouched mice (Lynch 1973; Bradley et al. 1974). Alternatively, behavioural responses, such as the selection and modification of suitable microhabitats, might have limited the effect of local climatic extremes on the energy metabolism of *S.campestris* from southern Africa (see Chapter 2). Indeed, Hayward (1965c) found that burrow temperatures of *Peromyscus* spp. from different localities were very similar despite considerable differences in macroclimatic conditions and this explained why BMR was nonadaptive to climate in these species (Hayward 1965b). Under similar circumstances there may have been no selection pressure for ecotypic adaptation in BMR, C_m or T_{lc} of pouched mice examined in the present study.

Thus, although *S.campestris* from throughout southern Africa exhibit extensive geographical variation in metabolic parameters, only their body mass and their capacity for NST appear to exhibit significant ecological adaptation.

CHAPTER 6: SEASONAL ACCLIMATION OF THERMOREGULATION IN POUCHED MICE FROM CONTRASTING HABITATS

Introduction

The large surface area to volume ratio of small mammals makes them particularly susceptible to increases in heat loss during winter when ambient temperature declines (Heldmaier & Steinlechner 1981). In response, these animals display a diverse array of structural, physiological and behavioural adaptations which counteract the increase in heat loss and help maintain body temperature (Wunder 1984). Some of these responses, such as the modification of activity, require no preparation and are available all year round. Others, such as thicker pelage and increased thermogenesis, need time to develop, and Hart (1971) estimated that it takes at least two weeks for small mammals to develop maximal responses to cold. This time delay might not pose a critical threat to the survival of animals living in warmer areas or during warmer times of the year when cold spells are infrequent, brief and mild. However, temperatures can be unpredictable during winter in colder localities, and periods of prolonged cold stress might occur too quickly to permit the full expression of heat production and thermal insulation. Under these circumstances small mammals need to prepare before temperatures fall and many species use a decline in photoperiod to detect the onset of winter because it isn't subject to random fluctuations on the scale exhibited by other environmental factors (Steinlechner & Heldmaier 1982).

Recently, a series of studies on white footed mice (*Peromyscus leucopus*) established that populations from high and low latitudes differed in their ability to respond to changes in photoperiod (Lynch, Heath & Johnston 1981; Heath & Lynch 1982; 1983). These findings suggest that the extent to which a population uses photoperiod depends upon the relative severity of seasonal changes in temperature and/or food availability in each locality (Lynch et al. 1981). However, because the magnitude of seasonal changes in photoperiod also varies with latitude it remains unclear whether these geographical differences in photoresponsivity are the result

of differences in climatic or photoperiodic conditions.

The present study aims to evaluate the effect of short photoperiod and cold on metabolism and thermoregulation of *S.campestris* from three localities experiencing different climatic conditions but similar photoperiods. This experiment might establish whether there are any geographical differences in seasonal acclimation of energy metabolism which can be attributed to the climatic conditions prevailing in their native locality.

Materials and methods

Animals

Seasonal acclimation of thermoregulation was studied in pouched mice which originated from three localities in South Africa and displayed different karyotypes (2n); *Nwanedzi* in the Kruger National Park (24°23'S 31°40'E: 2n=46), *Schuinklip* Farm near Pofadder (28°51'S 19°42'E: 2n=35) and *Vaalkop* Dam Nature Reserve near Brits (25°23'S 27°28'E: 2n=44). Climatic data from each locality was obtained from their nearest meteorological station and have been summarised in Table 21.

Six adult males and non-pregnant females from each locality were used. Those from *Vaalkop* were all wild-caught, while animals from *Schuinklip* and *Nwanedzi* included both wild-caught and first generation laboratory-bred individuals. Laboratory-bred mice were reared under long photoperiod (14L:10D) at 25°C and were at least four months old at the start of the present study. Wild-caught animals were exposed to similar conditions for at least four months before the present study began in order to eliminate the effects of local acclimatization. The pouched mice were kept separately in standard laboratory rat cages and provided with rat cubes, rabbit pellets (Epol, Vereniging) and water *ad libitum*, a diet occasionally supplemented with sunflower seeds, apple and carrots. Shredded paper and sawdust were provided for bedding and their cages were cleaned weekly, and dusted with insecticide (Karbadust: Sentrachem, Silverton. Containing 50g/kg Carbamate) to eradicate ectoparasites. Relative humidity was maintained above 45% to prevent ringtail (Ellison & Westlin-van Aarde 1990).

Table 21 Climatic conditions at pouched mouse localities examined in Chapter 6

Locality:	*Nwanedzi*	*Schuinklip* Farm	*Vaalkop* Dam
Weather Station:	Satara (24°24'S 31°47'E)	Pofadder (29°14'S 16°52'E)	Brits (25°35'S 27°49'E)
Altitude (m):	275m	989m	1158m
Mean Annual Rainfall(mm):	437mm	105mm	621mm
		Air Temperatures (Summer: November - February)	
Mean daily maximum (°C)	32,3	31,8	29,8
Mean daily minimum (°C)	19,8	15,7	16,2
Mean daily range (°C)	12,5	16,0	13,5
Mean of lowest monthly (°C)	12,6	8,6	6,6
		Air temperatures (Winter: May - August)	
Mean daily maximum (°C)	26,2	18,9	22,3
Mean daily minimum (°C)	9,2	6,0	3,0
Mean daily range (°C)	17,0	12,9	19,3
Mean of lowest monthly (°C)	1,2	0,2	-7,9

Acclimation

Throughout the study the pouched mice were housed in a windowless climate chamber in which photoperiod and temperature could be accurately controlled. Within this chamber they were subjected to a twelve week protocol comprising four weeks acclimation under long photoperiod (14L:10D) at 25 $\pm$1°C followed by 8 weeks under short photoperiod (10L:14D) in which the first four weeks were spent at 25 $\pm$1°C and the last four weeks at 10 $\pm$1°C. During the fourth week of acclimation under each photoperiodic/temperature regime the following metabolic parameters were recorded:

Oxygen consumption

Oxygen consumption ($VO_{2)}$ was measured at ambient temperatures (T_a) between 5,0°C and 34°C, as described in Chapter 5. Minimum observed metabolic rate (MOMR) was again taken as an estimate of basal metabolism (BMR: Simon 1987) and expressed per (mass in grams)^{0,774} to compensate for any variation in body mass (M_b), as indicated by Hayssen & Lacy's (1985) equation relating BMR to M_b in cricetids.

Body mass

Body mass (M_b) was measured after each VO_2 measurement using a PB3000 Mettler Scale (Mettler, Zurich). Animals were restrained within a cotton bag, and M_b was recorded to the nearest 0,1g.

Body temperature

Rectal temperature (T_b) was recorded following each VO_2 measurement using a chromel-alumel thermocouple connected to a 52K/J Fluke digital thermometer. The thermocouple was inserted approximately 30 mm into the rectum for a period not exceeding 15 sec.

Minimal thermal conductance

Minimal thermal conductance (C_m) was calculated at ambient temperatures below thermoneutrality from the slope of the regression line relating VO_2 and T_a, (as described in Chapter 6). Likewise, measured C_m was again expressed per (mass in grams)^{0,46} in order to exclude the effect of differences in body mass on C_m as suggested by Bradley & Deavers' (1980) equation relating C_m to M_b in cricetids.

Non-shivering thermogenesis

Non-shivering thermogenesis (NST) was measured in unanaesthetised pouched mice at T_as within the thermoneutral zone, as described in Chapter 6. Maximum VO_2 (VO_{2NA}), recorded following an injection of noradrenaline (NA), was taken as NST maximum. Regulatory NST (that exceeding basal metabolism; Jansky 1973) was then calculated as the difference between VO_{2NA} and MOMR. Both these measures of NST (VO_{2NA} and regulatory NST) were expressed per (mass in grams)^{0,546} in order to eliminate the effect of any intraspecific variation in M_b, as indicated by Heldmaier's (1971) equation relating VO_{2NA} to M_b in mammals.

Statistical analyses

All results are presented as mean $\pm$ one standard deviation. Mann-Witney U-tests (MWU), and two-way analyses of variance (ANOVA) were used for the statistical analysis of results (Sokal & Rohlf 1981).

Results

Body mass

The M_bs of pouched mice from all three localities under each acclimatory regime are summarised in Table 22. Animals from *Schuinklip*, which had a mean M_b ranging from 48,4 $\pm$7,1g to 49,2 $\pm$8,6g, were significantly lighter than those from *Nwanedzi* or *Vaalkop* with a M_b between 82,4 $\pm$13,6g and 96,3 $\pm$20,8g (ANOVA, $F>33$, d.f.= 2 & 30, $p<0,001$). Reducing photoperiod or temperature had no significant effect on M_b ($F<3,5$, d.f.= 1 & 30, $p>0,05$).

Metabolic rate

Fig. 16 displays the mean RMR and T_b of pouched mice from each locality recorded at T_as between 5°C and 34°C. All three populations displayed thermoneutral zones (TNZs) between T_a= 28-32°C below which VO_2 increased linearly with declining T_a. MOMR (see Table 22) was unaffected by the decline in photoperiod (ANOVA, F=3,758, d.f.= 1 & 30, p>0,05) while it increased significantly following four weeks at T_a= 10°C (F=13,631, d.f.= 1 & 30, p<0,01) when cold acclimated pouched mice from *Nwanedzi* exhibited a higher MOMR than animals from *Vaalkop* or *Schuinklip* (F=3,714, d.f.= 2 & 30, p<0,05).

Body temperature

In contrast to VO_2, mean T_b was highly variable between T_a=5°C and 34°C and although there was a uniform increase in T_b at T_as above 30°C there was a variable response to T_as below thermoneutrality such that some populations displayed an increase in T_b as T_a declined, while the T_b of others fell at lower T_as (see Fig. 16). Even at T_a=30°C, where T_b was relatively constant, this ranged between 34,0°C and 36,6°C (Fig. 16). Clearly, *S.campestris* are highly thermolabile which makes it difficult to determine a "normal" value of T_b for this species.

Minimal thermal conductance

Assuming that T_b at T_a=30°C represents a realistic estimate of "normal" T_b in *S.campestris* then the regression of VO_2 on T_a below the TNZ should intercept the X-axis between 34,0-36,6°C. However, for VO_2 between T_a= 5°C and 28°C the regression predicted a mean T_b of 33,1 ±0,7°C which was below the range in T_b measured at T_a= 30°C. This may have been the result of an increase in thermal conductance at lower temperatures as observed in Chapter 5. If values of VO_2 recorded at T_a= 5°C and 10°C are excluded once more, then the regressions of VO_2 on T_a between 15°C and 28°C predict a mean T_b of 36,2 ±1,4°C which lies within the range observed.

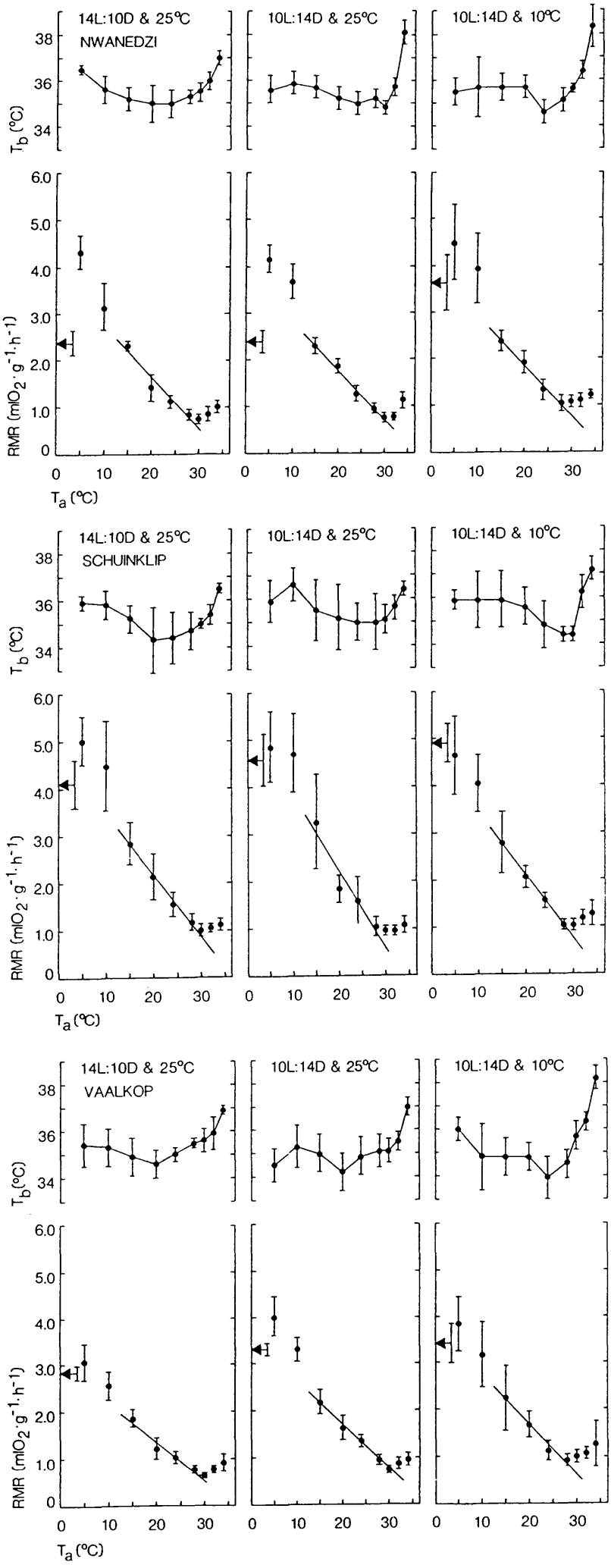
Table 22 A summary of metabolic measurements recorded for pouched mice from the three localities following acclimation to the different photoperiodic and temperature regimes

Experimental Condition	Metabolic measurement	Locality *Nwanedzi*	*Schuinklip*	*Vaalkop*
14L:10D & 25°C	M_b (g)	88,8 ± 16,8	48,9 ± 08,0	84,7 ± 12,9
	T_b at T_a= 30°C (°C)	35,5 ± 0,4	35,0 ± 0,2	35,6 ± 0,5
	MOMR ($mlO_2 \cdot M_b^{-0,774} \cdot h^{-1}$)	2,078 ± 0,321	2,179 ± 0,146	1,941 ± 0,072
	C_m ($mlO_2 \cdot M_b^{-0,46} \cdot h^{-1} \cdot °C^{-1}$)	1,229 ± 0,263	1,068 ± 0,234	0,894 ± 0,119
	VO_{2NA} ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	17,923 ± 0,978	23,817 ± 2,106	21,165 ± 1,199
	Reg. NST ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	9,960 ± 4,704	18,543 ± 2,065	15,831 ± 0,971
10L:14D & 25°C	M_b (g)	93,3 ± 11,1	49,2 ± 08,6	96,3 ± 20,8
	T_b at T_a= 30°C (°C)	34,8 ± 0,3	35,1 ± 0,6	35,1 ± 0,5
	MOMR ($mlO_2 \cdot M_b^{-0,774} \cdot h^{-1}$)	2,318 ± 0,202	2,172 ± 0,149	2,086 ± 0,191
	C_m ($mlO_2 \cdot M_b^{-0,46} \cdot h^{-1} \cdot °C^{-1}$)	1,222 ± 0,205	1,298 ± 0,404	1,075 ± 0,232
	VO_{2NA} ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	18,582 ± 1,538	26,682 ± 1,300	23,829 ± 1,878
	Reg. NST ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	12,075 ± 1,343	21,424 ± 1,386	17,932 ± 1,545
10L:14D & 10°C	M_b (g)	82,4 ± 13,6	48,4 ± 07,1	83,4 ± 16,3
	T_b at T_a= 30°C (°C)	35,6 ± 0,2	34,4 ± 0,3	35,7 ± 0,6
	MOMR ($mlO_2 \cdot M_b^{-0,774} \cdot h^{-1}$)	2,715 ± 0,190	2,327 ± 0,395	2,491 ± 0,336
	C_m ($mlO_2 \cdot M_b^{-0,46} \cdot h^{-1} \cdot °C^{-1}$)	1,158 ± 0,278	1,073 ± 0,291	1,109 ± 0,457
	VO_{2NA} ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	26,539 ± 2,580	28,398 ± 1,927	25,374 ± 1,677
	Reg. NST ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$)	19,120 ± 2,839	22,772 ± 2,256	18,562 ± 1,613

Figure 16 Overleaf: Mean body temperature (T_b - above) and resting metabolic rate (RMR - below) of 6 pouched mice from each locality measured at ambient temperatures (T_a) between 5-34°C under each acclimatory regime. The arrows indicate the mean VO_{2NA} following an injection of noradrenaline. Vertical lines indicate one standard deviation of the mean.

Regression lines describing the relationship between VO_2 on T_a's between T_a = 15°C and 28°C were fitted using the least-squares technique, and have the following equations:

Nwanedzi	14L:10D & 25°C:- $VO_2 = 3,811 - 0,109 \cdot T_a$	(r = -0,932)
Nwanedzi	10L:14D & 25°C:- $VO_2 = 3,895 - 0,106 \cdot T_a$	(r = -0,966)
Nwanedzi	10L:14D & 10°C:- $VO_2 = 3,988 - 0,107 \cdot T_a$	(r = -0,931)
Schuinklip	14L:10D & 25°C:- $VO_2 = 4,801 - 0,132 \cdot T_a$	(r = -0,887)
Schuinklip	10L:14D & 25°C:- $VO_2 = 5,482 - 0,163 \cdot T_a$	(r = -0,804)
Schuinklip	10L:14D & 10°C:- $VO_2 = 4,746 - 0,134 \cdot T_a$	(r = -0,886)
Vaalkop	14L:10D & 25°C:- $VO_2 = 2,959 - 0,080 \cdot T_a$	(r = -0,921)
Vaalkop	10L:14D & 25°C:- $VO_2 = 3,556 - 0,093 \cdot T_a$	(r = -0,923)
Vaalkop	10L:14D & 10°C:- $VO_2 = 3,783 - 0,105 \cdot T_a$	(r = -0,817)



When expressed per (mass in grams)^{0,46} (Table 22) there was no significant difference in C_m between the three populations (ANOVA, F<3,003, d.f.= 2 & 30, p>0,05), and C_m was not significantly affected by declines in either photoperiod (F=2,445, d.f.= 1 & 30, p>0,05) or T_a (F=0,614, d.f.= 1 & 30, p>0,05).

Non-shivering thermogenesis.

There was a significant increase in both VO_{2NA} (ANOVA, F=15,948, d.f.= 1 & 30, p<0,01) and regulatory NST (F=12.506, d.f.= 1 & 30, p<0,01) following the decline in photoperiod from 14L:10D to 10L:14D (Table 22). There was also a significant geographical effect on both these parameters (F>62,088, d.f.= 2 & 30, p<0,001) such that pouched mice from *Schuinklip* exhibited the highest levels of NST and those from *Nwanedzi* the lowest (Table 22). Both VO_{2NA} and regulatory NST also increased when T_a fell from 25°C to 10°C, although there was an interactive effect of temperature and locality (F>10,173, d.f.= 2 & 30, p<0,01) because animals from *Nwanedzi* displayed a much greater increase in NST capacity than those from either *Schuinklip* or *Vaalkop* (Table 22). In order to compare the increase in regulatory NST attributable to declines in photoperiod with those caused by falling T_a, these data have been summarised in Table 23. Pouched mice from *Nwanedzi* exhibited a significantly larger increase in regulatory NST when T_a fell from 25°C to 10°C than when photoperiod fell from 14L:10D to 10L:14D (MWU, U=3, $n_1=n_2=6$, p<0,02). In contrast, there were no significant differences between the increase in regulatory NST exhibited following changes in T_a and photoperiod by animals from either *Schuinklip* (U=10, $n_1=n_2=6$, p>0,05) or *Vaalkop* (U=13, $n_1=n_2=6$, p>0,05). These data suggest that, with respect to regulatory NST, pouched mice from *Nwanedzi* responded predominantly to T_a while those from *Schuinklip* and *Vaalkop* cued to both photoperiod and T_a.

Table 23 The proportion of the overall increase in regulatory NST attributable to the decline in photoperiod and ambient temperature

Locality:	*Nwanedzi*	*Schuinklip*	*Vaalkop*
Total increase in regulatory NST from 14L:10D & 25°C to 10L:14D & 10°C ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$):	7,590 ± 2,873	4,229 ± 2,154	2,731 ± 1,872
(% of total increase):	100%	100%	100%
Increase in regulatory NST from 14L:10D & 25°C to 10L:14D & 25°C ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$) :	0,544 ± 2,681	2,881 ± 2,341	2,101 ± 1,601
(%of total increase):	7,2%	68,1%	76,9%
Increase in regulatory NST from 10L:14D & 25°C to 10L:14D & 10°C ($mlO_2 \cdot M_b^{-0,546} \cdot h^{-1}$) :	7,046 ± 3,590	1,348 ± 1,894	0,630 ± 2,057
(%of total increase):	92,8%	31,9%	23,1%

Discussion

When confronted by a winter decline in ambient temperature small mammals must either avoid or resist the increase in heat loss if they are to maintain homeothermy (Wunder 1984). Mechanisms for avoiding heat loss can involve migrating to warmer climes, spending more time in warmer micro-habitats or constructing artificial nests in which to shelter from the cold. Alternative mechanisms for avoiding excessive heat loss include reducing thermal conductance or lowering body temperature and employing controlled hypothermia. In the absence of these thermoregulatory mechanisms, animals must resist increases in heat loss and this can be achieved by producing more heat through muscle contraction (shivering or exercise) or metabolic activity (BMR or regulatory NST) (Wunder 1984; see also Table 2). In the present study, pouched mice responded to

four weeks of cold acclimation by increasing metabolic heat production through higher levels of MOMR (≡ BMR) and regulatory NST. However, there was no significant change in thermal conductance and it appears that, within this timescale, *S.campestris* reacts to cold by resisting rather than avoiding increases in heat loss.

Higher levels of BMR often occur in winter as a result of an overall increase in metabolic activity and this can provide an important contribution to the higher levels of thermogenesis required at this time of year (Heldmaier & Steinlechner 1981). In addition, BMR interacts with thermal conductance to determine the lower critical temperature (T_{lc}), at the bottom of the TNZ, below which metabolism must increase if T_b is to be maintained (Scholander et al. 1950a). In the present study thermal conductance remained constant and the higher levels of MOMR produced an apparent decline in T_{lc} to around 26°C following cold acclimation (see Fig.16). This is advantageous because it would allow the pouched mice to maintain homeothermy at lower T_as without resorting to thermoregulatory heat production.

Despite these advantages, increasing BMR can be an inefficient mechanism for increasing thermogenesis because metabolism cannot be reduced below this fundamental threshold when there is no immediate heat drain to resist. Indeed, Jansky (1973) described BMR as "obligatory" NST, "which serves to maintain the basic energy demands of the homoiothermic (sic) organism". For this reason many small mammals show no seasonal changes in BMR (Pohl 1965; Klaus, Heldmaier & Ricquier 1988; Feist & Feist 1986) and use other avenues of heat production such as shivering or regulatory NST. Even among those species which display a winter increase in BMR many do so only when exposed to cold (eg. *Microtus ochrogaster*, Wunder 1984; and *S.campestris* in the present study), and do not respond to anticipatory cues such as photoperiod (Gettinger & Ralph 1985; Bartness & Wade 1984). In this way these species avoid an increase in obligatory heat production until it is actually required for thermoregulation. However, there are exceptions to this generalisation such as *P.sungorus* (Heldmaier & Steinlechner 1981), *Apodemus mystacinus* (Haim & Yahav 1982) and two diurnal murids from subtropical Africa,

Rhabdomys pumilio and *Lemniscomys (griselda) rosalia* (Haim 1982) which display an increase in BMR in response to a decline in photoperiod. As regards *P.sungorus*, these originate from localities such as Siberia which experience extreme cold during winter and their survival might thus require anticipatory improvements in every available avenue of heat production including BMR. As for *A.mystacinus* and the two diurnal species, it is unlikely that these require an increase in BMR for thermoregulatory purposes in view of the mild winter temperatures prevailing in their natural environments. Instead, the increased BMR of these species may be the result of a general increase in metabolic activity rather than a specific adaptation for improving heat production.

In contrast to BMR, regulatory NST is an essential component of cold tolerance in small mammals (Heldmaier et al. 1982) and, unlike BMR, is only expressed when necessary to maintain homeothermy in the cold (Jansky 1973). For this reason, improving the capacity for regulatory NST is less energetically expensive than increasing BMR and represents an economical alternative for anticipating an increase in cold stress (Wunder 1985). Thus many species anticipate cold and respond to a decline in photoperiod by increasing their potential for heat production via NST (Lynch, White, Grundel & Berger 1978; Haim & Fourie 1980; Haim 1982; Haim & Yahav 1982; Bartness & Wade 1984; Wunder 1984; Rafael & Vsiansky 1985). However, because the relative severity of seasonal changes in temperature varies geographically we might expect geographical variation in the use of photoperiod as an anticipatory cue. A series of studies on thermoregulation and reproduction in *P.leucopus* by Lynch and his co-workers have shown that "the extent to which a local population uses this cue (photoperiod) depends largely upon the population's necessity for accurately predicting its immediate environment" (Lynch et al. 1981; Heath & Lynch 1982, 1983). For this reason the geographical differences in responsivity to photoperiod discovered in the present study possibly reflect differences in the relative severity of winters in each locality. Certainly, pouched mice from *Nwanedzi*, which displayed the smallest increase in regulatory NST (Table 23) following the decline in photoperiod, came from an area

experiencing a warmer winter than animals from *Vaalkop* and *Schuinklip* (Table 21). Nevertheless, the animals from *Nwanedzi* generally had the highest levels of MOMR which might have helped compensate for their lower capacity for regulatory NST (see Table 22).

Although the cold-induced improvements in thermogenesis would help *S.campestris* counteract the increase in heat loss they would also result in higher energy requirements during winter, at a time when primary production is lowest and cold night-time temperatures restrict foraging. One mechanism for accommodating these increases in energy expenditure involves a reduction in body size, because smaller individuals require less food in spite of their higher mass-specific metabolic rate (Heldmaier & Steinlechner 1981). Indeed, many populations of small mammals, including *S.campestris* (Korn 1989), display a winter decline in mean body weight which would result in lower total energy requirements (Fuller, Stebbins & Dyke 1969; Iverson & Turner 1974; Klaus et al. 1988). These cycles in body weight could be caused by a reduction in individual body size and/or a shift towards a younger, smaller population during winter. However, Ellison et al. (in press) found that there was no significant difference in the relative age structure of *S.campestris* collected during wet and dry seasons (summer and winter) in the Transvaal (South Africa). At the same time, juvenile pouched mice stay small during their first winter in captivity and accrue significant energy savings without compromising their thermoregulatory ability (Ellison & Skinner 1991b). Petterborg (1978) obtained similar results for growing *Microtus montanus* and Yellon & Goldman (1984) found that young *P.sungorus* were significantly smaller if reared under short photoperiod. In the present study, however, there was no significant effect of photoperiod or temperature on the body mass of adult pouched mice, whilst adult specimens collected in autumn by Haim et al. (1991) actually put on weight in captivity. These results suggest that the annual cycle in mean body weight of *S.campestris* populations is caused solely by a reduction in the weight of young animals. This is supported by Ellison et al. (in press) who demonstrated that pouched mice from the youngest age classes were significantly smaller in winter

than in summer while older individuals maintained a relatively constant body weight throughout the year.

Despite the absence of a significant effect of acclimation on M_b, the relative energetic advantages of a lower M_b may still have been important for animals from *Schuinklip* which were significantly smaller than those from *Vaalkop* or *Nwanedzi* (Table 22). For example, while pouched mice from *Schuinklip* exhibited the highest levels of VO_{2NA} at T_a= 10°C the total energy they required for NST (VO_{2NA} expressed in $mlO_2 \cdot animal^{-1} \cdot h^{-1}$) was only 235.8 ±26.7 $mlO_2 \cdot animal^{-1} \cdot h^{-1}$ which was significantly lower than that for individuals from *Nwanedzi* (293.1 ±28.0, U= 2, $p<0.02$) or *Vaalkop* (282.6 ±29.5, U=4, $p<0.05$). *Schuinklip* animals inhabit by far the driest of the three localities and probably experience the least predictable food supply (Louw & Seely 1982). With limited energy available, this population would be under intense selection to reduce their total energy requirements, possibly leading to the smaller size observed. Likewise, the decline in food availability which occurs during winter is thought to have been the selective force responsible for the seasonal reduction in body mass of both *M.montanus* (Wunder et al. 1977) and *P.sungorus* (Heldmaier & Steinlechner 1981).

In conclusion, the present study confirms that pouched mice display considerable geographical variation in energy requirements together with differences in the use of photoperiod as an anticipatory cue for predicting the onset of winter. These differences appear to be related to the availability of energy and relative severity of climatic conditions in each respective locality.

CHAPTER 7: GEOGRAPHICAL VARIATION IN THE EXPRESSION OF SPONTANEOUS TORPOR BY POUCHED MICE

Introduction

Small mammals face a substantial increase in the rate of heat loss when subjected to cold stress as a result of their large surface area to volume ratio (Hart 1971). Although this should cause higher thermoregulatory costs, heat loss can be reduced by improving thermal insulation, or by lowering body temperature and entering torpor so that the temperature gradient between ambient and body core is reduced (Vogt & Lynch 1982). Torpor is characterised by a drop in body temperature below 30°C followed by spontaneous arousal to normothermia (Hudson 1978) and can be elicited either by food restriction ("induced torpor" eg. Buffenstein 1985) or cold stress ("spontaneous torpor" eg. Hill 1975). "Because small mammals from different environments do not experience identical seasonal fluctuations in stress-provoking stimuli it is reasonable to expect them to exhibit different tendencies toward torpor" (Tannenbaum & Pivorun 1988) and several studies have discovered geographical differences in the expression of torpor which are associated with differences in climatic conditions (Brenner & Lyle 1975; Hill 1975; Heath & Lynch 1983; Thompson 1985; Tannenbaum & Pivorun 1984).

S.campestris enter spontaneous torpor in response to cold stress (Ellison & Skinner in press) and, in common with other subtropical, semi-fossorial mammals (Downs and Perrin 1989), they also experience seasonal and geographical differences in temperatures above and below ground throughout their range in southern Africa (see Chapter 2). The aim of the present study was therefore to establish whether pouched mice from colder localities exhibit spontaneous torpor to a greater extent than those from milder areas.

Materials and methods

Animals

Torpor was examined in pouched mice which originated from six localities in southern Africa and displayed variable karyotypes (see Table 24 for details). Rainfall and air temperature data for each locality were obtained from their nearest meteorological station and are summarised in Table 25.

A total of 36 pouched mice, comprising six individuals from each locality, was studied. Eighteen individuals of each sex were used and approximately equal numbers of these were derived from wild-caught (n=17) and first generation laboratory-bred (n=19) stocks. Laboratory-bred mice were reared under long photoperiod (14L:10D) at 25°C and were at least four months old at the start of the present study. Wild-caught animals had been exposed to similar conditions for at least four months before the experiments began.

Throughout the present study the pouched mice were housed separately in standard laboratory rat cages and provided with rabbit pellets, rat cubes (Epol, Vereeniging) and water ad libitum, a diet occasionally supplemented with sunflower seeds and pieces of apple. Shredded paper and sawdust were provided for bedding and their cages were cleaned weekly, and dusted with insecticide to eradicate ectoparasites (Karbadust: Sentrachem, Silverton. Containing 50g/kg Carbamate).

Acclimation

Although previous studies have confirmed that *S.campestris* enter spontaneous torpor at cold temperatures (5-15°C; Ellison & Skinner in press), short photoperiod seems to facilitate torpor in this species because more individuals enter torpor when acclimated to short photoperiod than when acclimated to long photoperiod (Ellison, Skinner & Ferguson in prep.). In order to elicit torpor in as many animals as possible the pouched mice in the present study were acclimated to short photoperiod (10L:14D) at 25°C for four weeks prior to cold acclimation (10°C or 15°C) at the same photoperiod.

Table 24 Summary of the six pouched mouse populations examined in Chapter 7

Locality	(Grid Reference)	Karyotype (2n)	Mean body mass (M_b g)	Males/Females (n/n)
Letaba	(Kruger National Park, E.Transvaal, RSA: 23°56'S 31°35'E)	46	71,8 ±11,0	3/3
Nwanedzi	(Kruger National Park, E.Transvaal, RSA: 24°23'S 31°40'E)	46	81,9 ±11,3	4/2
Gam	(S.E.Bushmanland, Namibia: 20°27'S 20°44'E)	35	71,8 ±11,0	3/3
Pniel	(Barkly West, Kimberley, N.E.Cape, RSA: 28°35'S 24°31'E)	32	75,9 ±13,1	3/3
Schuinklip	(N.E. Pofadder, N.Cape, RSA: 28°51'S 19°42'E)	35	49,8 ± 6,3	2/4
Vaalkop	(Brits, Central Transvaal, RSA: 25°23'S 27°28'E)	44	79,9 ±18,0	3/3

Table 25 Climatic conditions at the pouched mouse localities examined in Chapter 7

Locality	(Meteorological Station)	Altitude (m)	Mean Annual Rainfall (mm)	Mean Daily Temperatures Summer (Nov.- Feb.) Air Max. (°C)	Summer (Nov.- Feb.) Air Min. (°C)	Winter (May - Aug.) Air Max. (°C)	Winter (May - Aug.) Air Min. (°C)
Letaba	(Letaba 23°51'S 31°35'E)	215m	387	33.2	20.8	27.0	9.5
Nwanedzi	(Satara 24°24'S 31°47'E)	437m	437	32.2	19.8	26.2	9.2
Gam	(Grootfontein 19°36'S 18°08'E)	1398m	611	30.7	17.5	24.8	6.1
Pniel	(Barkly West 28°32'S 24°32'E)	1204m	391	31.8	15.8	20.7	2.5
Schuinklip	(Pofadder 29°14'S 16°52'E)	989m	105	31.8	15.7	18.9	6.0
Vaalkop	(Brits 25°35'S 27°49'E)	1158m	621	29.8	16.2	22.3	3.0

Following four weeks cold acclimation the oxygen consumption (VO_2) of each pouched mouse was recorded for 24h at 10°C to determine the incidence, depth, duration and relative energy savings of spontaneous torpor in *S.campestris* from each locality.

Oxygen Consumption

VO_2 was measured using an open-circuit system, as described in Chapter 5. The mice were placed in large (13 l), perspex metabolic chambers, through which a flow of dried air (silica gel) was passed at a rate of 600ml·min^{-1}. The chambers were immersed in a constant temperature water bath (Labotec, Isando), and a thermocouple (J-Type: Grant Instruments, Cambridge) within the chamber was used for monitoring its temperature. The water bath was covered by a light-tight hood and short photoperiod was maintained using an electric light connected to an automatic time switch.

The pouched mice were transferred to these chambers together with their bedding, food and water (fresh apple), at least 24h prior to the start of measurement to allow them to become habituated to their new surroundings. VO_2 was subsequently recorded every 30 min for 24h.

Following full equilibration of the system, the air leaving the chambers was dried and its oxygen content determined using an Ametek S-3A/I oxygen analyser (Applied Electrochemistry, Pittsburgh) connected to a multi-channel data logger (1200 Series Squirrel: Grant Instruments, Cambridge). An in-line, three way valve (Air/Water 350 KPA: Ascoreg, Johannesburg) and time switch alternated the input to the oxygen analyser so that air from two chambers, containing separate pouched mice, could be recorded twice an hour using a single oxygen analyser. The oxygen analyser was calibrated before and after each measurement and all readings were corrected to standard temperature and pressure, dry (STPD). Gross VO_2 was converted to mass specific VO_2 using the mean of body mass measurements taken at the beginning and end of each experiment.

To compensate for any differences in M_b metabolic measurements were expressed in terms of [mass in grams]^{0,75} as suggested by Hart (1971). Average daily metabolic rate (ADMR) was calculated as the mean VO_2 measured over a full 24h. Minimum metabolic rate (VO_{2min}) was taken as the lowest value of VO_2 sustained for at least 15 minutes during this period.

Body Temperature

Body temperature (T_b) was measured in individuals from three localities (*Letaba*, *Gam* and *Pniel*) using small (1.2g) temperature telemeters (Model X: Minimitter, Sunriver) which provided accurate temperature measurement to within 0.5°C. Sterilised telemeters were surgically implanted in the peritoneal cavity of anaesthetised (75mg/kg *i.p.* Sagatal: Maybaker, Port Elizabeth) pouched mice under veterinary supervision, and the mice were given at least five days to recover from surgery before measurements were made. The telemeters were calibrated prior to implantation and at the end of each experiment, during which time there was negligible drift of telemeter calibration.

Signals from implanted telemeters were received using small AM radios placed within the metabolic chambers. It wasn't possible to record T_b continually because electromagnetic interference from laboratory equipment inhibited the reception of pulses from the minimitters (see also Ruf & Heldmaier 1987). However, daily torpor usually takes place at a predictable time of day (Tucker 1965; Lynch & Gendler 1980) and previous studies have revealed that torpor cycles of pouched mice always occur during the first few hours of photophase (Ellison et al. in prep.). For this reason we recorded T_b manually using headphones once an hour following lights on between 07h00 and 12h00.

Statistical analyses

All results are presented as mean $\pm$ one standard deviation. The Mann-Witney U-test (U - MWU), Chi-squared test (X^2 - CS) and Spearman's rank correlation coefficient (r_s - SCC) were used for the statistical analysis of results

(Sokal & Rohlf 1981).

Results

Body Mass

Although pouched mice from most localities exhibited a mean body mass (M_b) of 70-80g (Table 24) those from *Schuinklip* weighed only 49,8 ±6,3g and were significantly smaller than individuals from other localities (MWU: $U<2$, $n_1=n_2=6$, $p\leq0,01$). Females (M_b=67,6 ±14,2g) were also smaller than males (M_b=75,9 ±15,8g) whilst laboratory-bred mice (M_b=71,5 ±13,8g) were slightly smaller than wild-caught animals (M_b=73,8 ±16,4g). However, these differences were not significant (Sex: U=118,5, $n_1=n_2=18$, $p>0,05$. Stock: U=117,5, $n_1=17$ $n_2=19$, $p>0,05$).

Torpor Cycles

The T_bs of six pouched mice containing implanted telemeters (three from *Gam*, two from *Letaba* and one from *Pniel*) fell below 30°C and these animals were considered to have entered torpor (Hudson 1978). Their ADMRs were characterised by a distinct decline in VO_2 during the early morning which fell below 50% of resting levels for more than two hours and ended in a sharp increase in VO_2 which overshot resting levels temporarily during arousal (Fig. 17). Similar bouts of reduced metabolism were displayed by nine untelemetered pouched mice (three from *Nwanedzi*, three from *Schuinklip* and three from *Vaalkop*) and daily variation in VO_2 of these animals is also displayed in Fig. 17. Of those individuals which entered torpor, six were female and nine were male, whilst seven were laboratory-bred and eight were wild-caught. These proportions didn't differ significantly from parity (CS: Sex: X^2=0,013, d.f.= 1, $p>0,05$. Stock: X^2=0,002, d.f.= 1, $p>0,05$) and there was also no significant difference between the M_b of pouched mice which entered torpor (73,4 ±19,4g) and those remaining normothermic (70,6 ±12,1g. MWU: U=146, $n_1=15$ $n_2=21$, $p>0,05$).

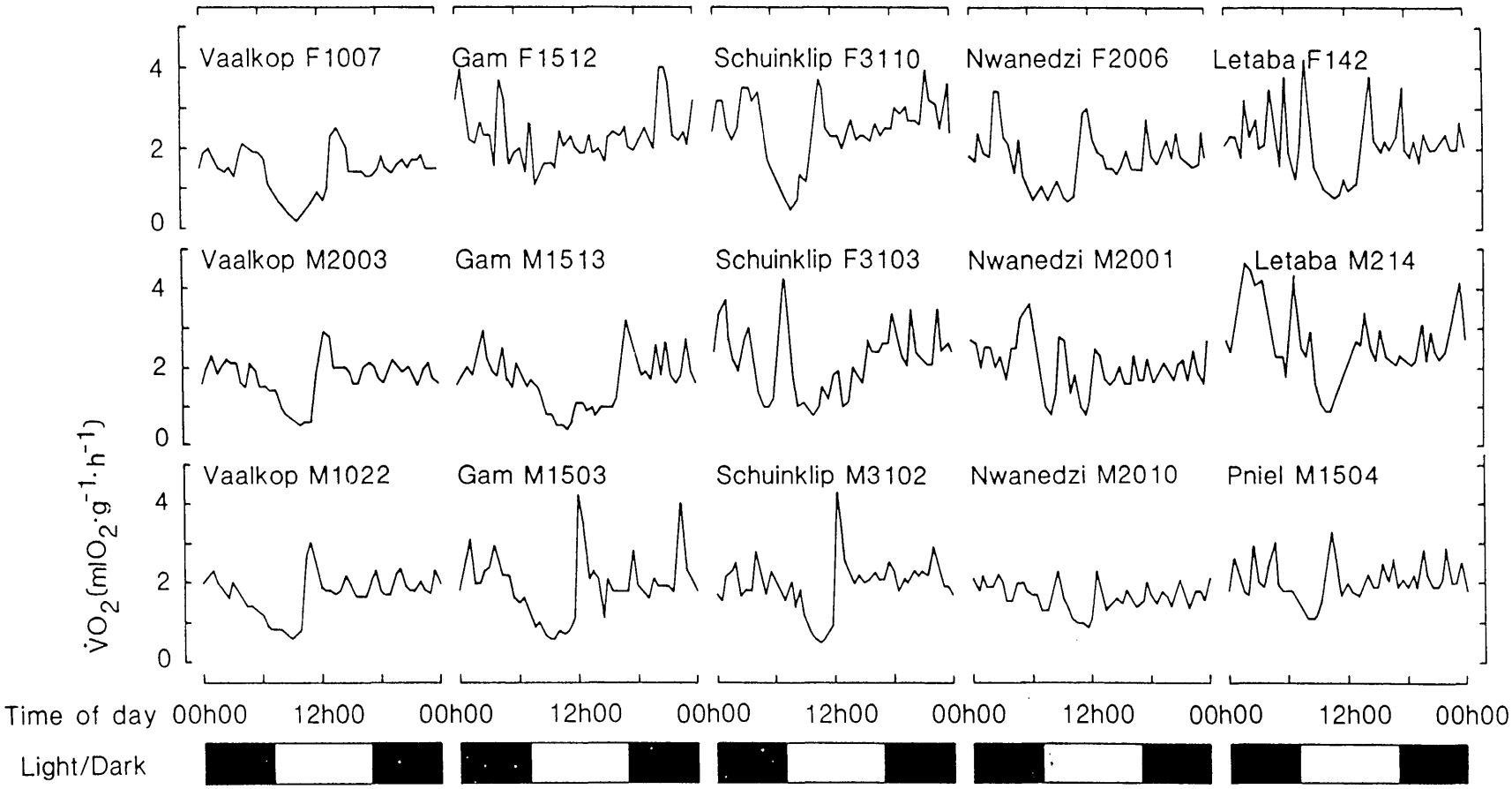


Figure 17 Oxygen consumption (VO_2) of 15 pouched mice recorded over 24 h at T_a=10°C. Each pouched mouse exhibited a bout of spontaneous torpor which was characterised by a distinct decline in VO_2 during the early morning which fell below 50% of resting levels for more than 2 h and ended in a sharp increase in VO_2 during arousal to normothermia.

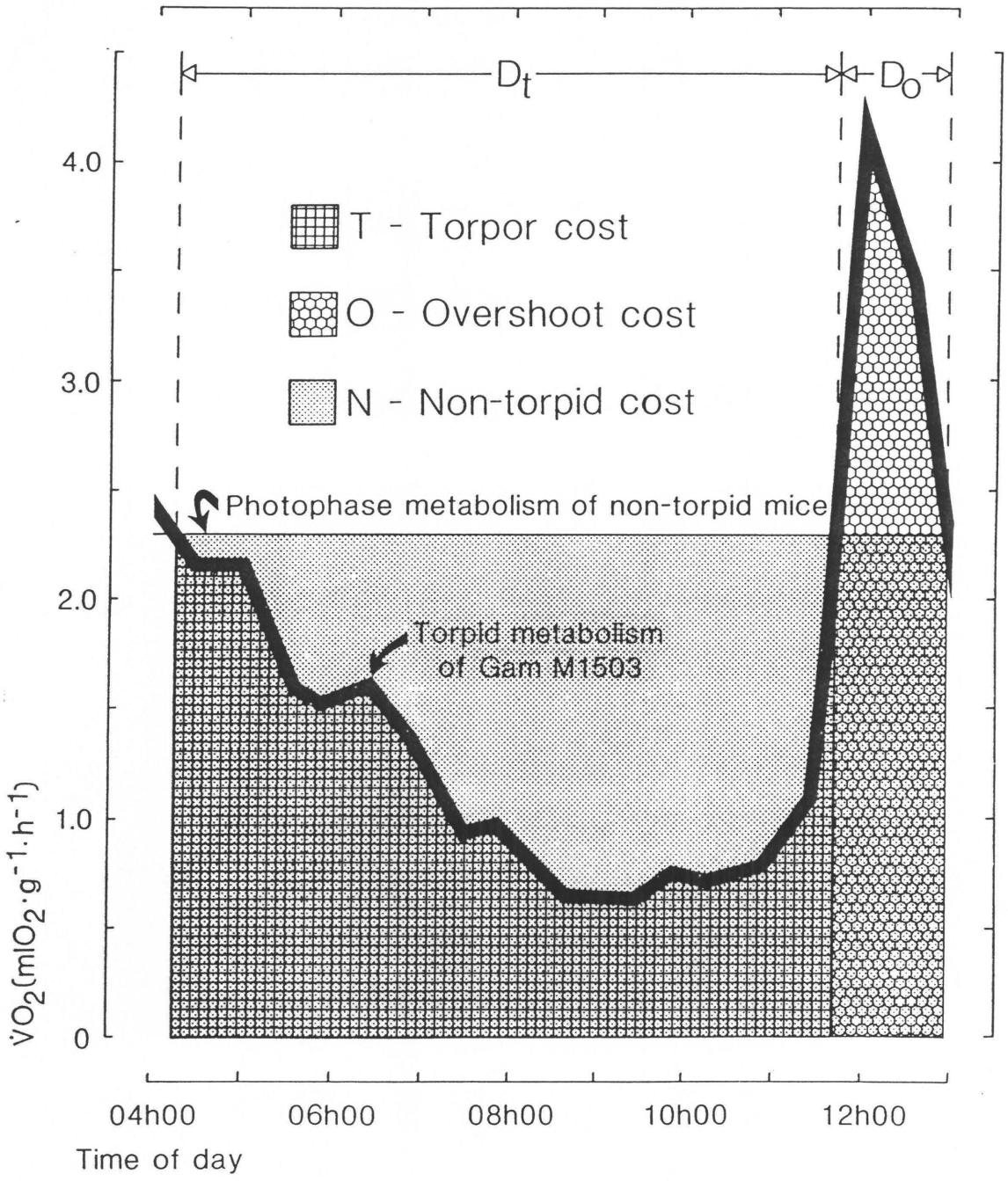


Figure 18 Calculating the duration of torpor (D_t), the duration of overshoot (D_o), the cost of torpor (T), the cost of overshoot (O) and the equivalent cost of non-torpid pouched mice during the same period (N) by comparing the oxygen consumption of a torpid individual (Gam 1503) to the mean oxygen consumption of non-torpid animals recorded during photophase.

In order to quantify the duration and relative energetic savings for each bout of torpor, the metabolism of torpid individuals was compared to that of euthermic pouched mice from the same locality (see Fig.18, and Ellison & Skinner in press). Torpor duration (D_t) was determined as the period during which the metabolism of torpid animals fell below the mean metabolism of non-torpid mice measured during photophase (the resting phase of *S.campestris*). In the same way the duration of overshoot (D_o) was calculated as the period in which the metabolism of the arousing animal exceeded the mean metabolism of non-torpid mice measured during photophase. The energy savings of each bout of torpor were then determined by calculating the total oxygen consumption during torpor (T), the total oxygen consumption during overshoot (O), and the equivalent oxygen consumption of non-torpid individuals over the same period of time (N). The energy savings (N-[T+O]) were then expressed as a percentage of N. These data are summarised in Table 26 together with VO_{2min} recorded.

As an indication of torpor depth, VO_{2min} was inversely correlated with torpor duration (D_t) (SCC: r_s=-0,705, n=15, p<0,01). However, there was no correlation between VO_{2min} and the energy saved during each bout of torpor (r_s=-0,243, n=15, p>0,05), although there was a positive correlation between energy savings and D_t (r_s=0,448, n=15, p<0,05). The duration of overshoot (D_o) was positively correlated with its cost (O) (r_s=0,850, n=15, p<0,001) whilst D_o and O were both inversely correlated with energy savings (D_o: r_s=-0,591, n=15, p<0,05. O: r_s=-0,729, n=15, p<0,01). Female pouched mice did not display deeper or longer bouts of torpor than male animals (MWU: U=26,5/25,5, n_1=6 n_2=9, p>0,05) and did not accrue greater energy savings than males (U=27, n_1=6 n_2=9, p>0,05). There was also no significant difference between laboratory-bred and wild-caught pouched mice with regard to these characteristics (U>15, n_1=7 n_2=8, p>0,05).

Table 26 A summary of metabolic measurements of eight pouched mice entering spontaneous torpor

Locality:	Sex: (M/F)	No:	Stock: (W/L)	M_b (g)	VO_{2min} ($ml O_2 \cdot M_b^{-0,75} \cdot h^{-1}$)	Torpor duration: (D_t - h)	Overshoot duration: (D_o - h)	Torpor cost: (T - $ml O_2 \cdot M_b^{-0,75}$)	Overshoot cost: (O - $ml O_2 \cdot M_b^{-0,75}$)	Non-torpid Equivalent (N - $ml O_2 \cdot M_b^{-0,75}$)	Energy Savings: (per bout of torpor)
Letaba	F	142	L	85,4	2,332	4,9	1,5	18,209	12,826	40,960	24,2%
Letaba	M	214	L	61,4	2,427	3,8	2,4	15,227	18,062	39,823	16,4%
Nwanedzi	F	2006	L	67,0	1,966	6,1	1,6	18,514	11,425	47,454	36,9%
Nwanedzi	M	2001	W	89,9	2,516	4,6	1,9	18,006	13,989	39,440	18,9%
Nwanedzi	M	2010	L	97,6	2,631	3,8	0,3	13,892	2,074	24,958	36,0%
Gam	M	1513	W	77,9	1,153	12,0	1,2	41,034	9,929	85,975	40,7%
Gam	M	1503	W	83,4	1,837	7,5	1,2	25,830	12,392	56,510	32,4%
Gam	F	1512	W	72,2	3,253	2,9	0,3	13,123	1,667	20,192	26,8%
Pniel	M	1504	W	66,9	3,020	4,8	1,2	19,379	8,488	36,404	23,5%
Schuinklip	F	3110	L	48,8	1,290	4,7	3,0	13,746	21,403	41,465	15,2%
Schuinklip	F	3103	L	44,0	2,130	8,5	1,6	27,723	12,543	54,291	25,8%
Schuinklip	M	3102	L	49,2	1,292	6,1	2,0	20,215	14,572	43,758	20,5%
Vaalkop	F	1007	W	55,0	0,651	8,5	1,7	22,530	9,915	58,808	44,8%
Vaalkop	M	2003	W	100,1	1,702	6,6	1,7	22,173	13,216	47,807	26,0%
Vaalkop	M	1022	W	102,0	1,964	8,8	1,6	32,915	12,289	59,686	24,3%

Geographical Differences

There were no significant geographical differences in the incidence of torpor among pouched mice in the present study (CS: X^2=3,800, d.f.=5, p>0,05). However, the expression of torpor was variable and individuals from localities with colder winters (ie. with lower minimum air temperatures) had significantly longer bouts of torpor (D_t) than those from warmer localities (SCC: r_s=-0,465, n=15, p<0,05). Despite the positive association between D_t and energy savings (see above) there was no significant correlation between minimum temperatures and energy saved (r_s=-0,062, n=15, p>0,05). There was also no correlation between locality temperature and VO_{2min} (r_s=0,350, n=15, p<0,05).

Discussion

The pouched mice examined in the present study displayed typical bouts of torpor which included entry, torpor itself and arousal to normothermia (Tucker 1965). The duration of each bout is thought to be determined by the depletion of nutrients and/or the production of metabolites (Twente & Twente 1965; Geiser 1988). As both of these factors are dictated by metabolic rate it is not surprising that the duration of torpor was inversely related to its depth (VO_{2min}). However, because VO_2 varied during torpor, VO_{2min} didn't always provide an accurate indication of torpid metabolism and there was a closer correlation between the duration and cost of torpor (T) (p<0,001) than between its depth and cost (p<0,05). This might explain why the duration of torpor correlated with energy savings while VO_{2min} did not. Meanwhile, the inverse relationship between the cost of arousal and energy saved supports the suggestion by Prothero & Jürgens (1986) that the cost of regaining homeothermy can limit the energetic benefits of short bouts of torpor.

Several studies have previously reported geographical variation in the incidence of torpor between different populations of identical or closely related rodent species (Brenner & Lyle 1975; Hill 1975; Heath & Lynch 1983; Thompson 1985; Tannenbaum & Pivorun 1984; 1987b; 1988). In each case, animals from warmer localities rarely entered torpor or did so in fewer numbers than those from

colder areas which suggests that selection for torpor was greater in more energetically stressful environments (Tannenbaum & Pivorun 1988). In the present study, there were no such geographical differences in the proportion of pouched mice entering torpor and a similar number of individuals from populations throughout southern Africa had the capacity to exhibit torpor. However, there were significant geographical differences in torpor duration which are similar to those reported for *Peromyscus* species by Tannenbaum & Pivorun (1984). In their study *P. maniculatus*, from a montane habitat, had significantly longer bouts of torpor than two lowland species (*P.leucopus* and *P.gossypinus*) even though, like *S.campestris* in the present study, there were no differences in torpor depth (as determined by minimum body temperature).

Although their original study was conducted upon acclimatized, wild-caught individuals, these authors subsequently confirmed the genetic basis for these differences by repeating the measurements on laboratory-bred animals (Tannenbaum & Pivorun 1987b). In the present study there is strong evidence that the differential expression of torpor by *S.campestris* is also genetically derived: first, wild-caught animals had been subjected to extensive acclimation and secondly, approximately equal proportions of laboratory-bred and wild-caught mice entered torpor. Indeed, the variable karyotype of *S.campestris* might possibly facilitate a genetic role in the differential selection of torpor as suggested by Lyman et al. (1983). They discovered significant differences in the frequency of torpor in two populations of Turkish hamsters, *Mesocricetus brandti*, from similar habitats, only 125 km apart, and suggested that these differences might be attributable to variation in karyotype (Lyman, O'Brien & Bossert et al. 1983). However, in view of Ferreira's (1990) findings (see Chapter 1 and Fig. 5) it seems unlikely that karyotypic diversity played a role in the differential expression of torpor reported here, although this possibility will be discussed further in Chapter 8.

CHAPTER 8: EVOLUTIONARY CONSIDERATIONS

Introduction

Despite the "usefulness" of optimality theory as a mechanism for quantifying adaptation (Diamond 1992) natural selection is directed towards survival, rather than perfection and only operates on organisms when they are "stressed" (Berry 1985; Schmidt-Nielsen 1986). Stresses can be either biological (eg. competition, predation, parasitism) or physical (eg. temperature, humidity, light intensity) in origin and involve "a cost to an organism which can be lessened behaviourally, biochemically, physiologically or morphologically" (Berry 1985: 225). Biological stresses are usually difficult to identify and assess because they involve complex interactions between different organisms (Krebs 1978; Berry 1985). However, physical stresses, such as climatic factors, often result in quantifiable energetic costs which can be measured directly (Grodzinski et al. 1975). For this reason, the preceeding chapters were able to identify a variety of behavioural, morphological and physiological mechanisms which limit the effect of climatic stresses on the energy expenditure of *Saccostomus campestris* throughout its range in southern Africa.

However, adaptations do not occur in isolation but interact with each other in one of two ways: either they reduce the impact of an environmental stress on other traits or they cause associated changes in closely related characters which result in positive ("pre-adaptive"), neutral, or even negative ("mal-adaptive") consequences (Gould & Johnston 1972; Gould & Lewontin 1979; Berry 1985). At the same time, the presence of a certain trait does not necessarily mean that selection has taken place, because it may simply be the result of phenotypic flexibility or pre-adaptation. The aim of this chapter is therefore to assess the relative importance of behaviour, morphology and physiology as avenues for energetic compatibility in *S.campestris*, by examining how different mechanisms interact, and to evaluate to what extent these beneficial traits are the result of selection.

Behavioural adaptations

Pouched mice display four behavioural adaptations which enable them to largely avoid unfavourable temperatures: They make use of subterranean burrows where diurnal, seasonal and geographical variation in ambient temperature is buffered (see Fig. 9 and Table 4). They are nocturnal and remain in their burrows during the heat of the day (Skinner & Smithers 1990). They hoard food throughout the year to minimise the duration of foraging above ground at night (Chapters 2 and 3). And finally, they reduce their activity following a decline in temperature (Ellison et al. in prep.) so that they spend even less time outside their nest when it is cold.

The coldest burrow temperatures recorded in Chapter 2 were around 15°C which are 10-15°C below the thermoneutral zone (Ellison & Skinner 1991b; Haim et al. 1991; Chapters 5 and 6). However, several studies have shown that small mammals warm up their nests by 8-21°C when they are inside (Sealander 1952; Stark 1963) so that pouched mice probably experience thermoneutral temperatures throughout the year within their burrows in southern Africa. Under these conditions there would be no necessity to improve the insulation of burrows using additional nesting behaviour, which might explain why little bedding material was found in many of the burrows examined (Table 8), and why there was no effect of photoperiod, temperature or locality on nest-building in captivity (Chapter 3).

Morphological adaptations

Traditionally, geographical variation in the body size of homeotherms has been interpreted as an adaptation to modify heat loss in response to differences in environmental temperature, because small individuals have a higher surface area to volume ratio and lose relatively more heat than larger animals. This view is central to Bergmann's and Allen's rules which predict that individuals from cooler environments should be larger (Bergmann's rule) and have smaller appendages (Allen's rule) to improve their thermal insulation (Allaby 1985). However, animals need sufficient food to attain and maintain a given body size. Consequently, rainfall (as an index of food availability) might be expected to mask the effect of

temperature in arid and semi-arid parts of southern Africa, where droughts are endemic (Tyson 1986), primary productivity is low and food resources are limiting (Louw & Seely 1982). This would explain why the body size of pouched mice from throughout the subregion displayed a stronger correlation with rainfall than with temperature while animals from localities with lower rainfall were smaller than those from areas that receive more rain (Chapters 4 and 5).

At the same time, it is possible that rainfall might assume increasing importance among species like *S.campestris* which behaviourally avoid unfavourable temperatures and experience limited geographical variation in thermal conditions within their burrows. This view is supported by the observation that there was no clear correlation between appendage size and temperature (as predicted by Allen's rule) even though this would not necessarily have counteracted the effect of a positive association between body size and rainfall.

Physiological adaptations

In common with other semi-fossorial mammals (eg. *Gerbillurus paeba*, Buffenstein 1984), *S.campestris* displayed a lower basal metabolic rate (BMR/MOMR) and a minimum thermal conductance (C_m) equal to or higher than that predicted for cricetid species of similar size (Table 18). Although these characteristics are thought to help limit the build up of heat within a burrow (McNab 1979), they would have contradictory effects on the environmental compatibility of pouched mice in southern Africa: A low MOMR would help reduce their energy requirements and would pre-adapt them to an arid/semi-arid region where food availability is limited (eg. Buffenstein 1984). In contrast, a normal or high C_m could be mal-adaptive because it would facilitate heat loss during periods of activity when they are exposed to cold nocturnal temperatures outside the burrow. However, it is difficult to evaluate the influence of a high or low C_m on heat loss during activity as this is also affected by changes in posture which occur during movement (Wunder 1975). For this reason, it is probably more relevant for pouched

mice to have a relatively high C_m as an adaptation to fossorial conditions, particularly because they spend most of their lives underground.

As a result of the positive correlation between rainfall and body size (Chapters 4 and 5) it would not have been surprising to find a negative correlation between rainfall and MOMR, C_m or nonshivering thermogenesis (NST) because all three are inversely proportional to body mass (Heldmaier 1971; Bradley & Deavers 1980; Hayssen & Lacy 1985). However, after correcting for geographical variation in body mass (as described in Chapter 5) there were no significant correlations between MOMR or C_m and body mass or rainfall. Although there was a significant correlation between NST and rainfall (see Table 20) this was probably not a residual effect of body mass because it was positive rather than negative (Chapter 5).

Despite significant intraspecific differences in energy metabolism between pouched mice from different localities in southern Africa (Chapter 5) there were no correlations between local climatic conditions and MOMR, C_m, or body temperature which are three parameters that represent the metabolism of resting animals. This suggests that none of these characteristics are adapted to climate in this species. Alternatively, it might simply indicate that pouched mice do not experience significant geographical variation in temperature when at rest inside their burrows so that there may be no selection pressure for ecotypic adaptation of their resting metabolism.

However, there are limits to which all species, including *S.campestris*, can avoid unfavourable conditions (Hill 1983), and pouched mice undoubtedly experience geographical variation in nocturnal temperatures when they are active above ground at night. These temperatures would normally be well below thermoneutrality (see Table 4) so that pouched mice would have to increase their heat production to maintain homeothermy. This would explain why the capacity for NST, an important form of heat production (Wunder 1984), was significantly higher among animals from cooler localities (Chapters 5 and 6). Likewise, pouched mice would also experience a reduction in nocturnal temperature during winter which

would explain why they are able to increase their capacity for NST following cold acclimation (Ellison & Skinner 1991b; Haim et al. 1991; Chapter 6). However, seasonal changes in temperature can be unpredictable, and many species use a decline in photoperiod to anticipate the onset of winter (Heldmaier et al. 1989), particularly in areas where conditions are harsh at this time of year. For this reason, pouched mice from colder, more seasonal environments displayed a larger increase in NST capacity in response to a decline in photoperiod than animals from milder areas (Chapter 6).

Finally, the adaptive significance of geographical variation in spontaneous torpor (Chapter 7) remains unclear. Because pouched mice accrue considerable savings in energy expenditure during these bouts of torpor, an increase in the duration of torpor at cooler localities might be interpreted as an adaptation to reduce the higher thermoregulatory costs in these environments. However, pouched mice enter torpor during periods of rest only at 15°C and below (Ellison & Skinner in press) yet these temperatures are much lower than those recorded at any of the burrows examined in South Africa, even during winter at the coldest locality (Chapter 2). For this reason it is tempting to suggest that the correlation between locality temperature and torpor duration is spurious, unless it indicates that pouched mice have experienced more severe conditions during their evolutionary history. Having evolved more than 3,5 million years ago (Denys 1988) *S.campestris* would have experienced at least 17 glacial periods since the middle of the Pliocene (Brain 1985) during which lower global temperatures might have favoured animals capable of spontaneous torpor.

The interaction between behaviour, morphology and physiology

From the above review it is clear that semi-fossoriality is the most important avenue for climatic adaptation in *S.campestris* because it appears to have reduced the importance of geographical variation in temperature as a selection force on nesting behaviour, body size and resting metabolism. However, pouched mice cannot completely escape variation in food availability or nocturnal ambient

temperatures in their local environment. For this reason they display additional ecotypic adaptations in body size, heat production and torpor which enable them to cope with geographical differences in primary productivity and ambient temperature within southern Africa.

The interaction between heredity and the environment

According to Gould & Johnston (1972: 480) "the *prima facie* case for selection is an excellent correspondence between environment and form, with a sensible adaptive explanation for the phenetic differences". Unfortunately, the environment can also cause non-genetic changes to phenotype which may be difficult to distinguish from heritable effects because they are often in the same direction as those caused by "selectively ordered genetic variation" (Gould & Johnston 1972). These environmental changes include temporary (acclimatory) effects on the phenotype of adults and permanent (epigenetic) changes on that of developing juveniles.

In adult pouched mice it is clear that body size, NST capacity and the incidence of torpor are all susceptible to modification by environmental conditions: Captive individuals were considerably larger than those in the wild (compare M_b in Tables 11 and 13 with those in Table 18), whilst young animals appear to lose weight during winter (Ellison et al. in press). NST capacity increased significantly following cold acclimation (Ellison & Skinner 1991b; Haim et al. 1991), and, in some populations, after a decline in photoperiod as well (Chapter 6). The incidence of torpor is facilitated by short photoperiod (Ellison et al. in prep.), and increases at lower temperatures (Ellison & Skinner in press).

Meanwhile, a large number of studies have shown that manipulating environmental conditions during development (particularly temperature and food availability) can influence adult body size (eg. Ogle 1934; Dauncey & Ingram 1986). Likewise, Mason (1974), Lynch et al. (1977), and Caputa & Kamari (1991a, b) have demonstrated that the early thermal history of rodents can cause long-term effects on the subsequent metabolism of adults, although Muraliidhara & Shetty (1986)

found that pre-weaning food deprivation did not result in any permanent changes in the basal metabolism or thermoregulatory thermogenesis of rats. Although I know of no studies that have examined the effect of ontogenetic environment on the expression of torpor, epigenetic factors cannot be ruled out as possible causes of the ecotypic differences found in the present study, because many of the pouched mice examined had grown up under natural conditions which might have differed from area to area.

In an attempt to minimise these effects many studies (including those in Chapters 5, 6 and 7) have measured the metabolism of wild-caught animals only after they had been maintained under standard laboratory conditions for prolonged periods of time (1-6 months: eg. Hayward 1965b; Bowers 1971; Bradley & Deavers 1980; Buffenstein & Jarvis 1985b). Although this cannot rule out the chance that epigenesis has had an effect, it does help to eliminate phenotypic changes caused by acclimatisation and makes it more likely that observed differences between populations are largely the result of heritable factors.

This possibility is particularly interesting for *S.campestris*, because it begs the question as to whether there is any connection between ecotypic adaptation and the high degree of karyotypic variability displayed by this species (Gordon 1986, see Fig. 4). In this context, two models of chromosomal speciation ("canalization" and "recombinational breakdown") have suggested that karyotypic changes might produce linkage groups which are adaptive to a particular environment (Sites & Moritz 1986). These models have been widely criticised because there is little empirical evidence for either of them (eg. King 1985; Sites & Moritz 1986). Nevertheless, a series of studies on geographical variation in mole rats (*Spalax ehrenbergi*) seem to support the concept of an adaptive karyotype as they found significant correlations between phenotype, karyotype and environmental factors (Nevo & Shkolnik 1974; Haim et al. 1984; Haim, Heth & Nevo 1985; Nevo et al. 1986; Yahav, Simson & Nevo 1988; 1989). Indeed, Nevo, Beiles & Ben-Shlomo (1984) have even suggested that genetic-environment correlations demonstrate "inferentially" (sic) their adaptive significance. However, *S.ehrenbergi* occasionally

displays more than one energetic phenotype per karyotype (eg. Haim et al. 1985; Nevo et al. 1986) which suggests that karyotype does not necessarily pre-determine the phenotypic response of different "chromosomal species". Likewise, *S.campestris* displays more than one energetic phenotype per diploid number (see Tables 18 and 26), and even exhibited more than one diploid number per locality (Gordon 1986, see Fig. 4). Therefore, even if ecotypic differences in energy metabolism of pouched mice are caused by genetic factors, it is clear that these are not the result of geographical variation in karyotype.

SUMMARY

The aim of the present study was to examine geographical variation in thermoregulation and energy metabolism of the pouched mouse (*Saccostomus campestris*) in southern African. This species is widely distributed throughout the subregion, and displays a broad habitat tolerance, occurring at altitudes from sea level to 1800m, and in areas experiencing mean annual rainfall between 100mm and 1200mm.

Field observations in South Africa confirmed that pouched mice are solitary, nocturnal and semi-fossorial. This lifestyle enables them to avoid the heat of the day and spend much of their lives in subterranean burrows where there is little diurnal, seasonal or geographical variation in temperature. Although they face cold temperatures when they are active above ground at night, they minimise the time spent foraging by collecting food in their cheek pouches and returning this to the nest where it is eaten in relative warmth and safety. However, neither photoperiod, temperature nor locality affect nest-building or food-hoarding behaviour in captivity, suggesting that these activities do not respond to seasonal or geographical changes in climate.

Geographical variation in body size was strongly correlated with rainfall and pouched mice from localities with lower rainfall were smaller than those from areas that receive more rain. This correlation might have been caused by food restriction in arid and semi-arid environments where primary production and food availability are low. However, this relationship was also apparent in captive animals which had access to food ad libitum. As smaller individuals require less food than larger ones, a reduction in body size might represent an adaptation to help reduce the energy requirements of pouched mice living in drier areas where food supplies are limited.

S.campestris had a lower than expected basal metabolic rate (BMR ≡ minimum observed metabolic rate: MOMR) and a minimal thermal conductance (C_m)

that was equal to or greater than that predicted for animals of similar size. These characteristics are thought to help burrow-dwelling species avoid over-heating within the confines of a humid burrow, although they would also have contradictory effects upon the environmental compatibility of pouched mice in southern Africa: A low BMR would help reduce the energy requirements of animals in this arid/semi-arid region where primary productivity and food availability is low. In contrast, a relatively high C_m would facilitate heat loss and increase the cost of thermoregulation in the cold, particularly during periods of nocturnal activity.

After correcting for geographical variation in body mass there were still significant differences in energy metabolism between pouched mice from nine localities in southern Africa which suggested that these populations were physiologically distinct. However, there were no significant correlations between local climatic conditions and MOMR, C_m or body temperature which are three characteristics that determine the metabolism of resting animals. This suggests that these parameters are not adapted to climate. Alternatively, if pouched mice experience little geographical variation in temperature when they are at rest inside their burrows, there might be insufficient selection pressure for ecotypic adaptation of their resting metabolism.

Nevertheless, pouched mice would still be subjected to some geographical variation in ambient temperature while active outside their burrows at night. This might explain why individuals from cooler environments displayed a higher capacity for non-shivering thermogenesis (NST) than those from warmer areas. NST is an important form of heat production in small rodents and would help pouched mice maintain homeothermy during nocturnal foraging.

Pouched mice would also experience a reduction in nocturnal temperature during winter. However, seasonal changes in temperature can be unpredictable and many species use a decline in photoperiod to anticipate the onset of winter, particu-

larly in areas where conditions are harsh at this time of year. For this reason pouched mice from colder, more seasonal localities displayed a larger increase in NST capacity in response to a decline in photoperiod, even though animals from throughout southern Africa displayed a similar improvement in heat production (MOMR and NST) following cold acclimation.

As pouched mice accrue substantial energy savings during bouts of spontaneous torpor, the increase in the duration of torpor observed at cooler localities would enable them to reduce the higher costs of thermoregulation in these areas. However, the functional significance of spontaneous torpor in this species remains unclear, because it only occurs during periods of rest at temperatures of 15°C and below which are lower than those normally encountered in pouched mouse burrows. It therefore appears that the correlation between locality temperature and torpor duration is spurious unless it indicates that pouched mice experienced more severe conditions at some stage in their evolutionary history.

The present study clearly demonstrates that pouched mice from southern Africa display a diverse array of behavioural, morphological and physiological mechanisms which make them energetically compatible with their environment. Of these, semi-fossoriality is possibly the most important, as it enables pouched mice to largely avoid unfavourable temperatures throughout the subregion. However, they cannot completely escape the effect of variation in nocturnal temperature or food availability and geographical differences in these factors may have resulted in ecotypic modifications of body size, NST, and torpor.

OPSOMMING

Die doel van die huidige studie was om die geografiese variasie in termoregulering en energiemetabolisme van die wangsakmuis *(Saccostomus campestris)* in suidelike Afrika te ondersoek. Hierdie spesies is wyd regdeur die substreek versprei, vertoon 'n breë habitatstoleransie en kom vanaf seevlak tot 1800 m voor in gebiede wat 'n gemiddelde jaarlikse reënval van tussen 100 mm en 1200 mm ondervind.

Veldwaarnemings in Suid-Afrika bevestig dat wangsakmuise alleenlopend, naglewend en half-fossoriaal is. Hierdie lewenstyl stel hulle in staat om die hitte van die dag te vermy, en hulle spandeer 'n groot deel van hul lewens in ondergrondse uitgrawings waar daar min daaglikse, seisoenale en geografiese variasie in temperatuur is. Alhoewel hulle koue temperature ervaar wanneer hulle snags aktief bo die grond is, verminder hul die tyd wat hul kos soek deur voedsel in hul wangsakke te versamel en dit na die nes saam te dra waar dit in relatiewe warmte en veiligheid geëet word. Nog fotoperiode, temperatuur of lokaliteit beïnvloed nesbou of kosstoorgedrag in gevangenskap, wat voorstel dat hierdie aktiwiteite nie op seisoenale of geografiese veranderinge in klimaat reageer nie.

Geografiese variasie in liggaamsgrootte was sterk met reënval korreleerd en wangsakmuise van lokaliteite met laer reënval was kleiner as die van gebiede wat meer reën ontvang. Hierdie korrelasie mag deur voedselbeperking in droë en halfwoestynagtige omgewings, waar primêre produksie en voedselbeskikbaarheid laag is, veroorsaak word. Hierdie verwantskap was egter ook opvallend in diere in gevangenskap wat toegang tot voedsel ad libitum gehad het. Aangesien kleiner individue minder voedsel as die wat groter is benodig, mag 'n afname in liggaamsgrootte 'n aanpassing verteenwoordig om die energiebehoeftes van wangsakmuise wat in droër gebiede waar voedselvoorrade beperk is woon, te verminder.

S.campestris het 'n laer as verwagte basale metaboliese tempo (BMT = minimum waargenome metaboliese tempo, MWMT) en 'n minimale termiese geleiding (G_m) wat gelykstaande of groter was as wat voorspel word vir diere van soortgelyke grootte. Hierdie eienskappe word beskou as hulp tot gat-lewende spesies om oorverhitting binne die beperkinge van 'n bedompige gat te vermy, alhoewel dit ook kontrasterende uitwerkinge op die omgewingsverenigbaarheid van wangsakmuise in suidelike Afrika sou hê; 'n lae BMT sou die energiebehoeftes van diere in hierdie droë/halfwoestynagtige gebied waar primêre produktiwiteit en voedelbeskikbaarheid laag is, help verminder. In teenstelling, 'n relatiewe hoë G_m sou hitteverlies in die hand werk en die koste van termoregulering in die koue verhoog, in besonder tydens tydperke van nagtelike aktiwiteit.

Na verstelling vir geografiese veranderlikheid in liggaamsmassa, was daar steeds betekenisvolle verskille in energiemetabolisme tussen wangsakmuise van nege lokaliteite in suidelike Afrika wat voorstel dat hierdie bevolkings fisiologies onderskeidend is. Daar was egter geen betekenisvolle verwantskappe tussen lokale klimaatskondisies en MWMT, G_m of liggaamstemperatuur nie wat drie eienskappe is wat die metabolisme van rustende diere bepaal. Dit doen aan die hand dat hierdie parameters nie tot klimaat aangepas is nie. Aan die ander kant, indien wangsakmuise min geografiese veranderlikheid in temperatuur ondervind wanneer hulle in hul gate rus, dan mag daar onvoldoende seleksiedruk vir ekotipiese aanpassings van hul rustende metabolisme wees.

Nieteenstaande hiervan sal wangsakmuise steeds aan sekere geografiese veranderlikheid in omgewingstemperatuur onderhewig wees terwyl aktief buite hul gate gedurende die nag. Dit mag verduidelik hoekom individue van koeler omgewings 'n hoër vermoë vir nie-bewingstermogenese (NBT) vertoon as diesulkes van warmer gebiede. NBT is 'n belangrike vorm van hitteproduksie in klein knaagdiere en sou wangsakmuise help om homeotermie gedurende nagtelike kos-soekery te handhaaf.

Wangsakmuise sou ook 'n afname in nagtelike temperature gedurende die winter beleef. Seisoenale verandering in temperatuur kan egter onvoorspelbaar wees en vele spesies gebruik 'n afname in fotoperiode om die aanvang van winter te voorsien, in besonder in gebiede waar omstandighede straf gedurende hierdie tyd van die jaar is. Om hierdie rede vertoon wangsakmuise van kouer, meer seisoenale gebiede 'n groter toename in NBT vermoë in reaksie op 'n afname in fotoperiode, alhoewel diere van regdeur suidelike Afrika 'n soortgelyke verbetering in hitteproduksie (MWMT en NBT) vertoon na akklimatisering aan koue.

Aangesien wangsakmuise beduidende energiebesparings gedurende rondes van spontane traagheid ophoop, sal die toename in die duur van traagheid wat by koeler lokaliteite waargeneem word hul in staat stel om die hoër koste van termoregulering in die gebiede te verminder. Die funksionele betekenis van spontane traagheid in hierdie spesies bly egter onseker aangesien dit slegs gedurende tydperke van rus by temperature van 15°C en laer plaasvind wat laer is as dit wat normaalweg in die gate van wangsakmuise aangetref word. Dit wil voorkom asof die verwantskap tussen lokaliteitstemperatuur en die duurte van traagheid vals is behalwe as dit aantoon dat wangsakmuise moeiliker omstandighede op 'n sekere stadium in hul evolusionêre geskiedenis ondervind het.

Die huidige studie toon duidelik aan dat wangsakmuise van suidelike Afrika 'n uiteenlopende slagorde van gedrags, morfologiese en fisiologiese meganismes vertoon wat hul energeties aanpasbaar aan hul omgewing maak. Hiervan is half-fossorialiteit moontlik die mees belangrike aangesien dit wangsakmuise in staat stel om grootliks ongustige temperature regdeur die substreek te vermy. Hulle kan egter nie heeltemal die uitwerking van veranderlikheid in nagtelike temperature of voedselbeskikbaarheid ontsnap nie, en geografiese verskille in hierdie faktore mag moontlik ekotipiese veranderinge in liggaamsgrootte, NBT en traagheid tot gevolg gehad het.

REFERENCES

ALCANTARA, M. 1991. Geographical variation in body size of the wood mouse *Apodemus sylvaticus* L. *Mammal Rev.* 21:143-150.

ALLABY, M. 1985. The Oxford dictionary of natural history. Oxford University Press, Oxford. 688pp.

ANDERSSON, M., & KREBS, J. 1978. On the evolution of hoarding behaviour. *Anim. Behav.* 26:707-711.

ANSELL, W.F.H. 1960. Mammals of Northern Rhodesia. Government Printer, Lusaka.

AR, A., 1987. Physiological adaptations to underground life in mammals. A case of mammalian neotony? In: Comparative physiology of environmental adaptations (ed.) Dejours, P., Vol. II, pp. 208-221, 8th ESCP Conference, Strasbourg 1986. Karger, Basel.

ARMITAGE, K.B., MELCHER, J.C. & WARD, J.M. 1990. Oxygen consumption and temperature regulation in yellow-bellied marmots from mesic and semi-arid environments. *J. Comp. Physiol. B* 160: 491-502.

ARNOLD, W., HELDMAIER, G., ORTMANN, S., POHL, H., RUF, T. & STEINLECHNER, S. 1991. Ambient temperatures in hibernacula and their energetic consequences for alpine marmots (*Marmota marmota*). *J. therm. Biol.* 16: 223-226.

BAKER, R.J. & BICKHAM, J.W. 1986. Speciation by monobrachial centric fusions. *Proc. Natl. Acad. Sci.* 83:8245-8248.

BARRY, W.J. 1976. Environmental effects on food hoarding in deermice (*Peromyscus*). *J. Mammal.* 57: 731-746.

BARTNESS, T.J. & WADE, G.N. 1984. Photoperiodic control of body weight and energy metabolism in Syrian hamsters (*Mesocricetus auratus*): Role of pineal gland, melatonin, gonads and diet. *Endocrinology* 114: 492-498.

BENNETT, N.C., JARVIS, J.U.M., & DAVIES, K.C. 1988. Daily and seasonal temperatures in the burrows of African rodent moles. *S. Afr. J. Zool.* 23: 189-195.

BERRY, R.J. 1985. The processes of pattern: genetical possibilities and constraints in co-evolution. *Oikos* 44:222-228.

BEST, T.L. 1982. Relationships of the burrows of Baja California kangaroo rats to ecogeographic and morphologic variation. *J. Mammal.* 63: 532-536.

BORUT, A., HAIM, A. & CASTEL, A. 1978. Non-shivering thermogenesis and implication of the thyroid in cold labile and cold resistant populations of the golden spiny mouse (*Acomys russatus*). In: Effectors of thermogenesis, (ed.s) Girardier, L. & Birkhauser, J., *Experientia Suppl.* 32: 219-227.

BOWERS, J.R. 1971. Resting metabolic rate in the cotton rat *Sigmodon*. *Physiol. Zool.* 44: 137-148.

BOYCE, M.S. 1978. Climatic variability and body size variation in the muskrats (*Ondatra zibethicus*) of North America. *Oecologia, Berlin* 36:1-19.

BOZINOVIC, F. & ROSENMANN, M. 1989. Maximum metabolic rate of rodents: physiological and ecological consequences on distributional limits. *Funct. Ecol.* 3: 173-181.

BRADLEY, S.R. & DEAVERS, D.R. 1980. A re-examination of the relationship between thermal conductance and body weight in mammals. *Comp. Biochem. Physiol.* 65A: 465-476.

BRADLEY, W.G., MILLER, J.S. & YOUSEF, M.K. 1974. Thermoregulatory patterns in pocket gophers: Desert and mountain. *Physiol. Zool.* 47: 172-179.

BRAIN, C.K. 1985. Temperature-induced environmental changes in Africa as evolutionary stimuli. In: Species and speciation, (ed.) Vrba, E.S., *Transvaal Mus. Monogr.* 4:45-52.

BRENNER, F.J. & LYLE, P.D. 1975. Effect of previous photoperiodic conditions and visual stimulation on food storage and hibernation in the eastern chipmunk (*Tamias striatus*). *Am. Midl. Nat.* 93: 227-234.

BRODY, S. 1945. Bioenergetics and growth. Reinhold Publ. Corp., New York.

BRONSON, F.H. 1987. Susceptibility of the fat reserves of mice to natural challenges. *J. Comp. Physiol. B* 157:551-554.

BRONSON, F.H., HEIDEMAN, P.D. & KERBESHIAN, M.C. 1991. Lability of fat stores in peripubertal wild house mice. *J. Comp. Physiol. B* 161:15-18.

BUFFENSTEIN, R. 1984. The importance of microhabitat in thermoregulation and thermal conductance in 2 Namib rodents - a crevice dweller, *Aethomys namaquensis*, and a burrow dweller, *Gerbillurus paeba*. *J. therm. Biol.* 9:235-242.

BUFFENSTEIN, R. 1985. The effect of starvation, food restriction and water deprivation on thermoregulation and average daily metabolic rates in *Gerbillus pusillus*. *Physiol. Zool.* 58: 320-328.

BUFFENSTEIN, R. & JARVIS, J.U.M. 1985a. The effect of water stress on growth and renal performance of juvenile Namib rodents. *J. Arid. Environ.* 9: 232-236.

BUFFENSTEIN, R. & JARVIS, J.U.M. 1985b. Thermoregulation and metabolism in the smallest African gerbil, *Gerbillus pusillus*. *J. Zool., Lond.* 205: 107-121.

BURNS, J.A., FLATH, D.L., & CLARK, T.W. 1989. On the structure and function of white-tailed prairie dog burrows. *Great Basin Nat.* 49: 517-524.

CAPUTA, M. & KAMARI, A. 1991a. Effect of warm rearing on temperature regulation in resting and exercising rats. *J. therm. Biol.* 16:357-361.

CAPUTA, M. & KAMARI, A. 1991b. Exercise performance of normothermic and hyperthermic rats: Effect of warm rearing. *J. therm. Biol.* 16:363-366.

CHAPPELL, M.A. 1985. Effects of ambient temperature and altitude on ventilation and gas exchange in deer mice (*Peromyscus maniculatus*). *J. Comp. Physiol. B* 155: 751-758.

CHOATE, T.S. 1972. Behavioural studies on some Rhodesian rodents. *Zool. Afr.* 7: 103-118.

CHEW, R.M., LINDBERG, R.G., & HAYDEN, P. 1967. Temperature regulation in the little pocket mouse, *Perognathus longimembris*. *Comp. Biochem. Physiol.* 21: 487-505.

COETZEE, C.G. 1970. The relative tail length of striped mice *Rhabdomys pumilio* Sparrman 1784 in relation to climate. *Zool. Afr.* 5: 1-6.

COPLEY, H. 1950. Small Mammals of Kenya. Highway Press, Nairobi.

DAUNCEY, M.J. & INGRAM, D.L. 1986. Acclimatization to warm or cold temperatures and the role of food intake. *J. therm. Biol.* 11: 89-93.

DE GRAAFF, G. 1981. The rodents of southern Africa. Butterworths, London.

DELANY, M.J. 1970. Variation and ecology of island populations of the long-tailed field mouse (*Apodemus sylvaticus* L.). *Symp. Zool. Soc. Lond.* 26:283-295.

DELANY, M.J. 1975. The rodents of Uganda. Trustees of the British Museum (Natural History), London.

DENYS, C. 1988. Apports de l'analyse morphométrique à la détermination des espèces actuelles et fossiles du genre *Saccostomus* (Cricetomyinae, Rodentia). *Mammalia* 5: 497-532.

DEPOCAS, F. & HART, J.S. 1957. Use of the Pauling oxygen analyzer for measurements of oxygen consumption of animals in open-cicuit system and in short-lag, closed-circuit apparatus. *J. Appl. Physiol.* 10: 388-392.

DENNIS, W. 1930. Rejection of wormy nuts by squirrels. *J. Mammal.* 11: 195-201.

DEWIT, C. 1972. An ecological study of a small mammal community with emphasis on the status of *Praomys* (*Mastomys*) *natalensis*. Unpublished M.Sc. thesis, University of Pretoria, Pretoria.

DIAMOND, J.M. 1992. The red flag of optimality. *Nature* 355:204-206.

DOI, K., & KUROSHIMA, A. 1982. Sexual difference in thermoregulatory ability of rats exposed to cold and heat. *J. therm. Biol.* 7: 99-105.

DOWNS, C.T. & PERRIN, M.R. 1989. An investigation of the macro- and micro-environments of four *Gerbillurus* species. *Cimbebasia* 11: 41-54.

DOWNS, C.T. & PERRIN, M.R. 1990a. Thermal parameters of four species of *Gerbillurus*. *J. therm. Biol.* 15: 291-300.

DOWNS, C.T. & PERRIN, M.R. 1990b. Field water-turnover rates of three *Gerbillurus* species. *J. Arid Environ.* 19: 199-208.

DUFOUR, B.A. 1978. Le terrier d'*Apodemus sylvaticus* L.: Sa construction en terrarium et son adaptation à des facteurs extremes et internes. *Behav. Proc.* 3: 57-76.

EARL, S.M.J.A. 1980. Development of behaviour in the pouched mouse *Saccostomus campestris*. Unpublished M.Sc. thesis, University of Pretoria, Pretoria.

EARL, Z. (S.M.J.A.) 1978. Postnatal development of *Saccostomus campestris*. *Afr. Small Mammal Newsletter* 2: 10-12.

EARL, Z. (S.M.J.A.) & NEL, J.A.J. 1976. Climbing behaviour in three African rodent species. *Zool. Afr.* 11: 181-190.

ELGAR, M.A. & HARVEY, P.H. 1987. Basal metabolic rates in mammals: allometry, phylogeny and ecology. *Funct. Ecol.* 1: 25-36.

ELLIOTT, L. 1978. Social behaviour and foraging ecology of the eastern chipmunk (*Tamias striatus*) in the Adirondack mountains. *Smithsonian Contrib. Zool.* 265: 1-107.

ELLISON, G.T.H. 1988. *Dorylus helvolus* (Formicidae: Dorylinae) preying on *Saccostomus campestris* (Rodentia: Cricetidae). *J. ent Soc. sth. Afr.* 51: 296.

ELLISON, G.T.H. 1990. A note on the small mammal fauna of Vaalkop Dam Nature Reserve. *Koedoe* 33: 114-116.

ELLISON, G.T.H. & SKINNER, J.D. 1990. Noradrenaline thermogenesis in conscious and anaesthetised pouched mice (*Saccostomus campestris*). *Comp. Biochem. Physiol.* 97A: 23-26.

ELLISON, G.T.H. & SKINNER, J.D. 1991a. Thermoregulation and torpor in African woodland dormice, *Graphiurus murinus*, following cold acclimation. *Z. Säugetierk.* 56: 41-47.

ELLISON, G.T.H & SKINNER, J.D. 1991b. Seasonal energy requirements and thermoregulation in growing pouched mice, *Saccostomus campestris* (Cricetidae). *Int. J. Biometeorol.* 35: 98-102.

ELLISON, G.T.H., SKINNER, J.D. In press. The influence of ambient temperature on spontaneous daily torpor of pouched mice (*Saccostomus campestris*: Rodentia - Cricetidae) from southern Africa. *J. therm. Biol.*

ELLISON, G.T.H. & WESTLIN-VAN AARDE, L.M. 1990. Ringtail in the pouched mouse (*Saccostomus campestris*). *Lab. Anim.* 24: 174-175.

ELLISON, G.T.H., BRONNER, G.N. & TAYLOR, P.J. In press. Is the annual cycle in body weight of pouched mice (*Saccostomus campestris*) the result of seasonal changes in adult size or population structure? *J. Zool., Lond.*

ELLISON, G.T.H., SKINNER, J.D. & FERGUSON, J.W.H. In prep. Interactive effects of photoperiod and temperature on daily activity and metabolism of pouched mice (*Saccostomus campestris*: Cricetidae).

FAHRI, L.E. 1987. Exposure to stressful environments. In: Comparative physiology of environmental adaptations (ed.) Dejours, P., Vol. II, pp. 1-14, 8th ESCP Conference, Strasbourg 1986. Karger, Basel.

FEIST, D.D. & MORRISON, P.R. 1981. Seasonal changes in metabolic capacity and norepinephrine thermogenesis in the Alaskan red-backed vole: environmental cues and annual differences. *Comp. Biochem. Physiol.* 69A: 697-700

FEIST, D.D. & FEIST, C.F. 1986. Effects of cold, short day and melatonin on thermogenesis, body weight and reproductive organs in Alaskan red-backed voles, *Clethrionomys rutilus dawsonii*. *J. Comp. Physiol.* 156: 741-746.

FERREIRA, S.M. 1990. Geographical variation in the pouched mouse, *Saccostomus campestris* (Rodentia, Cricetidae). Unpublished B.Sc. Honours Report, University of Pretoria, Pretoria.

FITZSIMMONS, F.W. 1920. The natural history of South Africa. Longmans, London.

FORMANOWICZ, D.R., BRADLEY, P.J. & BRODIE, E.D. 1989. Food hoarding by the least shrew (*Cryptotis parva*): Intersexual and prey type effects. *Am. Midl. Nat.* 122: 26-33

FULLER, W.A., STEBBINS, L.L. & DYKE, G.R. 1969. Overwintering of small mammals near Great Slave Lake, Northern Canada. *Arctic* 22: 34-55

GEISER, F. 1988. Reduction of metabolism during hibernation and daily torpor in mammals and birds: temperature effect or physiological inhibition? *J. Comp. Physiol. B* 158: 25-37.

GETTINGER, R.D. & RALPH, C.L. 1985. Thermoregulatory responses to photoperiod by kangaroo rats (*Dipodomys ordii*): influence of night lighting on non-shivering thermogenesis and resting metabolism. *J. Exp. Zool.* 234: 335-340.

GOERTZ, J.W., DAWSON, R.M. & MOWBRAY, E.E. 1975. Response to nest boxes and reproduction by *Glaucomys volans* in northern Louisiana. *J. Mammal.* 56: 933-939.

GORDON, C.J., FEHLER, K.S. & LONG, M.D. 1986. Relationship between autonomic and behavioural thermoregulation in the golden hamster. *Am. J. Physiol.* 251: R320-R324.

GORDON, D.H. 1986. Extensive chromosomal variation in the pouched mouse *Saccostomus campestris* (Rodentia, Cricetidae) from southern Africa: A preliminary investigation of evolutionary status. *Cimbebasia* 8: 37-47.

GORDON, S. 1972. Animal physiology: principles and adaptations. Macmillan, London.

GOULD, S.J. & JOHNSTON, R.F. 1972. Geographic variation. *Ann. Rev. Ecol. Syst.* 3:457-498.

GOULD, S.J. & LEWONTIN, R.C. 1979. The spandrels of San Marco and the Panglossian paradigm: a critique of the adaptionists' programme. *Proc. R. Soc. Lond. B* 205: 581-598.

GRODZINSKI, W., KLEKOWSKI, R.Z. & DUNCAN, A. 1975. Methods for ecological energetics. International Biological Programme Handbook no. 24., Blackwell Scientific, Oxford.

HAIM, A., 1982. Effects of long scotophase and cold acclimation on heat production in two diurnal rodents. *J. Comp. Physiol.* 148: 77-81.

HAIM, A. 1984. Adaptive variations in heat production within gerbils (genus *Gerbillus*) from different habitats. *Oecologia* 61: 49-52.

HAIM, A. & BORUT, A. 1976. Thermoregulation and nonshivering thermogenesis as factors limiting distribution of the golden spiny mouse (*Acomys russatus*). *Isr. J. Med. Sci.* 12: 896.

HAIM, A. & FOURIE, F. le R. 1980. Long scotophase increases heat production in *Rhabdomys pumilio* and *Praomys natalensis* (Rodentia). *S. Afr. J. Sci.* 76: 89.

HAIM, A. & YAHAV, S. 1982. Non-shivering thermogenesis in winter-acclimated and in long-scotophase and cold-acclimated *Apodemus mystacinus* (Rodentia). *J. therm. Biol.* 7: 193-195.

HAIM, A. & FAIRALL, N. 1986. Geographical variations in heat production and dissipation within two populations of *Rhabdomys pumilio* (Muridae). *Comp. Biochem. Physiol.* 84A: 111-112.

HAIM, A., HETH, G. & NEVO, E. 1985. Adaptive thermoregulatory patterns in speciating mole rats. *Acta Zool. Fenn.* 170: 137-140.

HAIM, A., SKINNER, J.D. & ROBINSON, T.J. 1987. Bioenergetics, thermoregulation and urine analysis of squirrels of the genus *Xerus* from an arid environment. *S. Afr. J. Zool.* 22: 45-49.

HAIM, A., ELLISON, G.T.H. & SKINNER, J.D 1988. Thermoregulatory circadian rhythms in the pouched mouse (*Saccostomus campestris*). *Comp. Biochem. Physiol.* 91A: 123-127.

HAIM, A., HETH, G., AVNON, Z. & NEVO, E. 1984. Adaptive physiological variation in non-shivering thermogenesis and its significance in speciation. *J. Comp. Physiol. B* 154: 145-147.

HAIM, A., RACEY, P.A., SPEAKMAN, J.R., ELLISON, G.T.H. & SKINNER, J.D. 1991. Seasonal acclimatization and thermoregulation in the pouched mouse *Saccostomus campestris*. *J. therm. Biol.* 16: 13-17.

HANNEY, P. 1965. The Muridae of Malawi. *J. Zool., Lond.* 146: 577-633.

HART, J.S. 1956. Seasonal changes in insulation of the fur. *Can. J. Zool.* 34: 53-57.

HART, J.S. 1971. Rodents. In: Comparative physiology of thermoregulation, (ed.) Causey Whittow, G., Vol. II, Mammals, pp. 1-149, Academic Press, London.

HARTUNG, T.G., & DEWSBURY, D.A. 1979. Nest-building behaviour in seven species of muroid rodents. *Behav. Neural. Biol.* 27: 532-539.

HAYSSEN, V. & LACY, R.C. 1985. Basal metabolic rates in mammals: taxonomic differences in the allometry of BMR and body mass. *Comp. Biochem. Physiol.* 81A: 741-745.

HAYWARD, J.S. 1965a. The gross body composition of six geographical races of *Peromyscus*. *Can. J. Zool.* 43:297-308.

HAYWARD, J.S. 1965b. Metabolic rate and its temperature-adaptive significance in six geographic races of *Peromyscus*. *Can. J. Zool.* 43: 309-323.

HAYWARD, J.S. 1965c. Microclimate temperature and its adaptive significance in six geographic races of *Peromyscus*. *Can. J. Zool.* 43: 341-350.

HEATH, H.W. & LYNCH, G.R. 1982. Intraspecific differences for melatonin-induced reproductive regression and the seasonal molt in *Peromyscus leucopus*. *Gen. Comp. Endocrinol.* 48: 289-295.

HEATH, H.W. & LYNCH, G.R. 1983. Intraspecific differences in the use of photoperiod and temperature as environmental cues in white-footed mice *Peromyscus leucopus*. *Physiol. Zool.* 56: 506-512.

HELDMAIER, G. 1971. Zitterfreie Wärmebildung und korpergröße bei Säugetieren. *Z. Vergl. Physiol.* 73: 222-248.

HELDMAIER, G. 1975. Metabolic and thermoregulatory responses to heat and cold in the Djungarian hamster, *Phodopus sungorus*. *J. Comp. Physiol.* 102: 115-122.

HELDMAIER, G., & STEINLECHNER, S. 1981. Seasonal control of energy requirements for thermoregulation in the Djungarian hamster (*Phodopus sungorus*), living under natural photoperiod. *J. Comp. Physiol.* 142: 429-437.

HELDMAIER, G., STEINLECHNER, S., & RAPHAEL, J. 1982. Nonshivering thermogenesis and cold resistance during seasonal acclimation in the Djungarian hamster. *J. Comp. Physiol.* 149: 1-9.

HELDMAIER, G., BÖCKLER, H., BUCHBERGER A., LYNCH, G.R., PUCHALSKI, W., STEINLECHNER, S. & WIESINGER, H. 1985. Seasonal acclimation and thermogenesis. In: Circulation, respiration and metabolism, (ed.) Gilles, R., pp. 431-501, Springer, Berlin

HELDMAIER, G., STEINLECHNER, S., RUF, T., WIESINGER, H. & KLINGENSPOR, M. 1989. Photoperiod and thermoregulation in vertebrates: body temperature rhythms and thermogenic acclimation. *J. Biol. Rhythms* 4: 251-265.

HELDMAIER, G., KLAUS, S. & WIESINGER, H. 1990. Seasonal adaptation of thermoregulatory heat production in small mammals. In: Thermoreception and temperature regulation, (ed.s) Bligh, J. & Voigt, K., pp. 235-243, Springer-Verlag, Berlin.

HERBERG, L.J., PYE, J. & BLUNDELL, J.E. 1972. Sex differences in the hypothalamic regulation of food hoarding: hormones versus calories. *Anim. Behav.* 20: 186-191.

HERREID, C.F., & KESSEL, B. 1967. Thermal conductance in birds and mammals. *Comp. Biochem. Physiol.* 21: 405-414.

HICKMAN, G.C. & MACHINÉ, C. 1986. Swimming behaviour in six species of African rodents (Cricetidae, Muridae). *Acta Theriol.* 31: 449-466.

HILL, R.W. 1972. Determination of oxygen consumption by use of the paramagnetic oxygen analyzer. *J. appl. Physiol.* 33: 261-263.

HILL, R.W. 1975. Daily torpor in *Peromyscus leucopus* on an adequate diet. *Comp. Biochem. Physiol.* 51A: 413-423.

HILL, R.W. 1983. Thermal physiology and energetics of *Peromyscus*; ontogeny, body temperature, metabolism, insulation and microclimatology. *J. Mammal.* 64: 19-37.

HORTON, J.S. & WRIGHT, J.T. 1944. The wood rat as an ecological factor in southern Californian watersheds. *Ecology* 25: 341-351.

HUDSON, J.W. 1978. Shallow daily torpor. In; Strategies in cold, (ed.s) Wang, L.C.H. & Hudson, J.W., pp. 67-108, Academic press, New York.

HUDSON, J.W. & DEAVERS, D.R. 1973. Metabolism, pulmo-cutaneous water loss and respiration of eight species of ground squirrels from different environments. *Comp. Biochem. Physiol.* 45A: 69-100.

IVERSON, S.L. & TURNER, B.N. 1974. Winter weight dynamics in *Microtus pennsylvanicus*. *Ecology* 55: 1030-1041.

JACOBSEN, N.H.G. 1977. An annotated checklist of the amphibians, reptiles and mammals of the Nylsvley Nature Reserve. *S. Afr. Nat. Sci. Prog. Rep.* 21: 1-65.

JAMES, F.C. 1970. Geographic size variation in birds and its relationship with climate. *Ecology* 51: 365-390.

JANSKY, L. 1973. Non-shivering thermogenesis and its thermoregulatory significance. *Biol. Rev.* 48: 85-132.

KERLEY, G.I.H. 1989. Diet of small mammals from the karoo, South Africa. *S. Afr. J. Wildl. Res.* 19: 67-72.

KERN, N.G. 1981. The influence of fire on populations of small mammals of the Kruger National Park. *Koedoe* 24: 125-157.

KING, M. 1985. The canalization model of chromosomal evolution: a critique. *Syst. Zool.* 34:69-74.

KING, J.A., MAAS, D. & WEISMAN, R.G. 1964. Geographic variation in nest size among species of *Peromyscus*. *Evolution* 18: 230-234.

KINGDON, J. 1984. East African mammals, Vol. IIB, hares and rodents, University of Chicago Press, Chicago.

KLAUS, S., HELDMAIER, G. & RICQUIER, D. 1988. Seasonal acclimation of bank voles and wood mice: nonshivering thermogenesis and thermogenetic properties of brown adipose tissue mitochondria. *J. Comp. Physiol.* 158: 157-164.

KLEIBER, M. 1947. Body size and metabolic rate. *Physiol. Rev.* 27: 511-541.

KORN, H. 1989. The annual cycle in body weight of small mammals from the Transvaal, South Africa, as an adaptation to a subtropical seasonal environment. *J. Zool., Lond.* 218: 223-231.

KOTEJA, P. 1987. On the relation between basal and maximum metabolic rate in mammals. *Comp. Biochem. Physiol.* 87A: 205-208.

KREBS, C.J. 1978. Ecology: The experimental analysis of distribution and abundance. 2nd edn, Harper and Row, New York.

KREBS, J.R. & McCLEERY, R.H. 1984. Optimization in behavioural ecology. In: Behavioural ecology, an evolutionary approach, (ed.) Krebs, J.R. & Davies, N.B., 2nd edn, Ch. 4, Blackwell Scientific, Oxford.

LEMEN, C.A. & FREEMAN, P.W. 1985. Tracking mammals with fluorescent pigments: a new technique. *J. Mammal.* 66: 134-136.

LEVENSON, H. 1990. Sexual size dimorphism in chipmunks. *J. Mammal.* 71: 161-170.

LOUW, G.N. & SEELY, M.K. 1982. Ecology of desert organisms. Longman, Harlow.

LOVEGROVE, B.G. 1986. Thermoregulation of the subterranean rodent genus *Bathyergus*, Bathyergidae. *S. Afr. J. Zool.* 21: 283-288.

LYMAN, C.P. 1954. Activity, food consumption and hoarding in hibernators. *J. Mammal.* 35: 545-552 .

LYMAN, C.P., O'BRIEN, R.C. & BOSSERT, W.H. 1983. Differences in tendency to hibernate among groups of Turkish hamsters (*Mesocricetus brandti*). *J. therm. Biol.* 8: 255-257.

LYNCH, C.D. 1983. The mammals of the Orange Free State. *Mem. nas. Mus. Bloemfontein* 18: 1-218.

LYNCH, G.R. 1973. Seasonal changes in thermogenesis, organ weights, and body composition in the white-footed mouse, *Peromyscus leucopus*. *Oecologia* 13: 363-376.

LYNCH, G.R., & GENDLER, S.L. 1980. Multiple responses to different photoperiods occur in the mouse, *Peromyscus leucopus*. *Oecologia* 45: 318-321.

LYNCH, G.R., LYNCH, C.B., DUBE, M. & ALLEN, C. 1976. Early cold exposure: Effects on behavioural and physiological thermoregulation in the house mouse, *Mus musculus*. *Physiol. Zool.* 49:191-199.

LYNCH, G.R., WHITE, S.E., GRUNDEL, R. & BERGER, M.S. 1978. Effects of photoperiod, melatonin administration and thyroid block on spontaneous daily torpor and temperature regulation in the white-footed mouse, *Peromyscus leucopus*. *J. Comp. Physiol. B* 125: 157-163.

LYNCH, G.R., HEATH, H.W. & JOHNSTON, C.M. 1981. Effect of geographic origin on the photoperiodic control of reproduction in the white-footed mouse, *Peromyscus leucopus*. *Biol. Reprod.* 25: 475-480.

MACLEAN, G.S., 1981. Factors influencing the composition of respiratory gases in mammal burrows. *Comp. Biochem. Physiol.* 69A: 373-380.

MANN, K.H. 1967. The approach through the ecosystem. In: The teaching of ecology, (ed.) Lambert, J.M., pp. 103-111, Blackwell Scientific, Oxford.

MASON, E.B. 1974. Metabolic responses of two species of *Peromyscus* raised in different thermal environments. *Physiol. Zool.* 47: 68-74.

MASUDA, A., & OISHI, T. 1988. Effects of photoperiod and temperature on body weight, food intake, food storage, and pelage colour in the Djungarian hamster, *Phodopus sungorus*. *J. Exp. Zool.* 248: 133-139.

MATTHEY, R. 1958. Les chromosomes et la position systematique de quelques Murinae Africains (Mammalia - Rodentia). *Acta Trop.* 15: 7-117.

McARTHUR, A.J. & CLARK J.A. 1988. Body temperature and the conservation of energy and water. *J. therm. Biol.* 13:9-13.

McNAB, B.K. 1971. On the ecological significance of Bergmann's rule. *Ecology* 52:845-854.

McNAB, B.K. 1979a. Climatic adaptation in the energetics of Heteromyid rodents. *Comp. Biochem. Physiol.* 62A: 813-820.

McNAB, B.K. 1979b. The influence of body size on the energetics and distribution of fossorial and burrowing mammals. *Ecology* 60: 1010-1021.

McNAB, B.K. 1980a. Food habits, energetics and population biology of mammals. *Am. Nat.* 116: 106-124.

McNAB, B.K. 1980b. On estimating thermal conductance in endotherms. *Physiol. Zool.* 53: 145-156.

McNAB, B.K. 1983. Energetics, body size and the limits to endothermy. *J. Zool., Lond.* 199: 1-29.

McNAB, B.K. 1984. Physiological convergence among ant-eating and termite-eating mammals. *J. Zool., Lond.* 203: 485-510.

McNAB, B.K. 1986a. The influence of food habits on the energetics of eutherian mammals. *Ecol. Monogr.* 56: 1-19.

McNAB, B.K. & MORRISON, P.R. 1963. Body temperature and metabolism in subspecies of *Peromyscus* from arid and mesic environments. *Ecol. Monogr.* 33: 63-82.

MILLER, G.A. & VIEK, P. 1944. An analysis of the rat's responses to unfamiliar aspects of the hoarding situation. *J. Comp. Psychol.* 37: 221-231.

MORENO, S. & DELIBES, M. 1982. Notes on the garden dormouse (*Eliomys*; Rodentia, gliridae) of northern Morocco. *Säugetierkunde Mitt.* 30: 212-215.

MORRISON, P. 1960. Some interrelations between weight and hibernation function. *Bull. Mus. Comp. Zool.* 124:75-91.

MURALIDHARA, D.V. & SHETTY, P.S. 1986. Effects of preweaning nutritional deprivation on basal metabolism and thermoregulatory thermogenesis in the rat. *Br. J. Nutr.* 56:615-624.

NEVO, E. & AMIR, E. 1964. Geographic variation in reproduction and hibernation patterns of the forest dormouse. *J. Mammal.* 45: 69-87.

NEVO, E. & SHKOLNIK, A. 1974. Adaptive metabolic variation of chromosomal forms in mole rats, *Spalax*. *Experientia* 30: 724-726.

NEVO, E., BEILES, A. & BEN-SHLOMO, R. 1984. The evolutionary significance of genetic diversity: Ecological, demographic and life history correlates. In: Evolutionary dynamics of genetic diversity, (ed.) Mani, S.S., *Lecture notes in Biomathematics* 53:13-213.

NEVO, E., BEILES, A., HETH, G. & SIMSEN, S. 1986. Adaptive differentiation of body size in speciating mole rats. *Oecologia* 69: 327-333.

NYBY, J., WALLACE, P., OWEN, K. & THIESSEN, D.D. 1973. An influence of hormones on hoarding behaviour in the Mongolian gerbil (*Meriones unguiculatus*). *Horm. Behav.* 4: 283-288.

OGLE, C. 1934. Climatic influences on the growth of the male albino mouse. *Amer. J. Physiol.* 107:635-640.

PERRIN, M.R. & KOKKIN, M.J. 1986. Comparative gastric anatomy of *Cricetomys gambianus* and *Saccostomus campestris* (Cricetomyinae) in relation to *Mystromys albicaudatus* (Cricetinae). *S. Afr. J. Zool.* 21: 202-210.

PETTERBORG, L.J. 1978. Effect of photoperiod on body weight in the vole, *Microtus montanus*. *Can. J. Zool.* 56: 431-435.

PETTIFER, H.L., & NEL, J.A.J. 1977. Hoarding in four southern African rodent species. *Zool. Afr.* 12:409-418.

PIENAAR, U. de V., RAUTENBACH, I.L., & DE GRAAF, G., 1980. The small mammals of the Kruger National Park. National Parks Board of Trustees, Pretoria.

POHL, H. 1965. Temperature regulation and cold acclimation in the golden hamster. *J. appl. Physiol.* 20: 405-410.

PRESCOTT-ALLEN, R. 1991. Caring for the Earth: a strategy for sustainable living. IUCN - UNEP - WWF, SADAG, Bellegarde - Valserine. 228 pp.

PROTHERO, J., & JURGENS, K.D. 1986. An energetic model of daily torpor in endotherms. *J. theor. Biol.* 121: 403-415.

RAFAEL, J. & VSIANSKY, P. 1985. Photoperiodic control of thermogenic capacity in brown adipose tissue of the Djungarian hamster. *J. therm. Biol.* 10: 167-170.

RAJENDRAM, E.A., BRAIN, P.F., PARMIGIANI, S. & MAINARDI, M. 1987. Effects of ambient temperature on nest construction in four species of laboratory rodents. *Boll. Zool.* 54: 75-81.

RANDALL, J.A. 1984. Territorial defense and advertisement by foot-drumming in bannertail kangaroo rats (*Dipodomys spectabilis*) at high and low population densities. *Behav. Ecol. Sociobiol.* 16: 11-20.

RAUTENBACH, I.L. 1978. A numerical re-appraisal of the southern African biotic zones. *Bull. Carnegie Mus. Nat Hist.* 6: 175-187.

RAUTENBACH, I.L. 1982. Mammals of the Transvaal. *Ecoplan Monog.* 1: 1-211.

REICHMAN, O.J. & FAY, P. 1983. Comparison of the diets of a caching and a non-caching rodent. *Amer. Nat.* 122: 576-581.

ROBERTS, J.C., HOCK, R.J. & SMITH, R.E.M. 1969. Effects of altitude on brown fat and metabolism of the deer mouse *Peromyscus*. Fed. Proc. 28: 1065-1072.

RUF, T. & HELDMAIER, G. 1987. Computerized body temperature telemetry in small animals: use of simple equipment and advanced noise suppression. *Comput. Biol. Med.* 17:331-340.

SCHECK, S.H. 1982. A comparison of thermoregulation and evaporative water loss in hispid cotton rat, *Sigmodon hispidus texianus*, from northern Kansas and south-central Texas. *Ecology* 63: 361-369.

SCHMIDT-NIELSEN, K. 1986. Physiological adaptation to the environment. In: Endocrine regulations as adaptive mechanisms to the environment, (ed.s) Assenmacher, I. & Boissin, J., pp. 13-17, Colloque International Centre d'études Biologiques des animaux sauvages, Editions du Centre National de la Recherche Scientifique, Paris.

SCHOFFENIELS, E. 1986. Adaptation and finality, or a boeing is not a submarine. In: Endocrine regulations as adaptive mechanisms to the environment, (ed.s) Assenmacher, I. & Boissin, J., pp. 19-27, Colloque International Centre d'études Biologiques des animaux sauvages, Editions du Centre National de la Recherche Scientifique, Paris.

SCHOLANDER, P.F. 1955. Evolution of climatic adaptation in homeotherms. *Evolution* 9: 15-26.

SCHOLANDER, P.F., WALTERS, V., HOCK, R. & IRVING, L. 1950a. Body insulation of arctic and tropical mammals and birds. *Biol. Bull.* 99: 237-258.

SCHOLANDER, P.F., HOCK, R., WALTERS, V. & IRVING, L. 1950b. Adaptation to cold in arctic and tropical mammals and birds in relation to body temperature, insulation and basal metabolic rate. *Biol. Bull.* 99: 259-271.

SCLATER, W.L. 1901. The mammals of South Africa. R.H. Porter, London. 241 pp.

SEALANDER, J.A. 1952. The relationship of nest protection and huddling to survival of *Peromyscus* at low temperatures. *Ecology* 33: 63-71.

SEARCY, W.A. 1980. Optimum body sizes at different temperatures: An energetics explanation of Bergmann's rule. *J. theor. Biol.* 83:579-594.

SHKOLNIK, A. & BORUT, A. 1969. Temperature and water relations in two species of spiny mice (*Acomys*). *J. Mammal.* 50: 245-255.

SHORTRIDGE, G.C. 1934. The mammals of South West Africa. Heinemann, London.

SIMON, E. 1987. Glossary of terms for thermal physiology. *Pfl. Arch. Europ. J. Physiol.* 410: 567-587.

SITES, J.W. & MORITZ, C. 1987. Chromosomal evolution and speciation revisited. *Syst. Zool.* 36:153-174.

SKINNER, J.D. & SMITHERS, R.H.N. 1990. The mammals of the Southern African subregion. University of Pretoria, Pretoria.

SMITH, C.C. & REICHMAN, O.J. 1984. The evolution of food caching by birds and mammals. *Ann. Rev. Ecol. Syst.* 15: 329-351.

SMITH, M.C. 1968. Red squirrel responses to spruce cone failure in interior Alaska. *J. Wildl. Mgmt.* 32: 305-317.

SMITHERS, R.H.N. 1971. Mammals of Botswana. *Mus. mem. Nat. Mus. Monum. Rhod.* 4: 1-340.

SOKAL, R.R. & ROHLF, F.J. 1981. Biometry. W.H. Freeman, San Francisco.

STARK, H.E. 1963. Nesting habits of the California vole, *Microtus californicus* and microclimatic factors affecting its nests. *Ecology* 44: 663-669.

STEINLECHNER, S., HELDMAIER, G. & BÖCKLER, H. 1983. The seasonal cycle of body weight in the Djungarian hamster: photoperiodic control and the influence of starvation and melatonin. *Oecologia* 60: 401-405.

STEINLECHNER, S. & HELDMAIER, G., 1982. Role of photoperiod and melatonin in seasonal acclimatization of the Djungarian hamster, *Phodopus sungorus*. *Int. J. Biometeorol.* 26: 329-337.

STUTTERHEIM, C.J. & SKINNER, J.D. 1973. Preliminary notes on the behaviour and breeding of *Gerbillurus paeba paeba* (A Smith 1834) in captivity. *Koedoe* 16:127-148.

SUMNER, F.B. 1909. Some effects of external conditions in the white mouse. *J. Exp. Zool.* 7: 97-155.

SWANEPOEL, P. 1972. The population dynamics of rodents at Pongola, Northern Zululand, exposed to dieldrin spraying. Unpublished M.Sc. thesis, University of Pretoria, Pretoria.

TANNENBAUM, M.G. & PIVORUN, E.B. 1984. Differences in daily torpor patterns among three southeastern species of *Peromyscus*. *J. Comp. Physiol. B* 154: 233-236.

TANNENBAUM, M.G. & PIVORUN, E.B. 1987a. Variation in hoarding behaviour in southeastern *Peromyscus*. *Anim. Behav.* 35: 297-299.

TANNENBAUM, M.G. & PIVORUN, E.B. 1987b. Differential effect of food restriction on the induction of daily torpor in *Peromyscus maniculatus* and *P.leucopus*. *J. therm. Biol.* 12: 159-162.

TANNENBAUM, M.G. & PIVORUN, E.B. 1988. Seasonal study of daily torpor in southeastern *Peromyscus maniculatus* and *Peromyscus leucopus* from mountains and foothills. *Physiol. Zool.* 61: 10-16.

THOMAS, K.R. 1974. Burrow systems of the eastern chipmunk (*Tamias striatus pipilans* Lowery) in Louisiana. *J. Mammal.* 55: 454-459.

THOMPSON, S.D. 1985. Subspecific differences in metabolism, thermoregulation, and torpor in the western harvest mouse, *Reithrodontomys megalotis*. *Physiol. Zool.* 58: 430-444.

THORINGTON, R.W. 1970. Lability of tail length of the white-footed mouse, *Peromyscus leucopus noveboracensis*. *J. Mammal.* 51: 52-59.

TUCKER, V.A. 1965. The relation between the torpor cycle and heat exchange in the California pocket mouse, *Perognathus californicus*. *J. Cell. Comp. Physiol.* 65: 405-414.

TWENTE, J.W. & TWENTE J.A. 1965. Effects of core temperature upon duration of hibernation of *Citellus lateralis*. *J. appl. Physiol.* 20: 411-416.

TYSON, P.D. 1986. Climatic change and variability in southern Africa. Oxford University Press, Cape Town. 128 pp.

USDA Soil Conservation Service, 1975. Soil taxonomy (A basic system of soil classification for making and interpreting soil surveys). Wiley, New York.

VAN CHI-BONNARDEL, R. 1973. The atlas of Africa. Éditions jeune Afrique, Paris.

VANDER WALL, S.B. 1990. Food hoarding in animals. University Press, Chicago.

VESEY-FITZGERALD, D.F. 1966. The habits and habitats of small rodents in the Congo river catchment region of Zambia and Tanzania. *Zool. Afr.* 2: 111-122.

VILJOEN, S. 1985. Comparative thermoregulatory adaptations of southern African tree squirrels from four different habitats. *S. Afr. J. Zool.* 20: 28-32.

VOGT, F.D. & LYNCH, G.R. 1982. Influence of ambient temperature, nest availability, huddling and daily torpor on energy expenditure in the white-footed mouse *Peromyscus leucopus*. *Physiol. Zool.* 55: 56-63.

WADE, G.N. & BARTNESS, T.J. 1984. Effects of photoperiod and gonadectomy on food intake, body weight, and body composition in Siberian hamsters. *Am. J. Physiol.* 246: R26-R30.

WALKER, E.P. 1964. Mammals of the World. John Hopkins Press, Baltimore.

WARD, J.M. & ARMITAGE, K.B. 1981. Circannual rhythms of food consumption, body mass and metabolism in yellow-bellied marmots. *Comp. Biochem. Physiol.* 69A: 621-626.

WATSON, C.R.B. 1987. The comparative ecology of two small mammal communities in the Kruger National Park. Unpublished M.Sc. thesis, University of Pretoria, Pretoria.

WEINER, J. 1989. Metabolic constraints to mammalian energy budgets. *Acta Theriol.* 34: 3-35.

WEINER, J. & HELDMAIER, G. 1987. Metabolism and thermoregulation in two races of Djungarian hamsters: *Phodopus sungorus sungorus* and *P.s.campbelli*. *Comp. Biochem. Physiol.* 86A: 639-642.

WESTLIN, L.M. & ELLISON, G.T.H. In prep. Reproductive flexibility in female pouched mice (*Saccostomus campestris*: Cricetomyinae).

WESTLIN-VAN AARDE, L.M. 1988. Reproduction in a laboratory colony of the pouched mouse, *Saccostomus campestris*. *J. Reprod. Fert.* 83: 773-778.

WESTLIN-VAN AARDE, L.M. 1989a. Pre- and post-natal development of pouched mice, *Saccostomus campestris*. *J. Zool., Lond.* 218: 497-501.

WESTLIN-VAN AARDE, L.M. 1989b. Social environment and reproduction in female pouched mice, *Saccostomus campestris*. *J. Reprod. Fert.* 86: 367-372.

WESTLIN-VAN AARDE, L.M. 1989c. Pregnancy, lactation and the oestrous cycle of the pouched mouse, *Saccostomus campestris*. *J. Reprod. Fert.* 87: 155-162.

WILLAN, K. 1990. Reproductive biology of the southern African ice rat. *Acta Theriol.* 35:39-51.

WITHERS, P.C. 1977. Measurement of VO_2, VCO_2, and evaporative water loss with a flow-through mask. *J. appl. Physiol.* 42: 120-123.

WOLFE, J.L. 1970. Experiments on nest-building in *Peromyscus*. *Anim. Behav.* 18: 613-615.

WONG, R. & JONES, C.H. 1985. A comparative analysis of feeding and hoarding in hamsters and gerbils. *Behav. Proc.* 11: 301-308.

WUNDER, B.A. 1974. The effect of activity on body temperature of Ord's kangaroo rat (*Dipodomys ordii*). *Physiol. Zool.* 47: 29-36.

WUNDER, B.A. 1978. Implications of a conceptual model for the allocation of energy resources by small mammals. In: Populations of small mammals under natural conditions (ed.) Snyder, D., Pymatuning Laboratory of Ecology, *Univ. Pittsburgh Spec. Publ. Ser.* 5: 68-75.

WUNDER, B.A. 1984. Strategies for, and environmental cueing mechanisms of, changes in thermoregulatory parameters of small mammals. In: Winter ecology of small mammals, (ed.) Merritt, J.F., *Carnegie Mus. Nat. Hist. Spec. Publ.* 10: 1-392.

WUNDER, B.A. 1985. Energetics and thermoregulation. In: Biology of New World *Microtus*. *Amer. Soc. Mammal. Spec. Publ.* 8: 812-844.

WUNDER, B.A., DOBKIN, D.S. & GETTINGER, R.D. 1977. Shifts of thermogenesis in the prairie vole (*Microtus ochrogaster*). *Oecologia* 29: 11-26.

YAHAV, S., SIMSON, S. & NEVO, E. 1988. Adaptive energy metabolism in four chromosomal species of subterranean mole rats. *Oecologia* 77: 533-536.

YAHAV, S., SIMSON, S. & NEVO, E. 1989. Total body water and adaptive water turnover rate in four chromosomal species of subterranean mole rats of the *Spalax ehrenbergi* superspecies in Israel. *J. Zool., Lond.* 218: 461-469.

YELLON, S.M. & GOLDMAN, B.D. 1984. Photoperiod control of reproductive development in the male Djungarian hamster (*Phodopus sungorus*). *Endocrinology* 114: 664-670.

YOM-TOV, Y. & NIX, H. 1986. Climatological correlates for body size of five species of Australian mammals. *Biol. J. Linn. Soc.* 29: 245-26.

YOUSEF, M.K., JOHNSON, H.D., BRADLEY, W.G. & SEIF, S.M. 1974. Tritiated water-turnover rate in rodents: desert and mountain. *Physiol. Zool.* 47: 153-162.

APPENDIX I: PUBLISHED AND ACCEPTED MANUSCRIPTS CITED IN THE THESIS

ELLISON, G.T.H. 1988. *Dorylus helvolus* L. (Formicidae: Dorylinae) preying on *Saccostomus campestris* (Rodentia: Cricetidae). *J. ent. Soc. sth. Afr.* 51: 296.

ELLISON, G.T.H. 1990. A note on the small mammal fauna of Vaalkop Dam nature reserve. *Koedoe* 33: 114-116.

ELLISON, G.T.H. & SKINNER, J.D. 1990. Noradrenaline thermogenesis in conscious and anaesthetised pouched mice (*Saccostomus campestris*). *Comp. Biochem. Physiol.* 97A: 23-26.

ELLISON, G.T.H. & SKINNER, J.D. 1991. Seasonal energy requirements and thermoregulation of growing pouched mice, *Saccostomus campestris* (Cricetidae). *Int. J. Biometeorol.* 35: 98-102.

ELLISON, G.T.H. & SKINNER, J.D. *in press*. The influence of ambient temperature on spontaneous daily torpor in pouched mice (*Saccostomus campestris*: Rodentia - Cricetidae) from southern Africa. *J. therm. Biol.*

ELLISON, G.T.H. & WESTLIN-VAN AARDE, L.M. 1990. Ringtail in the pouched mouse (*Saccostomus campestris*). *Lab. Anim.* 24: 205-206.

ELLISON, G.T.H., BRONNER, G.N. & TAYLOR, P.J. *in press.* Is the annual cycle in body weight of pouched mice (*Saccostomus campestris*) the result of seasonal changes in adult size or population structure? *J. Zool., Lond.*

HAIM, A., ELLISON, G.T.H. & SKINNER, J.D. 1988. Thermoregulatory circadian rhythms in the pouched mouse (*Saccostomus campestris*). *Comp. Biochem Physiol.* 91A: 123-127.

HAIM, A., RACEY, P.A., SPEAKMAN, J.R., ELLISON, G.T.H. & SKINNER, J.D. 1991. Seasonal acclimatization and thermoregulation in the pouched mouse *Saccostomus campestris*. *J. therm. Biol.* 16: 13-17.

to hard-to-catch and hence best-to-avoid species of *Papilio*. I agree with Vane-Wright (Vane-Wright, R. I. 1976. *Proceedings of the Royal Entomological Society of London* **41**: 1–7. Vane-Wright, R. I. 1984. *The biology of butterflies*. Ed. R. I. Vane-Wright and P. R. Ackery. *Royal Entomological Society of London*, Eleventh Symposium, London, pp. 251–253) that the male-like females are likely to be transvestites.

Acknowledgement: Fig. 1 was drawn by Derek Whiteley from specimens in my collection – Denis F. Owen, Department of Biology, Oxford Polytechnic, Headington, Oxford, U.K.

***Dorylus helvolus* L. (Formicidae: Dorylinae) preying on *Saccostomus campestris* (Rodentia: Cricetidae).**– During May 1987, at Vaalkop Dam Nature Reserve, (27° 30′ E; 25° 30′ S) an adult, male pouched mouse *(Saccostomus campestris)*, normally a nocturnal species, was observed lying outside its burrow at 16h00. When disturbed, the mouse entered its burrow but reappeared above ground five minutes later. Approximately 40 red ants *(Dorylus helvolus)* covered its tail, groin and rear legs, and the mouse was clearly in some distress. Subsequent excavation of the burrow system in July 1987 revealed a colony of *D. helvolus* 85 cm from the nesting chamber. A connecting tunnel from the present nest to a disused chamber, which was adjacent to the ant colony, had been blocked for 6,5 cm of its length with loose soil and small stones. Traces of a marker dye, used to track the mouse to its burrow, were found in both chambers and as *S. campestris* builds a single nesting chamber (personal observation) it is probable that the original nest had been abandoned and the connecting tunnel plugged following the attack by the ants in May. Although the male pouched mouse on this occasion survived, Sheppey (personal communication) observed an adult female pouched mouse retrieve six juveniles from a burrow in Nwanedzi (Kruger National Park). All were covered in *D. helvolus* and of four juveniles collected by him, two subsequently perished.

Whilst their hypogaeic habits have meant that the biology of *D. helvolus*, in common with most African dorylines, is poorly researched, their ability to tackle small vertebrate prey (Savage, T. S. 1847, *Transactions of the Entomological Society of London* **5**: 1–15) makes them capable predators of fossorial or semi-fossorial rodents. Their ability to handle such prey would be further enhanced in species that undergo torpor (such as *S. campestris*, personal observation) as arousal time from torpor might preclude any attempt by such rodents to escape. Future studies of prey remains in the nests of *D. helvolus* and other African dorylines might indicate the extent to which semi/fossorial rodents feature in their diet.

Acknowledgements: The author thanks Dr R. Kfir for identifying the ants and K. A. E. Sheppey for his comments. This work was supported by bursaries from the South African Council for Scientific and Industrial Research (CSIR) and the Department of National Education to whom thanks are expressed – G. T. H. Ellison, Department of Zoology, University of Pretoria, Pretoria 0002.

A note on the small mammal fauna of Vaalkop Dam Nature Reserve

G.T.H. ELLISON

Mammal Research Institute, Pretoria University, Pretoria, 0002 Republic of South Africa.

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Vaalkop Dam Nature Reserve (25°23'S; 27°28'E), 50 km northeast of Rustenburg, comprises 2 391 hectares of Mixed Bushveld (Acocks 1975, *Memoirs of the Botanical Survey of South Africa* 40: 1-128), and 1 045 hectares of water surface contained in the dam itself (at full capacity).

Small mammal trapping was conducted at the south western boundary of the reserve between 24 April-6 May 1987, and 14-21 April 1989. Standard Sherman live traps, baited with peanut butter and rolled oats, were set in pairs, at 10 m intervals, in two trap lines: 1. *Dam edge*: along the edge of a small farm dam near the Warden's residence, comprising thick grass cover interspersed with small stands of *Eucalyptus* spp. 2. *Open bushveld*: within mixed bushveld with sparse grass cover and occasional *Acacia* spp. trees. Nine hundred and thirty six trap-nights were recorded in 1987 and 1 254 in 1989. Trapping success averaged over 85 % during 1987, and fell to approximately 40 % during 1989. Eleven small mammal species were captured in 1987, and 12 in 1989. These are listed in Table 1 together with their relative abundance and habitat preference. (Representatives of all 14 species collected have been deposited at the Transvaal Museum, Pretoria, South Africa).

During 1987 *Mastomys natalensis* was the dominant component of the small mammal community comprising over 85 % of all animals caught. It was not uncommon to find two individuals in a single trap, and on one occasion three were captured in this way. Two years later in 1989, however, less than 15 % of animals captured were *M. natalensis*, and whilst *Aethomys chrysophilus* was the dominant species (46,5 %), additional species were more evenly represented within the traps; the three Soricidae species comprising 13,8% of captures, and *Lemniscomys rosalia* making up 11,5 %.

The present study provides further evidence that populations of *M. natalensis* exhibit extreme population fluctuations under natural conditions. These fluctuations might have masked the relative abundance of other species within the small mammal community and could confound attempts to determine species composition using standard live-trapping techniques.

Smithers (1983, *The Mammals of the Southern African Subregion,* Pretoria: Pretoria University) suggested that population explosions of *M. natalensis* occur following the onset of heavy rains at the end of a prolonged dry period, and that their numbers mask similar responses by gerbils and other rodents. Indeed, the rainfall pattern at Vaalkop Dam Nature Reserve suggests that climatic factors may have been responsible for the huge population of *M. natalensis* observed: annual rainfall increased steadily from 380 mm in 1984, 423 mm in 1985, to 634 in 1986, the year preceeding this study. However, Nel (1975, *Publikasies van die Universiteit van Pretoria, Nuwe Reeks* 97: 78-80) tentatively suggested that field studies of this species to date indicate the possibility that population cycles occur independently of climatic changes, in much the same way as those reported for

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Table 1
Small mammals recorded at Vaalkop Dam Nature Reserve

Family	Specific name		Common name	Numbers caught 1987 (936 Trapnights)	Numbers caught 1989 (1 254 Trapnights)	Habitat preference
Insectivora						
Soricidae	*Suncus lixus*	(Thomas, 1898)	Greater dwarf shrew			
	Crocidura hirta	Peters, 1852	Lesser red musk shrew	2	71	Dam edge
	Crocidura cyanea	(Duvernoy, 1838)	Reddish-grey musk shrew			
Macroscelididae	*Elephantulus brachyrhynchus*	(A. Smith, 1836)	Short-snouted elephant shrew	5	28	Open bushveld
Rodentia						
Gliridae	*Graphiurus murinus*	(Desmarest, 1822)	Woodland dormouse	0	3	*Acacia* spp. thicket
Cricetidae	*Tatera leucogaster*	(Peters, 1852)	Bushveld gerbil	10	31	Open Bushveld
	Steatomys pratensis	Peters, 1846	Fat mouse	1	11	Open Bushveld
	Saccostomus campestris	Peters, 1846	Pouched mouse	15	5	Open Bushveld
	Otomys irroratus	(Brants, 1827)	Vlei rat	1	2	Dam edge
Muridae	*Aethomys chrysophilus*	(De Winton, 1897)	Red veld rat	54	239	Open Bushveld
	Lemniscomys rosalia	(Thomas, 1904)	Single-striped mouse	5	59	Open Bushveld
	Mus minutoides	A. Smith, 1834	Pygmy mouse	4	3	Open Bushveld
	Mastomys natalensis (sp. 1)	(A. Smith, 1834)	Natal multimammate mouse	692	62	Ubiquitous
	Rhabdomys pumilio	(Sparrman, 1784)	Striped mouse	7	0	Open Bushveld
				Total 796 (85.0 %)	514 (41.0 %)	

northern hemisphere microtines. This aspect of *M. natalensis* biology is worthy of further research.

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NORADRENALINE THERMOGENESIS IN CONSCIOUS AND ANAESTHETISED POUCHED MICE (*SACCOSTOMUS CAMPESTRIS*)

G. T. H. ELLISON and J. D. SKINNER
Mammal Research Institute, Pretoria University, Pretoria 0002, South Africa. Telephone: (012) 420-2066

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Abstract—1. The metabolic response to injections of noradrenaline (NA) and saline (control) was investigated in conscious and anaesthetised (sodium pentobarbitone) pouched mice, *Saccostomus campestris*.

2. NA injection produced a calorigenic response which was significantly greater than that elicited by saline injection in both conscious and anaesthetised animals.

3. This calorigenic response was enhanced by motor activity in conscious pouched mice, but the exclusion of measurements recorded during visible activity eliminated the influence of movement.

4. Anaesthetised pouched mice underwent mild hypothermia and displayed a retarded metabolic response to NA injection which suggests that anaesthesia affects the expression of NA-induced thermogenesis.

5. The validity of proposed techniques for the measurement of NA thermogenesis is further discussed.

INTRODUCTION

Non-shivering thermogenesis (NST) or "chemical thermoregulation" has been defined as "a heat-production mechanism liberating chemical energy due to processes which do not involve muscular contractions", and predominates in cold-acclimated, hibernating, new-born and smaller mammals (Janský, 1973). NST can be sub-divided into "obligatory" NST, which comprises mostly of basal metabolism, and "regulatory" NST which occurs at ambient temperatures below the thermoneutral zone (Janský, 1973). Because noradrenaline (NA) induced NST substitutes for thermogenesis in the cold the artificial stimulation of NST using injections of exogenous NA has become the most commonly used technique for quantifying NST (Bockler *et al.*, 1982). To ensure a maximal calorigenic response, an optimal, mass-specific dose of NA must be used (as derived by Heldmaier, 1971) whilst muscle activity must be checked, either by curarization (Janský, 1973) or, more usually, by deep anaesthesia (Bockler *et al.*, 1982). However, it remains unclear what effect deep anaesthesia exerts on the expression of NA-induced NST, although Vybíral and Janský (1974) found that NA elicited a similar thermogenetic response in conscious and anaesthetised hamsters (*Mesocricetus auritus*) following correction for motor activity induced by a control (saline) injection. The aim of the present study was therefore to investigate the effect of NA and saline (control) injections on the metabolism of conscious and anaesthetised pouched mice (*Saccostomus campestris*), a small cricetid rodent endemic to sub-Saharan Africa (Smithers, 1983).

MATERIALS AND METHODS

The pouched mice used in the present study were 6 adult females collected in Hereroland, Namibia (20°27′S 20°44′E) during March 1989. Upon arrival in Pretoria they were maintained under long photoperiod (14L:10D) at 25°C for 3 months and were subsequently transferred to 12L:12D at 25°C and acclimated under these conditions for 8 weeks prior to and throughout the experiment. The pouched mice were housed separately in standard laboratory rat cages containing sawdust, sand and shredded paper. The cages were cleaned once a week and dusted with insecticide ("Karbadust": Sentrachem, Silverton. Containing 50 g/kg carbamate) to eradicate ectoparasites. The mice were provided with sunflower seeds, rat pellets (Epol, Vereniging) and water *ad libitum*, a diet occasionally supplemented with pieces of fresh apple.

Following acclimation the resting metabolic rate (RMR) and metabolic response to subcutaneous injections of saline (0.9% NaCl) and noradrenaline (NA; 1.5 mg/kg) were measured at 30°C using an open-flow system as described by Depocas and Hart (1957) and Hill (1972). The pouched mice were placed in clear perspex respiration chambers (700 ml) through which a flow of dried air (silica gel) was passed at a rate of 600 ml/min. The chambers were immersed in a constant temperature water bath (Labotec, Isando), and a chromel alumel thermocouple (52 K/J: Fluke, Everett) within the chamber was used for monitoring chamber temperature. Following equilibration of the system, oxygen consumption ($\dot{V}O_2$) was recorded using an Ametek S3A/I (Applied Electrochemistry, Pittsburgh) oxygen analyser connected to a multi-channel data logger (Grant Instruments, Cambridge). The oxygen analyser was calibrated before and after measurement, and $\dot{V}O_2$ was corrected to standard temperature and pressure (STP). In addition, the body mass (M_b) of the pouched mice was recorded to the nearest 0.1 g following each $\dot{V}O_2$ measurement.

RMR was measured prior to each test injection in conscious mice following a 2–3 hr stabilising period. $\dot{V}O_2$ was recorded every 3 min for half an hour, and five consecutive readings which did not differ by more than 0.03% were used for calculating RMR. Rectal temperature (T_b) was subsequently measured using a chromel alumel thermocouple inserted approximately 3 cm into the rectum for a period not exceeding 15 sec. Following RMR measurement the pouched mice were injected with saline or NA solutions

(3 ml/kg) whilst conscious or following anaesthesia with sodium pentobarbitone (75 mg/kg "Sagatal": Maybaker, Port Elizabeth, *i.p.*).

Conscious mice were immediately returned to the respiration chamber whereupon $\dot{V}O_2$ and activity were continuously recorded at 1 min intervals for 30 min (following saline injection) or 60 min (following NA injection). Pouched mice were deemed inactive if they did not move throughout a recording interval, and readings taken following at least 2 min quiescence were considered representative of inactivity. At the end of each test the T_b of conscious animals was determined using the chromel alumel thermocouple.

Following anaesthesia a J-type thermocouple (Grant Instruments, Cambridge), connected to the data logger, was inserted deep into the rectum of unconscious pouched mice and attached to the tail with tape. The mice were then returned to the respiration chamber for 30 min prior to injection with saline or NA. $\dot{V}O_2$ and T_b were subsequently recorded continuously every minute for a further 30 min (following saline injection) or 90 min (following NA injection).

The metabolic response to saline injection was conducted 3 days prior to that recorded following NA injection, and the pouched mice were assigned to two groups (3 individuals each) such that the first group was measured under anaesthesia, the second whilst conscious, and these tests were repeated 1 month later, the first group whilst conscious and the second under anaesthesia.

All results are presented as mean $\pm$ one standard deviation, and Wilcoxon's matched-pairs, signed-rank test was used for the statistical analysis of results (Sokal and Rohlf, 1981).

RESULTS

The mean M_b of the pouched mice prior to the first test was 80.0 $\pm$ 14.5 g and decreased significantly over the following month to 76.4 $\pm$ 14.4 g ($T = 0$, $N = 6$, $P < 0.05$). This decline in M_b was accompanied by a slight increase in RMR from 0.627 $\pm$ 0.087 ml O_2/g/hr before the first test to 0.752 $\pm$ 0.083 ml O_2/g/hr before the second, although this increase was not significant ($T = 2$, $N = 6$, $P > 0.05$). There was also no significant difference between RMR measured prior to saline and NA injection for either anaesthetised or conscious animals during each test period ($P > 0.05$).

Conscious animals

Figure 1 displays a typical metabolic response of a conscious pouched mouse to injections of saline and NA. Both these injections caused an increase in metabolic rate which was to some extent enhanced by motor activity. The mean of the highest values of oxygen consumption ($\dot{V}O_{2max}$) recorded for all the pouched mice was 1.68 $\pm$ 0.50 ml O_2/g/hr following saline injection, and 3.47 $\pm$ 0.41 ml O_2/g/hr after NA injection, both of which were significantly higher than RMR ($T = 0$, $N = 6$, $P < 0.05$). However, inactive $\dot{V}O_{2max}$ following saline injection was 0.77 $\pm$ 0.17 ml O_2/g/hr which was not significantly greater than

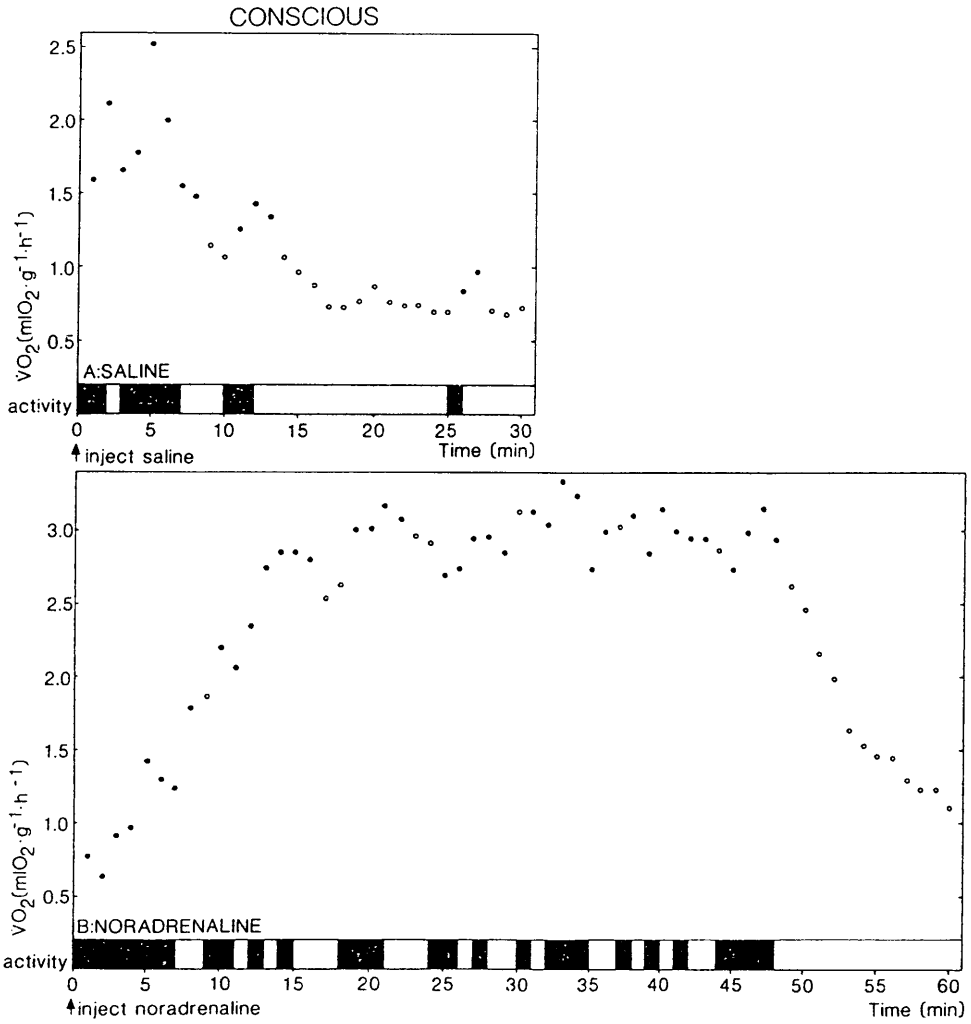

Fig. 1. Oxygen consumption ($\dot{V}O_2$) and activity of a conscious pouched mouse following injections of saline (above) and NA (below). Solid bars indicate activity during the period of measurement. Active readings of $\dot{V}O_2$ are indicated: ●, and readings following at least 2 min inactivity: ○.

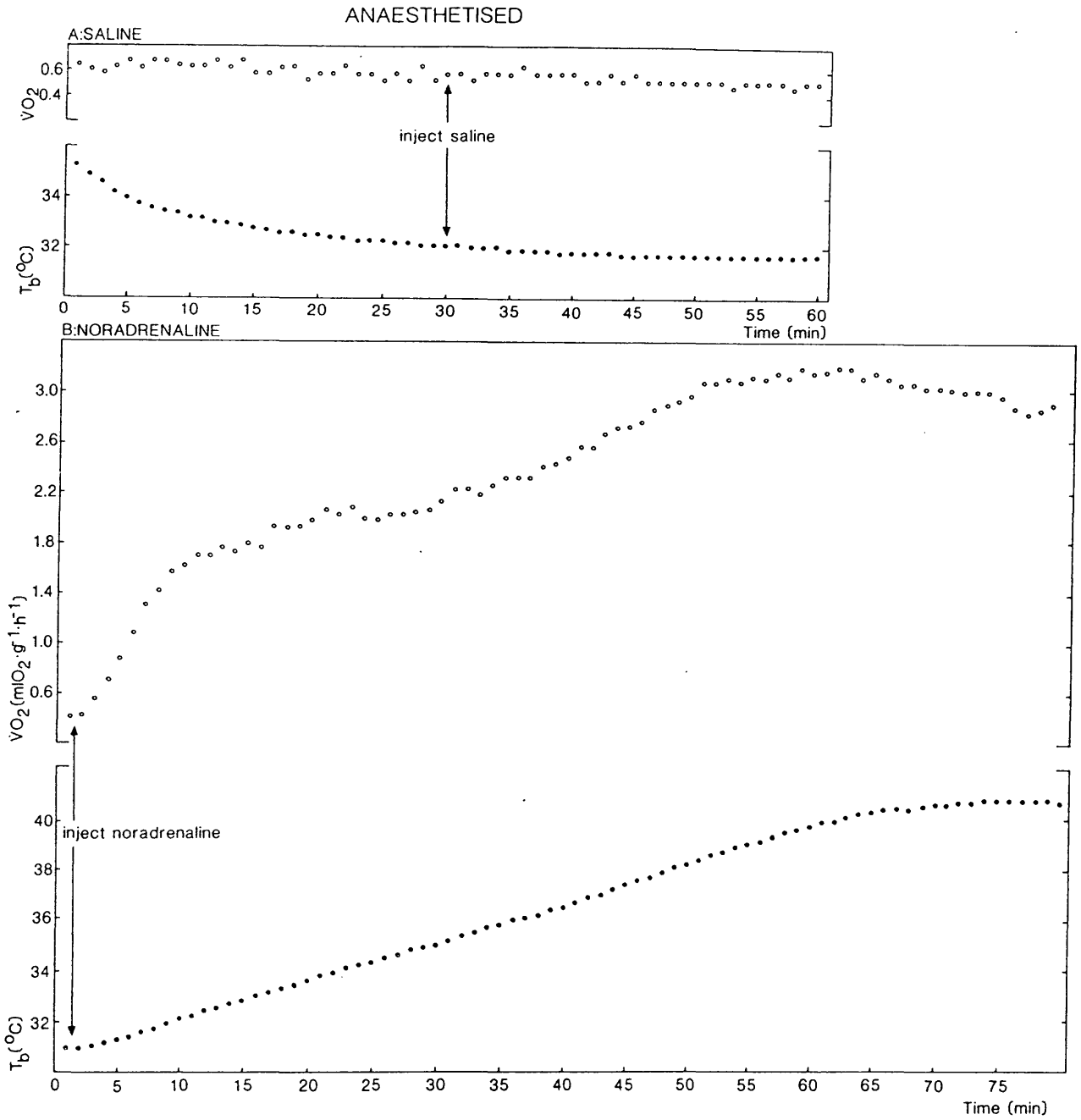

Fig. 2. Oxygen consumption ($\dot{V}O_2$) and body temperature (T_b) of an anaesthetised pouched mouse prior to and following injections of saline (above) and NA (below).

RMR ($T = 2$, $N = 6$, $P > 0.05$) whilst inactive $\dot{V}O_{2max}$ following NA injection (3.23 ± 0.46 ml O_2/g/hr) remained significantly greater than RMR ($T = 0$, $N = 6$, $P < 0.05$) and was elicited 20–40 min after administering NA. T_b measured after saline injection was 35.0 ± 0.5°C and 39.2 ± 1.1°C following an injection of NA, which compared to a mean T_b of 34.8 ± 0.4°C recorded after the measurement of RMR.

Anaesthetised animals

Figure 2 displays a typical metabolic response of an anaesthetised pouched mouse to injections of saline and NA. Anaesthetised pouched mice initially displayed a decline in $\dot{V}O_2$ accompanied by gradual hypothermia. Injection of saline did not arrest this decline and $\dot{V}O_2$ fell to 0.55 ± 0.18 ml O_2/g/hr and T_b to 33.0 ± 0.7°C. In contrast, NA injection produced a gradual increase in $\dot{V}O_2$ which peaked after 60–80 min with a mean $\dot{V}O_{2max}$ of 3.04 ± 0.42 ml O_2/g/hr and a corresponding T_b of 40.7 ± 0.8°C, both of which were significantly higher than RMR values ($T = 0$, $N = 6$, $P < 0.05$).

In order to compare the relative effects of saline and NA injection in anaesthetised and conscious pouched mice the metabolic responses to these injections are summarised in Fig. 3. $\dot{V}O_2$ recorded following NA injection fell between 432 and 515% RMR but there was no significant difference between these measures ($P > 0.05$). In contrast, saline injection produced values of $\dot{V}O_2$ between 78 and 249% RMR, and was significantly greater in active, conscious pouched mice, whilst the exclusion of active readings rendered this difference insignificant ($P < 0.05$).

DISCUSSION

The results of the present study clearly indicate that NA injection caused a calorigenic response in both

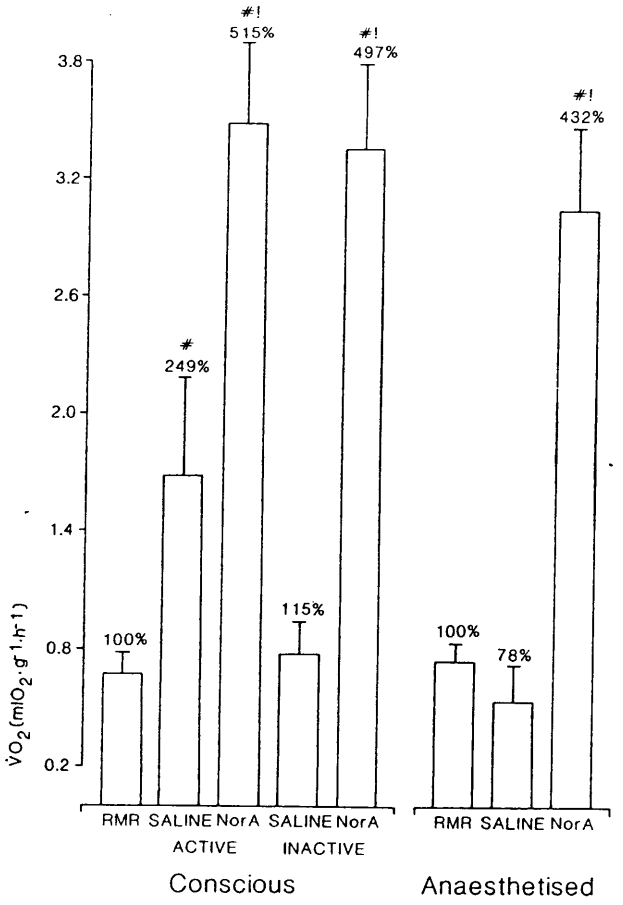

Fig. 3. A summary of oxygen consumption ($\dot{V}O_2$) recorded for conscious (active and inactive) and anaesthetised pouched mice prior to (RMR) and following saline and noradrenaline (NorA) injection. $\dot{V}O_2$ values significantly ($P < 0.05$) greater than RMR (#) and saline measurements (!) are indicated.

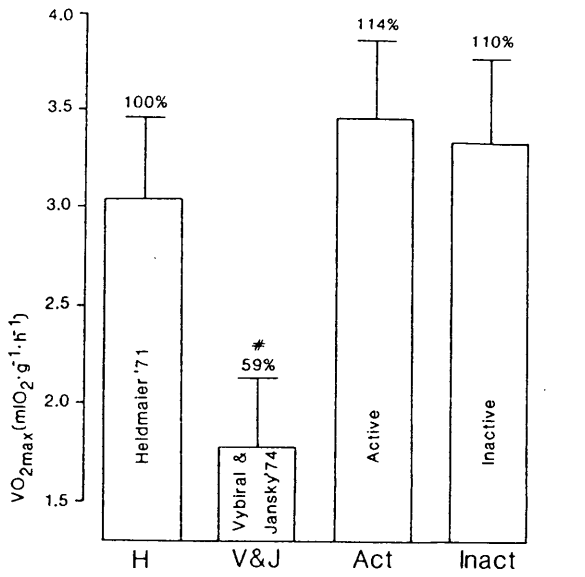

Fig. 4. A comparison of techniques for quantifying NA-induced thermogenesis ($\dot{V}O_{2max}$). Heldmaier's (1971) technique (H) for anaesthetised animals, Vybíral and Janský's (1974) technique (V&J) with correction for activity induced by saline injection in conscious animals, and measurements recorded for active (Act) and inactive (Inact) conscious pouched mice as described in the present study. # : V&J $\dot{V}O_{2max}$ measured was significantly ($P < 0.05$) lower than the other treatments.

conscious and anaesthetised pouched mice, which was significantly greater than that elicited by control injection of saline. Active (conscious) pouched mice also displayed an elevated $\dot{V}O_2$ following saline injection, but the effects of NA and activity did not appear to be synergistic.

Anaesthesia elicited two effects: a reduction in $\dot{V}O_2$ together with mild hypothermia prior to injection, and a delay in the metabolic response to NA. The former suggests a reduction in basal, "obligatory" NST or the elimination of muscle tone as a component of RMR. The latter indicates an effect on the expression of regulatory NST, possibly as a consequence of reduced metabolic rate prior to injection.

Two alternative methods have been proposed for estimating NST following NA injection: Heldmaier's (1971) technique involves the anaesthesis of subjects prior to injection, whilst Vybíral and Janský (1974) estimated NST as the metabolic response to NA minus that elicited by saline injection in conscious animals. These two interpretations are displayed in Fig. 4 for data collected in the present study, together with $\dot{V}O_{2max}$ elicited in conscious pouched mice including and excluding active readings. There was no significant difference between $\dot{V}O_{2max}$ recorded in conscious (active or inactive) pouched mice and that determined for anaesthetised animals ($P > 0.05$), whilst $\dot{V}O_{2max}$ determined by Vybíral and Janský's (1974) technique was significantly lower than these ($T = 0$, $N = 6$, $P < 0.05$). This, unexpected result may be due to differing levels of activity following saline and NA injection, which render this technique inaccurate.

We therefore conclude that NA-induced NST can be accurately quantified once activity has been excluded by anaesthesia (after Heldmaier, 1971) or by direct observation (as described in the present study).

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REFERENCES

Bockler H., Steinlechner St. and Heldmaier G. (1982) Complete cold substitution of noradrenaline-induced thermogenesis in the Djungarian hamster, *Phodopus sungorus*. *Experientia* **38**, 261–262.

Depocas F. and Hart J. S. (1957) Use of the Pauling oxygen analyser for measurements of oxygen consumption of animals in open-circuit system and in short-lag, closed-circuit apparatus. *J. appl. Physiol.* **10**, 388–392.

Heldmaier G. (1971) Zitterfreie wärmbildung und körpergröße bei säugetieren. *Z. vergl. Physiol.* **73**, 222–248.

Hill R. W. (1972) Determination of oxygen consumption by use of the paramagnetic oxygen analyser. *J. appl. Physiol.* **33**, 261–263.

Janský L. (1973) Non-shivering thermogenesis and its thermoregulatory significance. *Biol. Rev.* **48**, 85–132.

Smithers R. H. N. (1983) *The Mammals of the Southern African Subregion*. University of Pretoria Press, Pretoria. 283 pp.

Sokal R. R. and Rohlf F. J. (1981) *Biometry*. W. H. Freeman, San Francisco. 448 pp.

Vybíral S. and Janský L. (1974) Non-shivering thermogenesis in the golden hamster. *Physiol. Bohemoslov.* **23**, 235–243.

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Seasonal energy requirements and thermoregulation of growing pouched mice, *Saccostomus campestris* (Cricetidae)

G.T.H. Ellison and J.D. Skinner

Mammal Research Institute, University of Pretoria, Pretoria 0002, Republic of South Africa

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Abstract. Pouched mice (*Saccostomus campestris*) were born in captivity during January and March and subsequently maintained under long photoperiod (14 h light:10 h dark) at 25° C. During their first winter (July) and the following summer (January) the pouched mice were exposed to natural photoperiod in an unheated laboratory for 3 weeks prior to measurement. The pouched mice continued to grow during the study, and were significantly heavier after summer exposure than after winter exposure 6 months earlier. Although this increase in body mass would result in a decline in their surface area to volume ratio there was no significant decline in minimal thermal conductance (C_m) and winter-exposed pouched mice had a relatively lower C_m than expected. Meanwhile the smaller, winter-exposed animals displayed a significantly higher capacity for non-shivering thermogenesis, together with higher levels of basal metabolism than summer individuals. These differences were not solely attributable to the contrasting body mass of each group and it is therefore clear that *S. campestris* can increase thermoregulatory heat production, and modify heat loss following exposure to short photoperiod and cold during their first winter. Despite the significant increase in metabolism, the overall energy requirements of small, winter-exposed animals were significantly lower than those for heavier pouched mice following exposure to summer conditions. These results suggest that growing pouched mice can effectively adapt to lower temperature conditions during their first winter, yet accrue considerable overall savings in total energy requirements as a result of their smaller body mass.

Key words: Seasonal acclimation – Thermoregulation – Energetics – *Saccostomus*

Introduction

Previous studies have shown that many Holarctic populations of small mammals exhibit a decline in mean body mass during winter (Fuller et al. 1969; Iverson and Turner 1974; Klaus et al. 1988) either as a result of a reduction in adult body mass, or an increase in the number of small juvenile animals entering the population at the end of the breeding season. One might expect that smaller individuals, having relatively higher thermal conductances as a result of their increased surface area to volume ratio, might experience higher energetic demands during cold periods associated with winter. However, Heldmaier and Steinlechner (1981) demonstrated that adult *Phodopus sungorus* lost weight and increased thermoregulatory heat production during winter, yet displayed significantly lower total energy requirements compared to heavier summer animals.

These energetic savings led Korn (1989) to postulate that annual cycles of body weight amongst small mammals from the subtropical Transvaal might also represent an adaptation to reduce total energy requirements during the cool dry season, although he did not differentiate between adult and juvenile individuals. The aim of the present study was to investigate thermoregulation and energy requirements of growing pouched mice (*Saccostomus campestris*) a wet-season breeder which displays a significantly lower mean body mass during winter (Korn 1989).

Materials and methods

A pair of pouched mice, collected at Nwanedzi (E. Transvaal, Republic of S. Africa; 24°23′ S 31°40′ E) during December 1986, were bred in captivity at Pretoria (25°45′ S 28°12′ E) and gave birth to two litters of ten pups (11 female and 9 male) on 25th January and 22nd March 1987. These mice were weaned at 21 days of age, and housed in a controlled climate chamber under long photoperiod (14 h light:10 h dark) at 25° C. During their first winter (July 1987) and the following summer (January 1988) they were removed from the climate chamber and exposed to natural photoperiod in an unheated laboratory for a period of 3 weeks prior to the measurement of metabolic parameters. In July 1987, laboratory temperatures varied between a maximum of 18.2° C and a minimum of 9.8° C, whilst in January 1988 temperatures were higher, the maximum being 28.4° C and the minimum 24.0° C.

Throughout the present study, pouched mice were housed separately in standard laboratory rat cages and provided with rat pellets (Epol, Vereeniging) and water ad libitum, a diet occasionally sup-

Offprint requests to: G.T.H. Ellison

plemented with sunflower seeds, apple and carrots. Shredded paper was provided for bedding, and their cages were cleaned weekly and dusted with insecticide containing 50 g/kg carbamate (Karbadust; Sentrachem, Silverton) to eradicate ectoparasites. After 3 weeks of laboratory exposure during winter and summer the metabolic parameters described below were measured in five pouched mice, chosen at random from the 20 offspring.

Oxygen consumption. Oxygen consumption ($\dot{V}O_2$) was measured at ambient temperatures (T_a) between 5° and 34° C, using an open circuit system (as described by Depocas and Hart 1957; Hill 1972). Usually, the $\dot{V}O_2$ of any one pouched mouse was only recorded at a single T_a per day, but occasionally, at T_a values around the thermoneutral zone (24°–32° C), $\dot{V}O_2$ was measured at two consecutive T_a values daily. Measurements were made during daylight, the inactive phase of *S. campestris*, and whilst each animal was at rest.

The pouched mice were placed in a clear perspex respiration chamber (volume 700 ml) through which a flow of dried air (prior passage through silica gel; Holpro, Johannesburg) was passed at a rate of 600 ml/min. The respiration chamber was immersed in a constant temperature water bath (Labotec, Isando) and a chromel-alumel thermocouple (52 K/J; Fluke, Everett) placed within the chamber was used for monitoring T_a. After a 2–3 h stabilising period and following full equilibration, $\dot{V}O_2$ was recorded using an Ametek S-3A/I (Applied Electrochemistry, Pittsburgh) oxygen analyser, and readings were taken every 3 min for half an hour. For T_a values between 15° and 34° C, five consecutive readings which did not differ by more than 0.03% were used for calculating resting metabolic rate (RMR). At 5° and 10° C, $\dot{V}O_2$ was variable, and the mean of ten consecutive 3-min readings, recorded whilst each animal was at rest, was used for computing RMR. All $\dot{V}O_2$ measurements were corrected to standard temperature and pressure, dry (STPD) and the oxygen analyser was calibrated before and after each measurement.

Body weight. Body weight (M_b) was measured prior to the transfer of animals from the climate chamber, and after each $\dot{V}O_2$ measurement using a PB3000 Mettler Scale (Mettler, Zurich). Animals were restrained within a cotton sack, and M_b was recorded to the nearest 0.1 g.

Body temperature. Rectal temperature (T_b) was recorded following each $\dot{V}O_2$ measurement using a chromel-alumel thermocouple connected to a 52 K/J Fluke digital thermometer. The thermocouple was inserted approximately 3 cm into the rectum for a period not exceeding 15 s.

Minimal thermal conductance. Minimal thermal conductance (C_m) was calculated at ambient temperatures below thermoneutrality using the formula of Scholander et al. (1950), where $C_m = \dot{V}O_2/(T_b - T_a)$.

Non-shivering thermogenesis. Using the technique of Ellison and Skinner (1990) non-shivering thermogenesis (NST) was measured in conscious pouched mice at T_a 28° C during winter and at T_a 30° C during summer. Following at least five consecutive 3-min $\dot{V}O_2$ readings which did not differ by >0.015%, 1.5 mg/kg noradrenaline (NA) (L-noradrenaline bitartrate; Sigma, St Louis) was injected into the mice subcutaneously. Maximum noradrenaline-induced $\dot{V}O_2$ ($\dot{V}O_{2\,NA}$) for 45 min during the response to NA was taken as maximum NST, and the change in T_b following injection of NA (ΔT_b) was calculated from T_b recordings before ($T_{b\,min}$) and 45 min after ($T_{b\,max}$) injection; thus $\Delta T_b = T_{b\,max} - T_{b\,min}$.

Statistical analysis. All results are presented as mean ± one standard deviation. Mann-Whitney U-tests (MWU), two-way analyses of variance (ANOVA) and variance ratio tests (VRT) for linear regression were used for the statistical analysis of the results (Sokal and Rohlf 1981).

Results

Body weight

Following winter exposure the mean M_b of pouched mice fell from 66.0 ± 8.2 g to 63.6 ± 5.3 g. In contrast, the mean M_b of pouched mice increased following summer exposure from 85.0 ± 15.6 g to 89.8 ± 11.4 g. These changes in body weight were not significant (MWU; winter, $U' = 18$, $P > 0.05$; summer, $U' = 15$, $P > 0.05$; $n = 5$) and nor was the difference in body weight of pouched mice prior to each season's exposure ($U' = 21$, $P > 0.05$, $n = 5$). However, the mean M_b of pouched mice following summer exposure was significantly higher than that of mice following exposure to winter conditions ($U' = 25$, $P < 0.05$, $n = 5$).

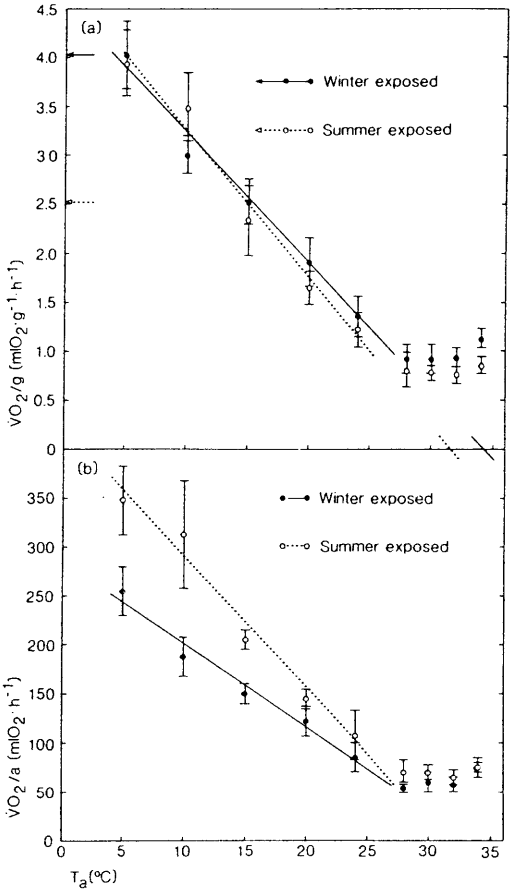

Fig. 1. **a** Mean mass-specific oxygen consumption ($\dot{V}O_2$/g) and **b** mean overall oxygen consumption ($\dot{V}O_2$/a) of growing pouched mice following winter (●) and summer (○) exposure at ambient temperatures (T_a) between 5° and 34° C. *Vertical lines* indicate one standard deviation of the mean. *Arrows* in **a** indicate maximum specific oxygen consumption ($\dot{V}O_{2\,NA}$) of winter- (←) and summer (◂···) exposed pouched mice following injection of noradrenaline. Regression lines describing the relationship of $\dot{V}O_2$/g and $\dot{V}O_2$/a on T_a values below 28° C were fitted using the least-squares technique, and have the following equations: **a** winter, $\dot{V}O_2/g = 4.537 - 0.133\ T_a$ ($r = -0.964$); summer, $\dot{V}O_2/g = 4.740 - 0.150\ T_a$ ($r = -0.961$); **b** winter, $\dot{V}O_2/a = 286.4 - 8.5\ T_a$ ($r = -0.950$); summer, $\dot{V}O_2/a = 424.8 - 13.5\ T_a$ ($r = -0.939$)

Oxygen consumption

The mass-specific $\dot{V}O_2$ ($\dot{V}O_2$/g) of winter- and summer-exposed pouched mice between T_a 5° C and 34° C is illustrated in Fig. 1a. Both groups of mice exhibited a broad thermoneutral zone (TNZ) between T_a 28° C and 32° C, whilst the RMR at thermoneutrality (≡Basal Metabolic Rate, BMR) of smaller, winter-exposed pouched mice (0.914±0.115 ml O_2/g per h) was significantly higher than that of summer animals (0.786±0.018 ml O_2/g per h) (ANOVA, $F=8.897$, $df=1, 24$; $P<0.01$). When compared to the predicted BMR for Cricetids of similar body weight (Hayssen and Lacy 1985) both winter- and summer-exposed pouched mice displayed levels of basal metabolism that were lower than those predicted; 71.0% (winter) and 66.1% (summer).

Below the TNZ, the RMRs of both groups of pouched mice increased linearly with decreasing T_a (Fig. 1a) such that $\dot{V}O_2/g=4.537-0.133\ T_a$ ($r-0.964$) for winter-exposed animals, and $\dot{V}O_2/g=4.740-0.150\ T_a$ ($r=-0.961$) for pouched mice in summer. Despite the difference in mean M_b, there were no significant differences in weight-specific RMR below thermoneutrality (VRT, $F_s=0.026$, $df=1, 5.3$; $P>0.05$). However, expressing the data as oxygen consumption/animal ($\dot{V}O_2$/a) (Fig. 1b), clearly revealed the lower *overall* RMR of smaller, winter-exposed pouched mice at T_a values below 28° C. Indeed the regression of $\dot{V}O_2$/a against T_a for pouched mice in winter was significantly lower than that for summer animals ($F_s=10.026$, $df=1, 707.2$; $P<0.01$) (linear regression equations: winter, $\dot{V}O_2/a=286.4-8.47\ T_a$; summer, $\dot{V}O_2/a=424.8-13.5\ T_a$).

Body temperature

The T_b of pouched mice at T_a values between 5° and 34° C are displayed in Fig. 2. The mean T_b of winter- and summer-exposed pouched mice at thermoneutrality was similar, being 35.5±0.5° C and 35.5±0.3° C respectively. Both groups exhibited a rise in T_b above T_a 28° C, whilst summer-exposed pouched mice retained a constant mean T_b of 35.6±0.3° C at T_a values below 28° C. In contrast, the T_b of pouched mice in winter fell to 34.8±0.3° C below T_a 28° C , which was significantly lower than that for summer animals (ANOVA, $F=12.337$, $df=1, 40$; $P<0.01$).

Minimal thermal conductance

Because thermal conductance at temperatures below thermoneutrality was variable (see Fig. 3) it is not surprising that the regressions of $\dot{V}O_2$/g on T_a in Fig. 1 did not extrapolate to T_b as predicted by Newton's Law (Scholander et al. 1950). Variation in conductance might be the result of labile T_b values or changes in pelage insulation caused by piloerection and/or shivering at lower temperatures. Therefore, we used the lowest value

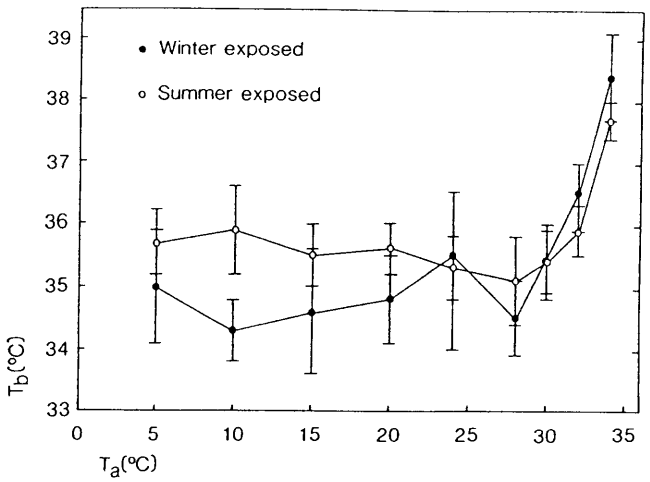

Fig. 2. Mean body temperature (T_b) of growing pouched mice following winter (●) and summer exposure (○) at ambient temperatures (T_a) between 5° and 34° C. *Vertical lines* indicate ± one standard deviation of the mean

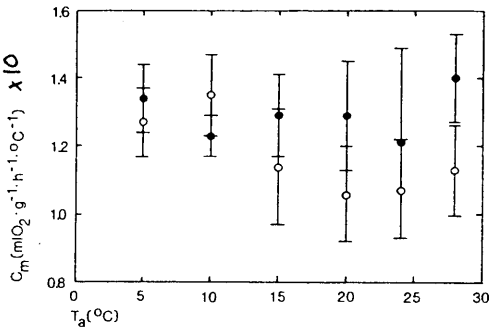

Fig. 3. Mean minimal thermal conductance (C_m) of growing pouched mice measured at ambient temperatures (T_a) below thermoneutrality (5°–28° C) following winter (●) and summer exposure (○). *Vertical lines* indicate ± one standard deviation of the mean

of conductance recorded below thermoneutrality in order to provide an estimate of minimal thermal conductance (C_m; McNab 1980). Using these data there was no significant difference between C_m of winter- ($C_m=0.121\pm0.028$ ml O_2/g per h per ° C) and summer- ($C_m=0.106\pm0.014$ ml O_2/g per h per ° C) exposed pouched mice (MWU, $U=9$, $n_1=n_2=5$, $P>0.05$).

Non-shivering thermogenesis

Following NA injection, the mass-specific $\dot{V}O_{2\,NA}$ of winter-exposed pouched mice (4.095±0.308 ml O_2/g per h) was significantly higher than that for summer animals (2.521±0.293 ml O_2/g per h) (MWU, $U'=25$, $n=5$, $P<0.05$). When compared to RMR at T_a 5° C in Fig. 1a, $\dot{V}O_{2\,NA}$ clearly accounted for 100% of cold thermogenesis in winter (RMR = 4.016±0.341 ml O_2/g per h) but only 65% during summer (RMR = 3.897±0.323 ml O_2/g per h). In addition, the mean increase in T_b following NA injection was higher in winter-exposed pouched mice

(mean $\Delta T_b = 4.0 \pm 0.3°$ C) compared to summer animals (mean $\Delta T_b = 3.0 \pm 0.5°$ C).

Discussion

Wunder (1984) considered the two most important stresses facing small mammals in winter to be increased cold and reduced food availability. Small mammals respond to these environmental constraints with a variety of different adaptations comprising mechanisms of either "resistance" (increased metabolic heat production) or "avoidance" (migration, microclimate selection, torpidity and increased insulation) (Wunder 1984).

The significantly higher levels of BMR and NST (resistance) displayed by winter-exposed *S. campestris* in the present study are similar to those described for other rodents both during winter (Lynch 1973; Wunder et al. 1977; Klaus et al. 1988) and following cold acclimation (Haim 1982; Heldmaier et al. 1982; Feist and Feist 1986). Although BMR and NA-induced $\dot{V}O_{2\,NA}$ are inversely related to body mass (Hayssen and Lacy 1985; Heldmaier 1971), the observed increases in both parameters exceeded those predicted on the basis of body mass alone: predicted BMR was 8.2% (Hayssen and Lacy 1985) higher for smaller, winter-exposed pouched mice compared to the 20.0% observed, and predicted $\dot{V}O_{2\,NA}$ was 17.0% (Heldmaier 1971) higher during winter compared to the 62.4% observed. Clearly, the enhanced metabolic heat production of winter-exposed *S. campestris* was not solely due to their lower body mass.

Because smaller animals exhibit a greater surface area to volume ratio, it was surprising that C_m was not significantly higher for smaller *S. campestris* following winter exposure. Indeed, the observed difference in C_m (14.2%) was lower than that predicted for the difference in body mass according to Bradley and Deavers' (1980) equation for Cricetids (19.8%). Thus winter-exposed pouched mice displayed a relative *improvement* in insulation, enhanced by the significantly lower T_b recorded at temperatures below thermoneutrality during winter which would have reduced the temperature gradient between their body core and the environment. Had these animals maintained a constant T_b of 35.5° C below the TNZ, their C_m would have been 117.0% of that observed for summer-exposed animals. This percentage increase in C_m is still lower than that predicted by Bradley and Deavers (1980), and it therefore appears that winter-exposed pouched mice displayed enhanced insulation (avoidance) possibly through an increase in pelage length or density, as reported for cold-acclimated *Phodopus sungorus* (Heldmaier and Steinlechner 1981) and *Mus musculus* (Al-Hilli and Wright 1988).

Genuine weight reductions in wild populations of several rodent species have been reported during winter (Fuller et al. 1969; Iverson and Turner 1974; Klaus et al. 1988; Korn 1989). At first sight, this weight loss might be interpreted as an indication of lower food quality/availability during winter, or as a decrease in the proportion of fully grown adults within a population. However, Heldmaier and Steinlechner (1981) demonstrated that the decline in body mass of adult *P. sungorus* during winter occurred in the presence of ad libitum food. These hamsters displayed an increase in heat production (BMR and NST) and a decrease in heat loss (C_m) following cold acclimation, yet had lower overall energy requirements compared to larger individuals during summer (Heldmaier and Steinlechner 1981). This can be explained by the fact that although mass-specific metabolic rate is an accurate description of heat production or "metabolic turnover rate" (MTR; Kleiber 1975), the energy needs of the same individual are better expressed as the total metabolism per animal (TM or $\dot{V}O_2$/a). Thus TM indicates the amount of energy that each animal needs to "gather, consume and assimilate (in order) . . . to maintain themselves" (Wunder et al. 1977) and does not mask differences in body weight. For this reason smaller animals can accrue considerable savings in total energy requirements even when MTR is enhanced to maintain homeothermy during seasonally cold periods.

Recently, Korn (1989) demonstrated a clear annual cycle in the mean body mass of *S. campestris* collected within the Transvaal, with lowest body masses recorded during the cool, dry winter. He also suggested that such changes in body mass would lower the overall energy requirements of individuals within the population during this time of year, although it was unclear whether the lower mean body mass in winter was due to a reduction in individual body weight of adult animals, or a shift in the population structure towards one dominated by smaller animals from the previous breeding season (Korn 1989).

In the laboratory, *S. campestris* reaches 90% of its asymptotic body mass at the age of ca. 140 days (Westlin-van Aarde 1989); thus winter-exposed pouched mice in the present study had already exceeded 90% of their adult mass, yet were significantly heavier following exposure to summer conditions 6 months later. Although the smaller pouched mice displayed higher levels of metabolic heat production following exposure to winter, their total metabolism below thermoneutrality was significantly lower than that for larger, summer-exposed animals. Thus it appears that growing *S. campestris* remain small during their first winter and accrue considerable energetic savings despite an increase in thermogenic capacity following cold exposure. This might explain the predominance of small individuals occurring during winter in their natural environment (Korn 1989).

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References

Al-Hilli F, Wright EA (1988) The effects of environmental temperature on the hair coat of the mouse. J Therm Biol 13:21–24

Bradley SR, Deavers DR (1980) A re-examination of the relationship between thermal conductance and body weight in mammals. Comp Biochem Physiol 65A:465–476

Depocas F, Hart JS (1957) Use of the Pauling oxygen analyzer for measurements of oxygen consumption of animals in open-circuit system and in short-lag, closed-circuit apparatus. J Appl Physiol 10:388–392

Ellison GTH, Skinner JD (1990) Noradrenaline thermogenesis in conscious and anaesthetised pouched mice (*Saccostomus campestris*). Comp Biochem Physiol 97A:23–26

Feist DD, Feist CF (1986) Effects of cold, short day and melatonin on thermogenesis, body weight and reproductive organs in Alaskan red-backed voles, *Clethrionomys rutilus dawsonii*. J Comp Physiol 156:741–746

Fuller WA, Stebbins LL, Dyke GR (1969) Overwintering of small mammals near Great Slave Lake, Northern Canada. Arctic 22:34–55

Haim A (1982) Effects of long scotophase and cold acclimation on heat production in two diurnal rodents. J Comp Physiol 148:77–81

Hayssen V, Lacy RC (1985) Basal metabolic rates in mammals: taxonomic differences in the allometry of BMR and body mass. Comp Biochem Physiol 81A:741–754

Heldmaier G (1971) Zitterfreie Wärmebildung und Körpergröße bei Säugetieren. Z Vergl Physiol 73:222–248

Heldmaier G, Steinlechner S (1981) Seasonal control of energy requirements for thermoregulation in the Djungarian hamster (*Phodopus sungorus*), living under natural photoperiod. J Comp Physiol 142:429–437

Heldmaier G, Steinlechner S, Raphael J (1982) Nonshivering thermogenesis and cold resistance during seasonal acclimation in the Djungarian hamster. J Comp Physiol 149:1–9

Hill RW (1972) Determination of oxygen consumption by use of the paramagnetic oxygen analyzer. J Appl Physiol 33:261–263

Iverson SL, Turner BN (1974) Winter weight dynamics in *Microtus pennsylvanicus*. Ecology 55:1030–1041

Klaus S, Heldmaier G, Ricquier D (1988) Seasonal acclimation of bank voles and wood mice: nonshivering thermogenesis and thermogenetic properties of brown adipose tissue mitochondria. J Comp Physiol 158:157–164

Kleiber M (1975) Metabolic turnover rate: A physiological meaning of the metabolic rate per unit body weight. J Theor Biol 53:199–204

Korn H (1989) The annual cycle in body weight of small mammals from the Transvaal, South Africa, as an adaptation to a subtropical seasonal environment. J Zool Lond 218:223–231

Lynch GR (1973) Seasonal changes in thermogenesis, organ weights and body composition in the white-footed mouse, *Peromyscus leucopus*. Oecologia (Berlin) 13:363–376

McNab BK (1980) On estimating thermal conductance in endotherms. Physiol Zool 53:145–156

Scholander PF, Walters V, Hock R, Irving L (1950) Body insulation of some arctic and tropical mammals and birds. Biol Bull 99:237–258

Sokal RR, Rohlf FJ (1981) Biometry. WH Freeman, San Francisco

Westlin-van Aarde LM (1989) Pre- and post-natal development of pouched mice, *Saccostomus campestris*. J Zool Lond 218:497–501

Wunder BA (1984) Strategies for, and environmental cueing mechanisms of, seasonal changes in thermoregulatory parameters of small mammals. In: Merritt JF (ed) Winter ecology of small mammals. Carnegie Mus Nat Hist Spec Publ 10:165–172

Wunder BA, Dobkin DS, Gettinger RD (1977) Shifts of thermogenesis in the prairie vole (*Microtus ochrogaster*). Oecologia (Berlin) 29:11–26

Journal of thermal Biology (*in press*).

THE INFLUENCE OF AMBIENT TEMPERATURE ON SPONTANEOUS DAILY TORPOR IN POUCHED MICE (*SACCOSTOMUS CAMPESTRIS*: RODENTIA - CRICETIDAE) FROM SOUTHERN AFRICA

G.T.H. ELLISON and J.D. SKINNER

Mammal Research Institute, University of Pretoria, Pretoria 0002

Republic of South Africa.

Abstract - 1. 18 pouched mice (*Saccostomus campestris*), from three different localities in southern Africa, responded to a daily decline in ambient temperature (T_a) from 20°C to 5°C by increasing metabolic heat production, thermolability and entering bouts of torpor.

2. The incidence of torpor increased with decreasing T_a although torpor was only observed in a total of 8 individuals at temperatures below those prevailing in their winter burrows.

3. The minimum body temperature (T_{bmin}) of torpid individuals ranged between 30.0°C & 21.0°C, but metabolic heat production during torpor increased to maintain T_{bmin} as T_a declined.

4. Torpor duration was correlated with minimum oxygen consumption (VO_{2min}), but the energy savings associated with torpor were not correlated to either torpor duration, T_{bmin} or VO_{2min}.

5. Individuals from all three localities entered torpor, while there appeared to be some geographical variation in the expression of torpor.

6. Male pouched mice had significantly lower values of T_{bmin} and VO_{2min} than females and 6/9 males entered torpor compared to 2/9 females.

Key Word Index - Spontaneous torpor; pouched mice; *Saccostomus campestris*; geographic variation; sexual difference.

INTRODUCTION

Strict homeothermy and high levels of activity can be self-defeating for small mammals in an energetically stressful environment (Brown & Bartholomew 1969). For this reason, a wide variety of small rodents enter short periods of facultative hypothermia, or daily torpor, and in this way reduce their overall metabolic expenditure when confronted with energetic stress in the form of low ambient temperatures (eg. Heldmaier & Steinlechner 1981, Maclean 1981, Thompson 1985). Hudson (1978) defined such torpor as a drop in body temperature below 30°C followed by a spontaneous arousal to normothermic body temperature.

In addition to cold-elicited or "spontaneous" torpor, additional studies have demonstrated that food restriction can elicit "induced" torpor (Hill 1975) even in normothermic species such as *Mus musculus* (Hudson & Scott 1979). In view of their limited fat reserves (Bronson 1987) it is not surprising that small mammals enter torpor when food restricted and induced torpor may represent an emergency mechanism rather than a controlled adjustment of energy balance (Ruf *et al.* 1991).

The pouched mouse, *Saccostomus campestris*, is a small cricetid rodent which displays torpid behaviour in the laboratory as a result of cold (Haim *et al.* 1988, unpublished data) but not food restriction (Earl 1980). However, spontaneous torpor has yet to be fully described in this species, and the present study aimed to investigate whether declining temperature could elicit torpor in pouched mice from three different localities in southern Africa.

MATERIALS AND METHODS

Animals

The pouched mice used in the present study originated from three localities in southern Africa and displayed different karyotypes (2n) as follows: *Letaba* (Kruger National Park: RSA, 23°56'S 31°35'E, 2n=46), *Gam* (S.E. Bushmanland: Namibia, 20°27'S 20°44'E, 2n=35) and *Pniel* (Barkly West: RSA, 28°35'S 24°31'E, 2n=32). Rainfall and air temperature data for each locality were obtained from the nearest meteorological station, together with soil temperatures 0.3m below ground which provide an indication of burrow temperatures. These climatic data are summarised in Table 1.

Table 1. Climatic Conditions for Pouched Mice Localities

Locality	(Meteorological Station)	Altitude	Mean Annual Rainfall	Mean Daily Temperatures: Summer (November-February) Air Max.	Air Min.	Soil 0.3m at 08h00[1]	Winter (May-August) Air Max.	Air Min.	Soil 0.3m at 08h00[1]
Letaba	(Letaba 23°51'S 31°35'E)	215m	387mm	33.2°C	20.8°C	28.1°C[2]	27.0°C	9.5°C	20.1°C[2]
Gam	(Grootfontein 19°36'S 18°08'E)	1398m	611mm	30.7°C	17.5°C	29.8°C	24.8°C	6.1°C	18.8°C
Pniel	(Barkly West 28°32'S 24°32'E)	1204m	391mm	31.8°C	15.8°C	26.9°C[2]	20.7°C	2.5°C	13.2°C[2]

Note 1. The 0.3m sub-soil temperatures approximate that prevailing in a *S.campestris* nest (which lie 0.22-0.86m below ground, n=10, pers.obs.).

Note 2. Sub-soil temperature data were not available for Letaba or Barkly West Met. Stations, and data for these were provided by Phalaborwa (23°56'S 31°09'E) and BJ Vorster (28°48'S 24°46'E) Met. Stations.

Table 2. A summary of metabolic measurements of 8 pouched mice entering torpor.

Locality:	No:	Sex: (M/F)	Stock: (W/L)	T_a: (°C)	VO_{2min}: ($ml O_2 \cdot g^{-1} \cdot h^{-1}$)	Tbmin: (°C)	Torpor duration: (D_t - h)	Overshoot duration: (D_o - h)	Torpor cost: (T - $ml O_2 \cdot g^{-1}$)	Overshoot cost: (O - $ml O_2 \cdot g^{-1}$)	Non-torpid Equivalent: (N - $ml O_2 \cdot g^{-1}$)	Energy Savings: (per bout of torpor)	Energy Savings: (per day)
Letaba	142	F	L	10	0.767	24.5	4.9	1.5	5.990	4.219	14.143	27.8%	13.5%
Letaba	214	M	L	10	0.867	25.5	3.8	2.4	5.440	6.453	13.751	13.5%	-7.8%
Letaba	142	F	L	5	0.996	24.5	incomplete	incomplete	-	-	-	-	-
Letaba	214	M	L	5	1.152	24.5	incomplete	incomplete	-	-	-	-	-
Gam	1513	M	W	15	0.456	25.0	13.2	1.9	12.051	2.107	26.262	46.1%	38.5%
Gam	1503	M	W	15	0.324	22.5	6.9	1.1	5.133	2.821	13.961	43.0%	28.4%
Gam	1513	M	W	10	0.388	21.0	12.0	1.2	13.807	3.341	30.188	43.2%	39.0%
Gam	1503	M	W	10	0.608	22.5	7.5	1.2	8.550	4.102	19.842	36.2%	28.2%
Gam	1512	F	W	10	1.116	27.0	2.9	0.3	4.502	0.572	7.090	28.3%	14.1%
Gam	1513	M	W	5	0.800	30.0	incomplete	incomplete	-	-	-	-	-
Gam	1512	F	W	5	1.308	26.0	incomplete	incomplete	-	-	-	-	-
Pniel	1504	M	W	10	1.056	27.5	4.8	1.2	6.776	2.968	12.264	20.5%	11.9%
Pniel	1504	M	W	5	1.142	26.0	incomplete	incomplete	-	-	-	-	-
Pniel	1013	M	L	5	1.366	26.5	incomplete	incomplete	-	-	-	-	-
Pniel	1011	M	L	5	1.396	29.0	incomplete	incomplete	-	-	-	-	-

A total of 18 pouched mice, comprising 3 males and 3 females from each locality, was studied. Equal proportions of these mice were derived from wild-caught and first generation laboratory-bred stocks. Laboratory-bred animals were reared under long photoperiod (14L:10D) at 25°C and were at least 4 months old at the beginning of the study. Wild-caught mice were maintained under the same conditions for at least 4 months before the experiments began.

Throughout the study the pouched mice were housed separately in standard laboratory rat cages and provided with rabbit pellets, rat cubes (Epol, Vereniging) and water *ad libitum*. Shredded paper and sawdust were provided for bedding and their cages were cleaned weekly, and dusted with insecticide to eradicate ectoparasites (Karbadust: Sentrachem, Silverton. Containing 50g/kg Carbamate).

Acclimation

Previous studies have shown that *S.campestris* undergo spontaneous torpor in response to cold (10°C) temperatures although more individuals display torpor following acclimation to short photoperiod than when acclimated to long photoperiod (unpublished data). In order to elicit torpor in as many individuals as possible the pouched mice were subjected to a series of artificial conditions in which first photoperiod and then temperature declined: 14L:10D & 25°C followed by 10L:14D & 25°C and then 10L:14D & 15°C. In this way the mice were gradually acclimated to short photoperiod and a temperature which was similar to the mean winter temperature of each locality (Table 1). The pouched mice spent 4 weeks under each set of conditions and remained healthy throughout the experiment.

Following acclimation to 15°C the oxygen consumption and body temperature of each pouched mouse was recorded at intervals over 3 1/2 days to establish whether spontaneous torpor occurred when ambient temperature (T_a) was gradually reduced from 20°C through 15°C and 10°C to 5°C.

Oxygen Consumption

Oxygen consumption (VO_2) was measured using an open-circuit system (as described by Depocas & Hart 1957, Hill 1972, and Withers 1977: Equation 3a) and assuming an RQ of 0.85. The mice were placed in large (13 l), perspex metabolic chambers, through which a flow of dried air (silica gel) was passed at a rate of 600ml·min^{-1}. The chambers were immersed in a constant temperature water bath (Labotec, Isando), and a thermocouple (J-Type: Grant Instruments, Cambridge) within the chamber was used for monitoring T_a. The water bath was covered by a light-tight hood and short photoperiod was maintained using an electric light connected to an automatic time switch.

The pouched mice were transferred to these chambers together with their bedding, food and water (fresh apple), 10h before measurements began so that they could become accustomed to their new surroundings. Measurements subsequently started in the middle of the dark phase (24h00) and continued for 3 1/2 days during which T_a was progressively lowered from 20°C to 5°C. In this way the VO_2 of each animal was recorded for 24h at T_a=20°C, T_a=15°C, & T_a=10°C and for 12h at T_a=5°C.

Following full equilibration of the system, the air leaving the chambers was dried and its oxygen content determined using an Ametek S-3A/I oxygen analyser (Applied Electrochemistry, Pittsburgh) connected to a multi-channel data logger (1200 Series Squirrel: Grant Instruments, Cambridge). An in-line, three way valve (Air/Water 350 KPA: Ascoreg, Johannesburg) and time switch alternated the input to the oxygen analyser so that air from two chambers, containing separate pouched mice, could be recorded twice an hour using a single oxygen analyser. The oxygen analyser was calibrated before and after each measurement and all readings were corrected to standard temperature and pressure, dry (STPD). Gross VO_2 was converted to mass specific VO_2 using the mean of body mass (M_b) measurements taken at the beginning and end of each 3 1/2 day run.

Average daily metabolic rate (ADMR) was calculated as the mean VO_2 measured over 24h at any one temperature. In this way ADMR was calculated at T_a=20°C, 15°C and 10°C but not at T_a=5°C because VO_2 was only recorded for 12h. In contrast, minimum metabolic rate (VO_{2min}) was taken as the lowest value of VO_2 sustained for at least 15 minutes at each of the four T_as.

Body Temperature

Body temperature (T_b) was determined using small (1.2g) temperature telemeters (Model X: Minimitter, Sunriver) which provided accurate temperature measurement to within 0.5°C. Sterilised telemeters were surgically implanted in the peritoneal cavity of anaesthetised (75mg/kg *i.p.* Sagatal: Maybaker, Port Elizabeth) pouched mice under veterinary supervision, and the mice were given at least 5 days to recover from surgery. The telemeters were calibrated prior to implantation and at the end of each experiment, during which time there was negligible drift of telemeter calibration.

Signals from implanted telemeters were received using small AM radios placed within the metabolic chambers. Unfortunately it wasn't possible to record T_b continually because background from laboratory equipment interfered with the reception of pulses from the minimitters. However, daily torpor usually takes place at a predictable time of day (Tucker 1965; Lynch and Gendler 1980) and previous studies have revealed that torpor cycles of pouched mice always occur during the first few hours of photophase (unpublished data). For this reason we recorded T_b manually using headphones once an hour following lights on between 07h00 and 12h00.

All results are presented as mean ± one standard deviation. Spearman's rank correlation coefficient (r_s - SCC), Mann-Witney U-test (U - MW), and two/three way analyses of variance with no repeated measures (F - 2/3-ANOVA) were used for statistical analysis of results (Sokal and Rohlf 1981).

RESULTS

Body Mass

The mean body mass (M_b) of pouched mice in the present study was 73.0 ± 10.5g (*Letaba*: M_b= 71.4 ± 8.4g, *Gam*: M_b= 71.8 ± 11.0g, *Pniel*: M_b= 75.9 ± 13.1g) and there was no significant effect of either locality (2-ANOVA, F= 0.310, d.f.= 2 & 12, p>0.05) or sex (F= 0.036, d.f.= 1 & 12, p>0.05) on body mass. Laboratory-bred pouched mice (M_b= 74.6 ± 8.8g) were slightly heavier than wild-caught individuals (M_b= 71.4 ± 12.4g) although this difference was not significant (MWU, U= 35,5, n_1=n_2= 9, p>0.05).

Metabolic rate

The average daily metabolic rate (ADMR) and minimum oxygen consumption (VO_{2min}) of all the pouched mice recorded at each T_a are summarised in Fig. 1. Both ADMR and VO_{2min} increased with decreasing temperature (3-ANOVA, ADMR F= 31.664, d.f.= 3 & 36, p<0.001 VO_{2min} F= 8.517, d.f.= 3 & 48, p<0.001) while female pouched mice displayed higher values of VO_{2min} than males (F= 5.784, d.f.= 1 & 48, p<0.05). In addition, there was an interactive effect of sex and locality on ADMR (F= 7.033, d.f.=2 & 36, p<0.05) such that female pouched mice from *Pniel* and *Gam* had higher values of ADMR than males while the reverse was true for pouched mice from *Letaba*.

Body temperature

In contrast to VO_2 the mean of minimum body temperatures (T_{bmin}) measured during the first 6 hours of photophase declined with decreasing temperature, from 34.0 ± 0.7°C at T_a=20°C, to 32.4 ± 3.3°C at 15°C and stabilising at 30.3 ± 4.4°C and 30.5 ± 3.3°C at T_a=10°C & 5°C respectively (3-ANOVA, F= 5.767, d.f.= 3 & 48, p<0.01). In addition male pouched mice displayed lower values of T_{bmin} than females at all T_as studied (F= 5.684, d.f.= 1 & 48, p<0.05). There was, however, no significant effect of locality on T_{bmin} (F= 1.607, d.f.= 2 & 48, p>0.05), and there was no significant correlation between body mass of pouched mice and their lowest Tb measured during the study (SCC, r_s=0.218, n=18, p>0.05).

Torpid Pouched Mice

A total of 8 pouched mice (2 *Letaba*, 3 *Gam* & 3 *Pniel*) displayed T_{bmin} values ≤ 30°C during the study and were considered to enter torpor. The ADMRs of these animals are displayed in Fig.s 2 (*Letaba*), 3 (*Gam*), and 4 (*Pniel*) which confirm that VO_{2min} always occurred during the first 6 hours of photophase, at the same time that T_{bmin} was recorded. Torpor cycles were characterised by a distinct decline in VO_2 for more than 2 h, ending in a sharp increase in VO_2 which overshot resting levels temporarily during arousal. No pouched mice entered torpor at T_a=20°C, whilst the incidence of torpor increased with declining T_a from 2 at T_a=15°C, to 6 at T_a=10°C, and 7 at T_a=5°C.

In order to quantify the duration and relative energetic savings for each bout of torpor, the metabolism of torpid individuals was compared to that of euthermic pouched mice from the same locality (see Fig.5). Torpor duration (D_t) was determined as the period during which the metabolism of torpid animals fell below the mean metabolism of non-torpid mice measured during photophase (the resting phase of *S.campestris*). In the same way the duration of overshoot (D_o) was calculated as the period in which the metabolism of the arousing animal exceeded the mean metabolism of non-torpid mice measured during photophase. The energy savings of each bout of torpor were determined by calculating the total oxgen consumption during torpor (T), the total oxygen consumption during overshoot (O), and the equivalent oxygen consumption of non-torpid individuals over the same period of time (N). The energy savings (N-[T+O]) were then expressed as a percentage of N. Finally, the daily energy savings associated with torpor were calculated by subtracting the ADMR of torpid pouched mice from the mean ADMR of non-torpid animals and expressing the difference as a percentage of euthermic ADMR. These data, together with T_{bmin} and VO_{2min}, are summarised in Table 2.

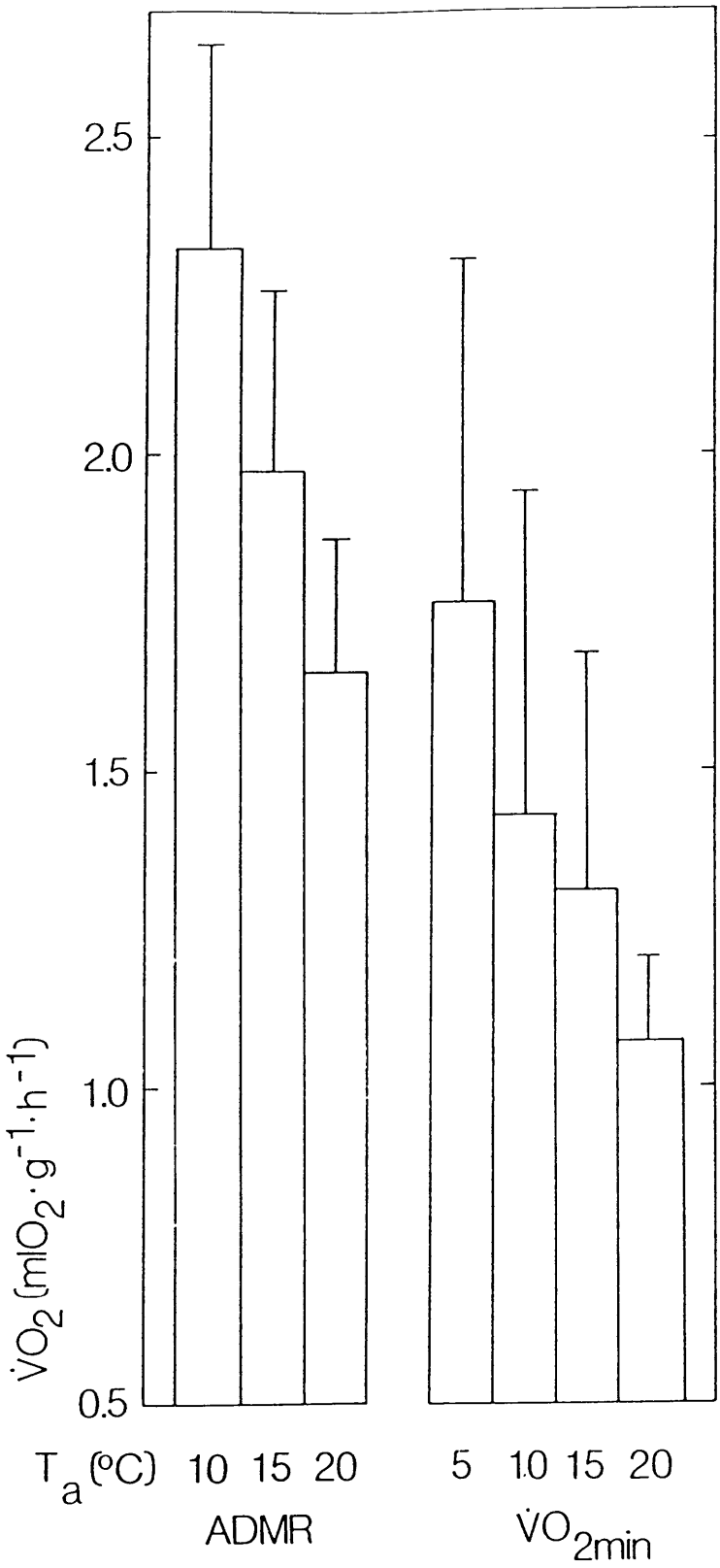

Figure 1. Mean oxygen consumption (VO_2, $mlO_2 \cdot g^{-1} \cdot h^{-1}$) of all pouched mice measured during 24h exposure to T_a = 20°C, 15°C & 10°C, and 12h exposure to 5°C. ADMR refers to the mean VO_2 recorded at each T_a.VO_{2min} refers to the mean minimum oxygen consumption measured at each T_a.

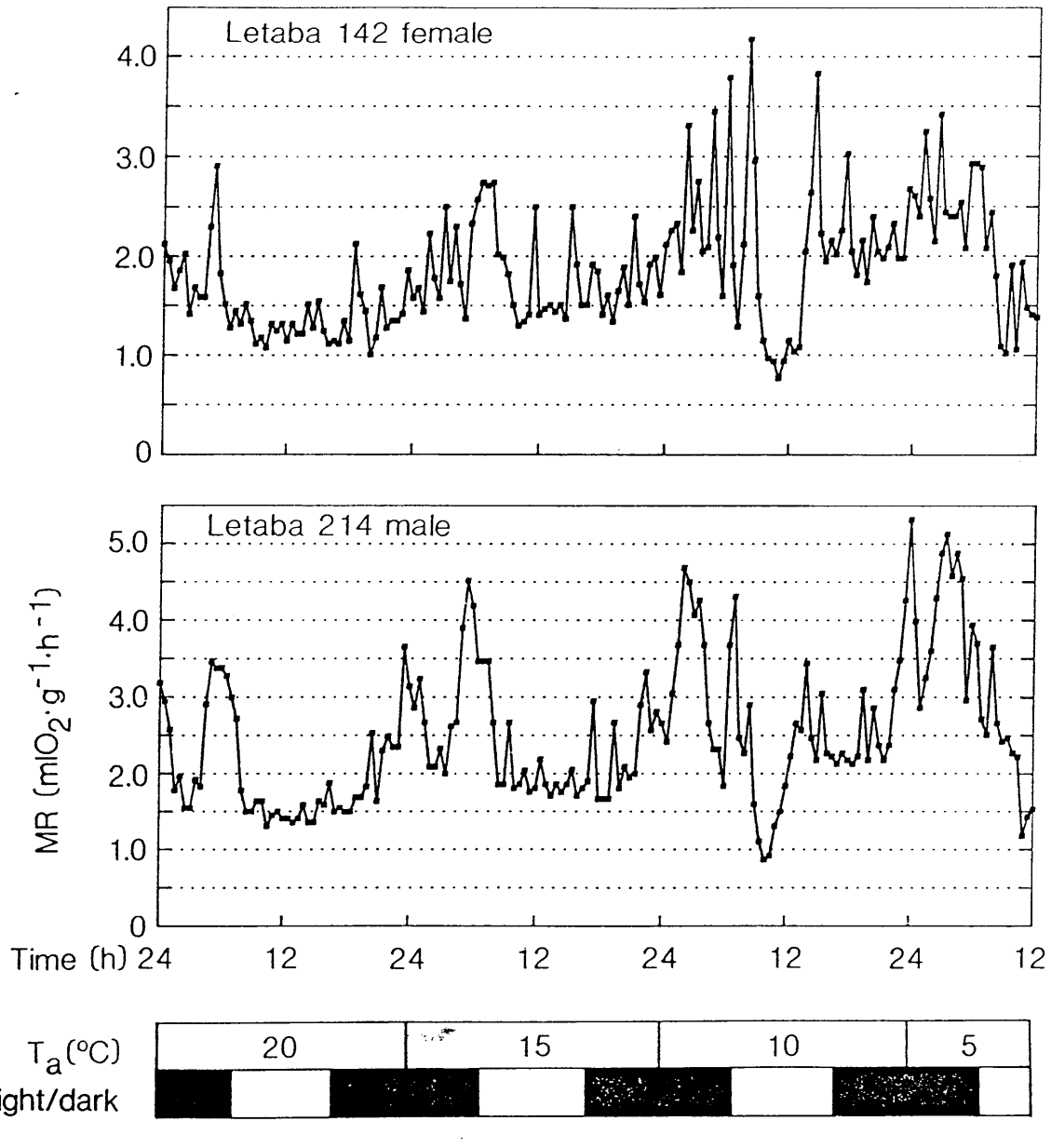

Figure 2. Oxygen consumption ($mlO_2 \cdot g^{-1} \cdot h^{-1}$) of 2 pouched mice from *Letaba* which exhibited torpor cycles during exposure to declining ambient temperature (T_a). Periods of light and dark, and T_a are indicated at the bottom of the Figure.

Letaba 142 displayed torpor at both T_a = 10°C & 5°C.

Letaba 214 displayed torpor at both T_a = 10°C & 5°C.

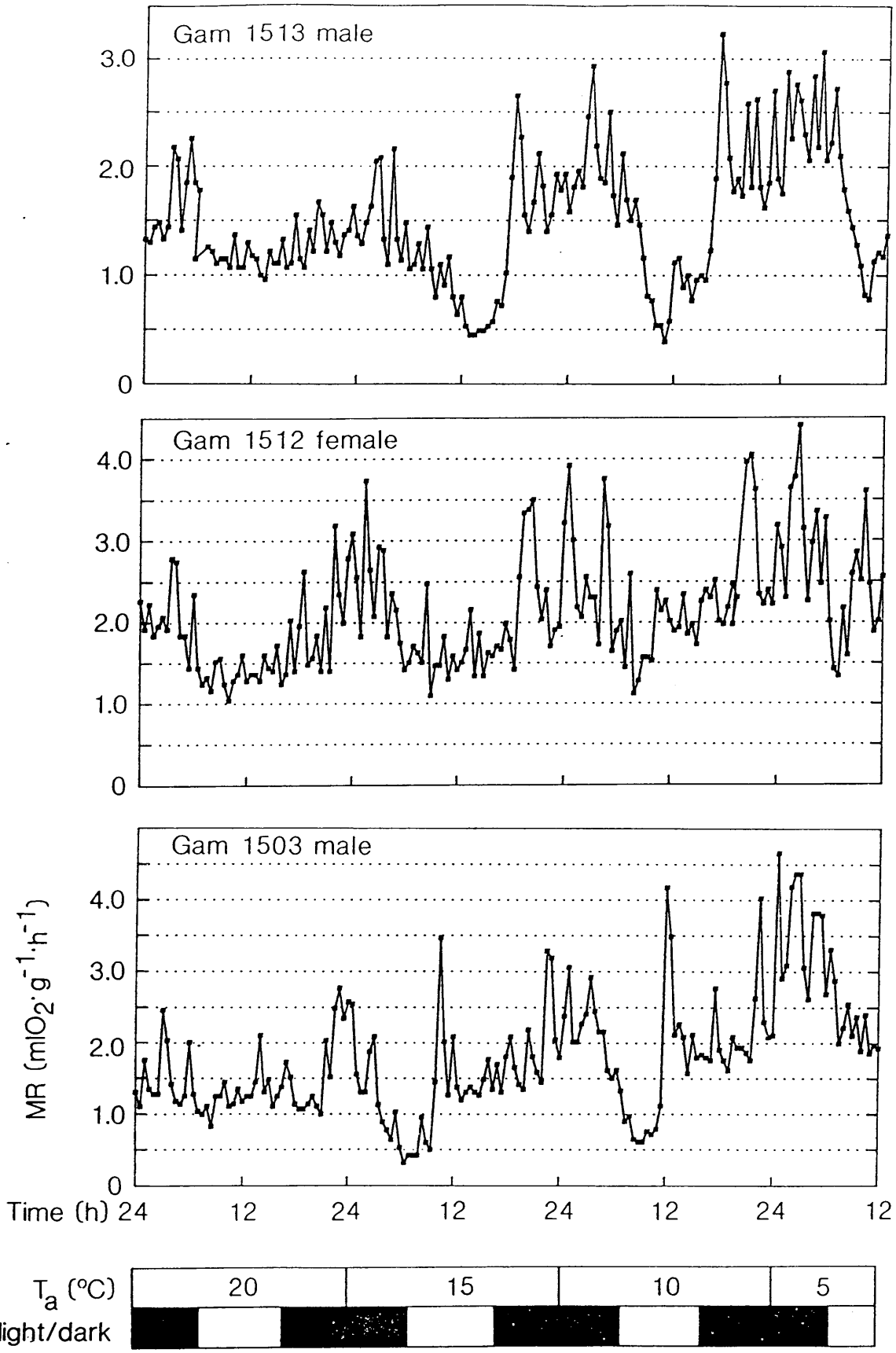

Figure 3. Oxygen consumption ($mlO_2 \cdot g^{-1} \cdot h^{-1}$) of 3 pouched mice from *Gam* which exhibited torpor cycles during exposure to declining ambient temperature (T_a). Periods of light and dark, and T_a are indicated at the bottom of the Figure.

Gam 1513 displayed torpor at T_a = 15°C, 10°C & 5°C.
Gam 1512 displayed torpor at both T_a = 10°C & 5°C.
Gam 1503 displayed torpor at both T_a = 15°C & 10°C.

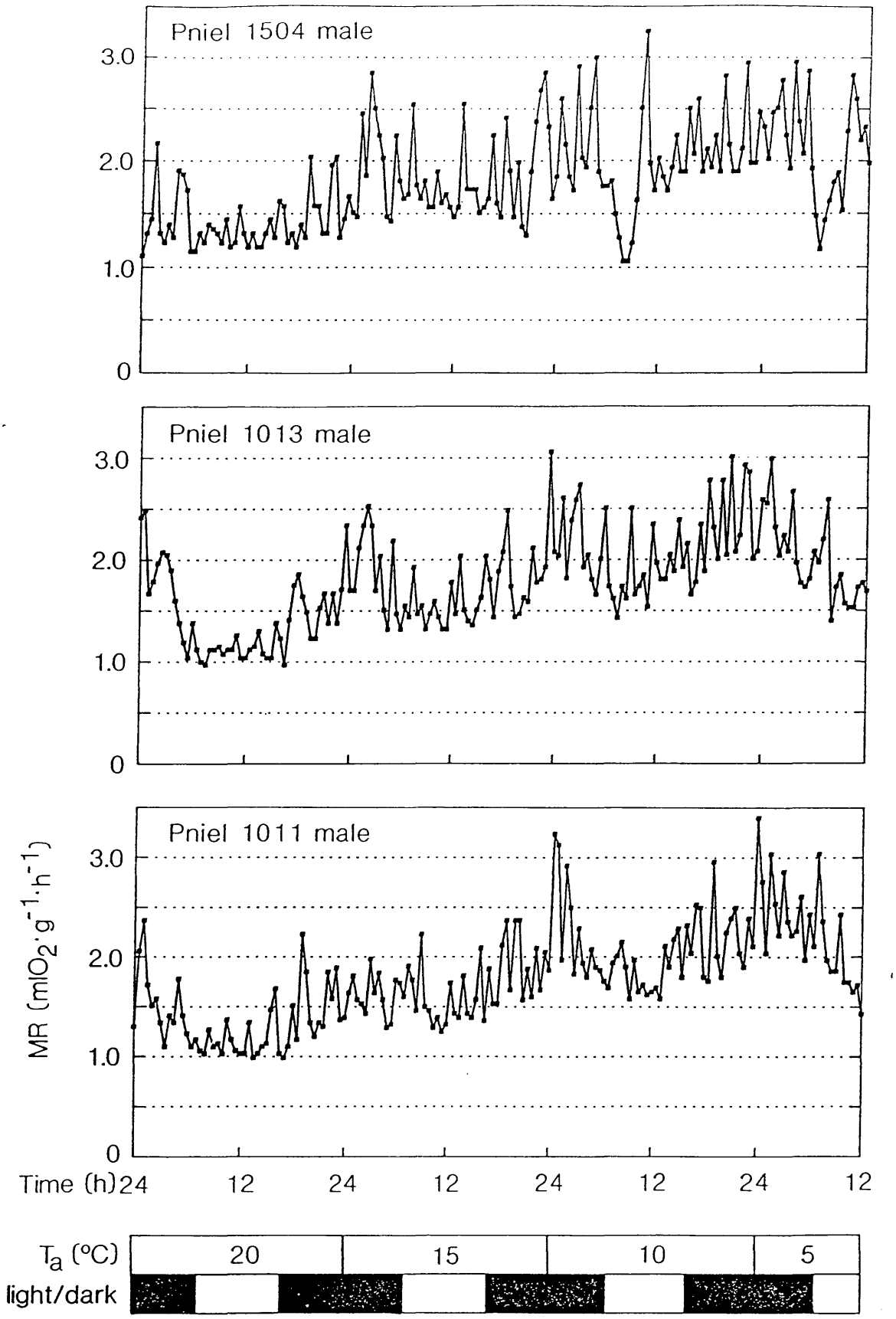

Figure 4. Oxygen consumption ($mlO_2 \cdot g^{-1} \cdot h^{-1}$) of 3 pouched mice from *Pniel* which exhibited torpor cycles during exposure to declining ambient temperature (T_a). Periods of light and dark, and T_a are indicated at the bottom of the Figure.

Pniel 1504 displayed torpor at both T_a = 10°C & 5°C.

Pniel 1013 displayed torpor at T_a = 5°C.

Pniel 1011 displayed torpor at T_a = 5°C.

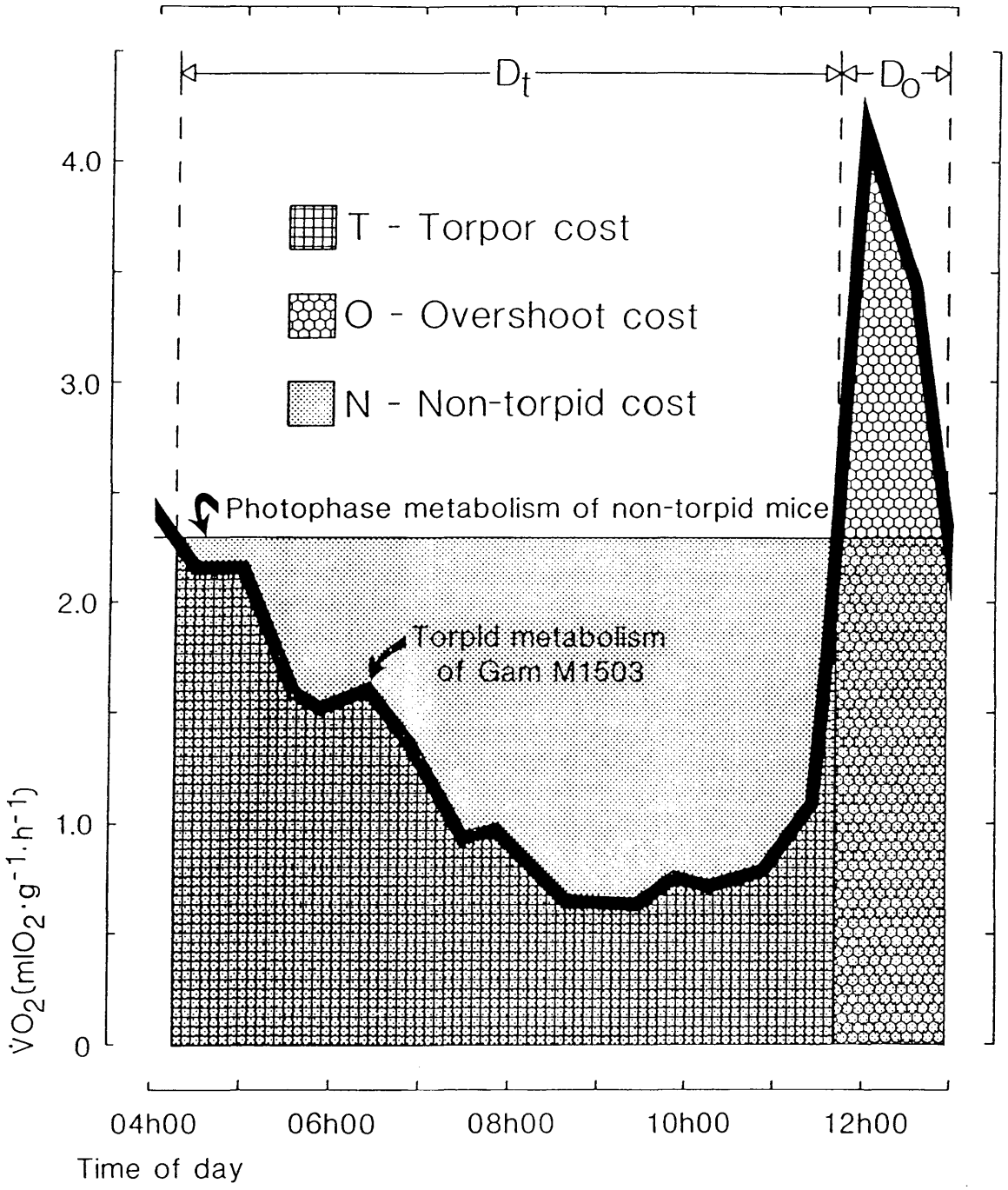

Figure 5. Calculating the duration of torpor (D_t), the duration of overshoot (D_o), the cost of torpor (T), the cost of overshoot (O) and the equivalent cost of non-torpid individuals during the same period (N) by comparing the oxygen consumption of a torpid individual (*Gam* 1503 at 10°C) to the mean oxygen consumption of non-torpid animals recorded during photophase.

Comparing VO_{2min} to the T_{bmin} of torpid pouched mice (Fig.6) reveals that mean T_{bmin} remained relatively constant as T_a declined whilst VO_{2min} increased steadily with falling T_a. At T_a=10°C, VO_{2min} was positively correlated with T_{bmin} (SRC, r_s= 0.943, n=6, p<0.01) and inversely correlated with the duration of individual torpor bouts (r_s= -0.943, n=6, p<0.01). There was also a perfect correlation between the savings associated with each bout of torpor and daily energy savings (r_s= 1.000). However, there was no significant correlation between these savings and either VO_{2min} (r_s= -0.600, n=6, p>0.05), T_{bmin} (r_s= -0.714, n=6, p>0.05), nor was there a correlation between energy savings and torpor duration (r_s= 0.657, n=6, p>0.05) or the cost of the arousal overshoot (r_s= -0.371, n=6, p>0.05).

Finally, it is clear from Table 2 that pouched mice from *Gam* entered torpor at higher T_as, experienced the longest bouts of torpor with the lowest T_bs, and accrued the greatest energetic savings associated with torpor. In contrast, there was no clear distinction between pouched mice from *Letaba* and *Pniel* with respect to incidence, duration, depth or energetic savings of torpor. As regards sexual differences, only 2 females entered torpor compared to 6 males, whilst of the 8 animals entering torpor, an equal proportion were derived from laboratory-bred and wild-caught stocks.

DISCUSSION

It is evident from the present study that *S.campestris* from throughout southern Africa respond to decreasing ambient temperature by increasing metabolic heat production, with or without spontaneous bouts of torpor. Even those pouched mice which remained euthermic throughout the study displayed a high degree of thermolability and lowered their minimum body temperature by 2-3°C as ambient temperature fell from 20°C to 5°C. In contrast, torpid pouched mice generally exhibited body temperatures well below 30°C although, in common with other torpid rodents, there was considerable variation in the extent to which T_b fell (Buffenstein 1985) and T_{bmin} during torpor ranged between 30.0°C and 21.0°C. However, the mean Tbmin of torpid individuals was independent of T_a whilst the minimum metabolic rate during torpor increased with decreasing ambient temperature. This suggests that T_{bmin} was under strict thermoregulatory control during torpor and that metabolic heat production increased to maintain Tb as ambient temperature fell.

The incidence of torpor increased as T_a declined which indicates that torpidity was elicited in response to rising energetic costs (Gaertner *et al.* 1973; Lynch *et al.* 1978). However, pouched mice only entered torpor at temperatures below those prevailing in their winter burrows (Table 1) and therefore it appears that spontaneous torpor might be unusual in nature, and only occurs under cold stress. Meanwhile, even at the lowest temperature, T_a= 5°C, only seven of the eighteen pouched mice exhibited torpor. Indeed, torpor is but one of a suite of possible adaptations to cope with the energetic cost of cold exposure (Wunder 1984) and among species which employ daily torpor not all individuals have the capacity to do so (Hudson 1978). Instead these individuals display alternative mechanisms such as a change in activity or food intake (Müller *et al.* 1988, Ruf *et al.* 1991).

The duration of torpor is thought to be related to the build-up of metabolic wastes or the depletion of metabolic substrates (Twente & Twente 1965). Both these factors are determined by the rate of metabolism, which might explain why the duration of torpor at T_a=10°C was inversely correlated with the VO_{2min} of pouched mice. However, it remains unclear why neither VO_{2min}, T_{bmin}, nor torpor duration were correlated with the energetic savings associated with torpor. In part this might be due to the assumption that the metabolism of torpid pouched mice would have been the same as that of euthermic individuals had torpor not occurred. As one can never accurately predict what the euthermic metabolism of a torpid animal should be (Hill 1975), calculating the energy savings accrued during torpor by comparing the metabolism of euthermic and torpid animals might be unrealistic.

The sexual differences in metabolism described in the present study are similar to those reported for *Mesocricetus brandti* by Lyman *et al.* (1983), for *Phodopus sungorus* by Elliott *et al.* (1987) and Ruf *et al.* (1991) and for *Tarsipes rostratus* by Withers *et al.* (1990). All these studies reported that females were less likely to enter torpor than males, which infers that selection operates in favour of males which display reduced metabolic expenditure during cold conditions. Alternatively, these sexual differences might simply reflect contrasting thermoregulatory abilities in the cold, as shown for female Wistar rats which responded to cold stress more efficiently than males by producing less heat and having better body insulation (Doi & Kuroshima 1982).

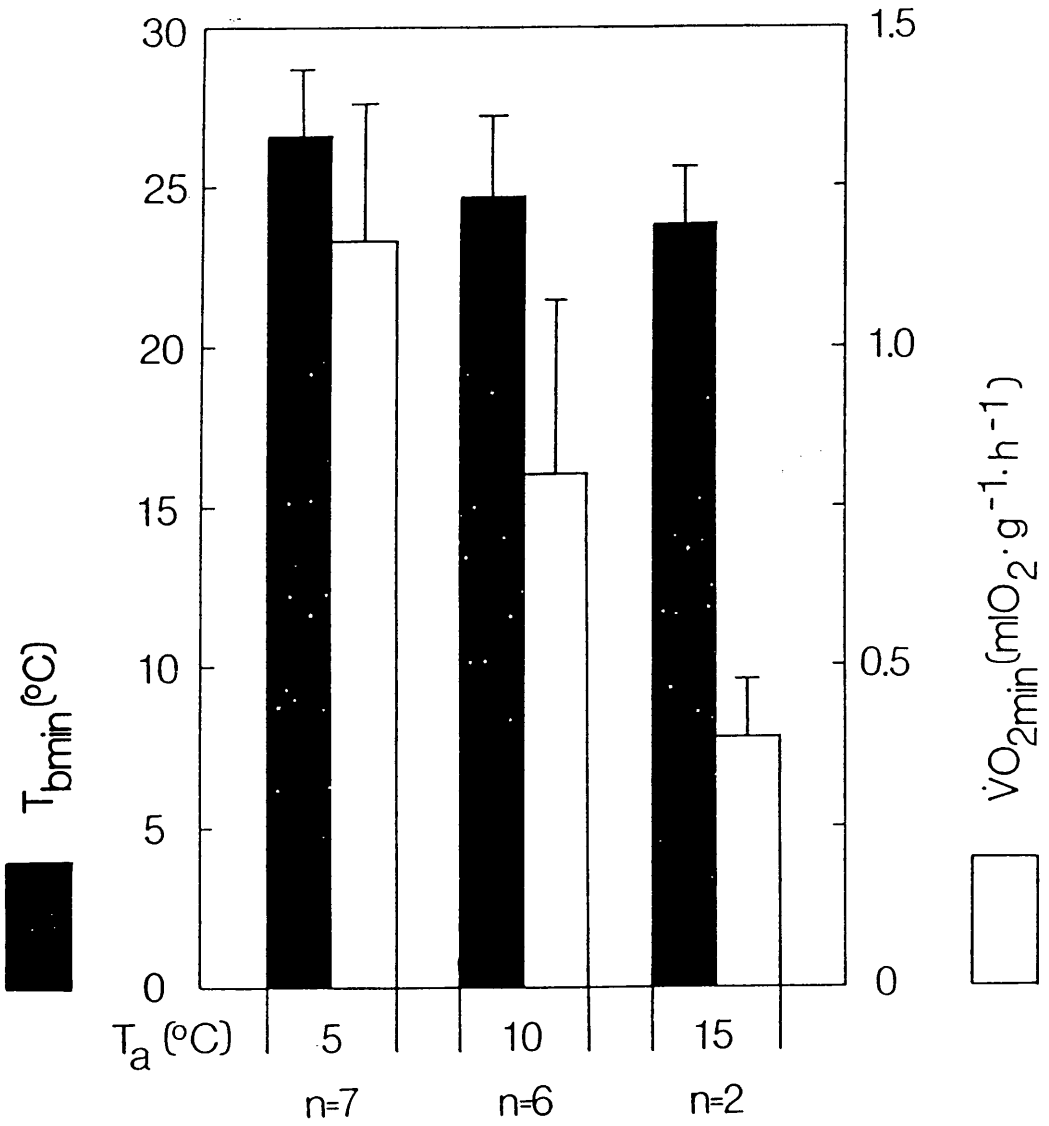

Figure 6. Minimum oxygen consumption (VO_{2min}, $mlO_2 \cdot g^{-1} \cdot h^{-1}$) and minimum body temperature (T_{bmin}, °C) recorded during torpor cycles of pouched mice at T_a= 15°C, 10°C & 5°C.

For the study on *T.rostratus* the sexual differences in torpor use appear to be based upon differences in body mass (Withers *et al.* 1990) but there was no sexual dimorphism evident in the present study, nor in those of Lyman *et al.* (1983), Elliott *et al.* (1987) and Ruf *et al.* (1991). There was also no correlation between the body mass of pouched mice and the lowest T_b recorded in the present study.

Although too few pouched mice became torpid in the present study to assess whether geographical differences exist, there was clearly a tendency for individuals from *Gam* to enter deeper, longer bouts of torpor at higher T_a's than those from either *Letaba* or *Pniel*. At the same time, equal numbers of laboratory-bred and wild-caught pouched mice entered torpor which suggests that spontaneous torpor in *S.campestris* is an heritable character which is not dependent upon prior exposure to specific conditions prevailing in their natural environment.

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REFERENCES

Bronson F.H. (1991) Susceptibility of the fat reserves of mice to natural challenges. *J. Comp. Physiol. B* **157**, 551-554.

Brown J.H. and Bartholomew G.A. (1969) Periodicity and energetics of torpor in the kangaroo mouse, *Microdipodops pallidus. Ecology* **50**, 705-709.

Buffenstein R. (1985) The effect of starvation, food restriction and water deprivation on thermoregulation and average daily metabolic rates in *Gerbillus pusillus. Physiol. Zool.* **58**, 320-328.

Depocas F. and Hart J. S. (1957) Use of the Pauling oxygen analyser for measurements of oxygen consumption of animals in open-circuit system and in short-lag, closed-circuit apparatus. *J. Appl. Physiol.* **10**, 388-392.

Doi K. and Kuroshima A. (1982) Sexual difference in thermoregulatory ability of rats exposed to cold and heat. *J. therm. Biol.* **7**, 99-105.

Earl S.M.J.A. (1980) Development of behaviour in the pouched mouse, *Saccostomus campestris*. Unpubl. MSc Thesis, University of Pretoria.

Elliott J.A., Bartness T.J. and Goldman B.D. (1987) Role of short photoperiod and cold exposure in regulating daily torpor in Djungarian hamsters. *J. Comp. Physiol. A* **161**, 245-253.

Gaertner R.A., Hart J.S. and Roy O.Z. (1973) Seasonal spontaneous torpor in the white-footed mouse, *Peromyscus leucopus. Comp. Biochem. Physiol.* **45A**, 169-181.

Geiser F. (1988) Reduction of metabolism during hibernation and daily torpor in mammals and birds: temperature effect or physiological inhibition? *J. Comp. Physiol. B* **158**, 25-37.

Haim A., Ellison G.T.H. and Skinner J.D. (1988) Thermoregulatory circadian rhythms in the pouched mouse (*Saccostomus campestris*). *Comp. Biochem. Physiol.A* **91**, 123-127.

Heldmaier G. and Steinlechner S. (1981) Seasonal pattern and energetics of short daily torpor in the Djungarian hamster *Phodopus sungorus. Oecologia (Berlin)* **48**, 265-270.

Hill R.W. (1972) Determination of oxygen consumption by use of the paramagnetic oxygen analyser. *J. Appl. Physiol.* **33**, 261-263.

Hill R.W. (1975) Daily torpor in *Peromyscus leucopus* on an adequate diet. *Comp. Biochem. Physiol.* **51A**, 413-423.

Hudson J.W. (1978) Shallow daily torpor: a thermoregulatory adaptation. In: Wang L.C.H. and Hudson J.W. (eds), Strategies in cold. Academic Press, New York San Francisco London, p 67-108.

Hudson J.W. and Scott I.M. (1979) Daily torpor in the laboratory mouse, *Mus musculus var. albino. Physiol. Zool.* **52**, 205-218.

Lyman C.P., O'Brien R.C. and Bossert W.H. (1983) Differences in tendency to hibernate among groups of Turkish hamsters (*Mesocricetus brandti*). *J. therm. Biol.* **8**, 255-257.

Lynch G.R. and Gendler S.L. (1980) Multiple responses to different photoperiods occur in the mouse, *Peromyscus leucopus*. *Oecologia (Berlin)* **45**, 318-321.

Lynch G.R., Vogt F.D. and Smith H.R. (1978) Seasonal study of spontaneous daily torpor in the white-footed mouse, *Peromyscus leucopus*. *Physiol. Zool.* **51**, 289-299.

Maclean G.S. (1981) Torpor patterns and microenvironment of the eastern chipmunk, *Tamias striatus*. *J. Mamm.* **62**, 64-73.

Müller E.F., Pfender P. and Beck M. (1988) The benefits of hunger-induced torpor in gerbils and spiny mice. *Veröffentlichungen der Universität Innsbruck* **167**, 52.

Ruf T., Klingenspor M., Preis H. and Heldmaier G. (1991) Daily torpor in the Djungarian hamster (*Phodopus sungorus*): interactions with food intake, activity and social behaviour. *J. Comp. Physiol. B* **160**, 609-615.

Sokal R.R. and Rohlf F.J. (1981) Biometry. W H Freeman & Co, San Francisco.

Thompson S.D. (1985) Subspecific differences in metabolism, thermoregulation, and torpor in the western harvest mouse, *Reithrodontomys megalotis*. *Physiol. Zool.* **58**, 430-444.

Tucker V.A. (1965) The relation between the torpor cycle and heat exchange in the California pocket mouse, *Perognathus californicus*. *J. Cell. Comp. Physiol.* **65**, 405-414.

Twente J.W. and Twente J.A. (1965) Effects of core temperature upon duration of hibernation of *Citellus lateralis*. *J. Appl. Physiol.* **20**, 411-416.

Withers P.C. (1977) Measurement of VO_2, VCO_2, and evapourative water loss with a flow-through mask. *J. Appl. Physiol.* **42**, 120-123.

Withers P.C., Richardson K.C. and Wooller R.D. (1990) Metabolic physiology of euthermic and torpid honey possums, *Tarsipes rostratus*. *Aust. J. Zool.* **37**, 685-693.

Wunder B.A. (1984) Strategies for, and environmental cueing mechanisms of, seasonal changes in thermoregulatory parameters of small mammals. In: Merritt JF (ed) Winter ecology of small mammals. *Carnegie Mus. Nat. Hist. Spec. Publ.* **10**, 165-172.

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Ringtail in the pouched mouse (*Saccostomus campestris*)

G. T. H. ELLISON & L. M. WESTLIN-VAN AARDE

Mammal Research Institute, Pretoria University, Pretoria 0002, Republic of South Africa

Summary

Laboratory colonies of the pouched mouse (*Saccostomus campestris*) were housed in solid bottom cages and fed a varied diet containing excess fatty acids. Ringtail was only initiated in animals of all ages, from populations originating from different areas of South Africa, when the relative humidity fell below 30%. The incidence of ringtail was curtailed by maintaining relative humidity above 45% in animal houses.

Keywords: Ringtail; Pouched mouse; Relative humidity; Environmental control; Laboratory animal housing

Ringtail has been reported in three species of rodents maintained under laboratory conditions; the Norwegian rat, *Rattus norvegicus* (Njaa *et al.*, 1957), the house mouse, *Mus musculus* (Nelson, 1960) and the white-tailed hamster, *Mystromys albicaudatus* (Stuhlmann & Wagner, 1971). The incidence of the condition increases as relative humidity falls below 40% (Njaa *et al.*, 1957; Totton, 1958), although Flynn's (1967) study of ringtail in *R. norvegicus* suggested that the condition is only present in young rats (<9 days old) when these are housed in cages with wire mesh bottoms at relative humidities below 60%. The addition of 5% corn-oil (which has a high content of unsaturated fatty acids) to the rats' diet delayed the onset of ringtail and hastened recovery, whilst some strains of laboratory rats (Wistar, Sprague–Dawley) were more susceptible to ringtail than others (Long–Evans, Sherman) (Flynn, 1967).

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The present report concerns the incidence of this condition in a fourth species, the pouched mouse, *Saccostomus campestris* (Rodentia, Cricetomyinae). *S. campestris* is a docile animal, well suited for use in the laboratory (Davis, 1963) and has been employed in bilharzia research in southern Africa for several years (Pitchford & Visser, 1970).

Materials and methods

Pouched mice were housed separately in polypropylene rat cages with solid bases (Labotec, Isando). They were provided with rat pellets (Epol, Vereniging) and water *ad libitum* and the diet was occasionally supplemented with sunflower seeds, apples, carrots and lettuce. Shredded paper and sawdust (pine and meranti shavings) were provided for bedding and the cages were cleaned twice weekly.

Laboratory bred and wild-caught pouched mice were maintained under the following conditions:

Breeding Colony. 14L : 10D at 22±2°C. Relative humidity was not controlled, and ranged between <25% (winter) and >45% (summer)
Wild-caught animals. 14L : 10D at 25±2°C. Relative humidity controlled at either <30% *or* >45%
Wild-caught animals. 10L : 14D at 10±2°C. Relative humidity controlled at either <30% *or* >45%.

The incidence of ringtail was monitored continuously in all three groups of pouched mice.

Results and discussion

Over 50% of pouched mice in the breeding colony developed ringtail during winter when the

relative humidity fell below 25%. In contrast, ringtail was not observed during summer when the mean relative humidity exceeded 40%.

The wild pouched mice were collected from throughout South Africa (Pofadder, Rustenburg, Satara, Beaufort West, and Kimberley), and these populations differ genetically (Gordon, 1986). However, all five populations developed ringtail after 2 months in captivity when housed at a relative humidity of less than 30%. Furthermore, the wild pouched mice in group 3, maintained under short photoperiod at 10°C, were more susceptible to ringtail than those in group 2 under long photoperiod at 25°C. Both adult and juvenile pouched mice displayed the condition and affected animals lost up to 80% of their tail length, although they remained healthy in all other respects.

The incidence of ringtail was curtailed by maintaining the relative humidity above 45% in animal houses and the condition was not observed in any of the three groups maintained at this RH irrespective of photoperiod or temperature.

In contrast to Flynn's (1967) report of ringtail in *R. norvegicus*, the present study suggests that ringtail in *S. campestris* was caused by low relative humidity alone as the condition was apparent in animals of all ages from a variety of different populations when housed in cages with solid bottoms. However, the effect of diet on the incidence of ringtail in the pouched mouse requires further research as components of the diet used in this study (Epol rat pellets and sunflower seeds) contain ample quantities of fatty acids.

Conclusion

We recommend that pouched mice should be held in captivity at a relative humidity in excess of 45%.

Acknowledgments

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References

Davis DHS (1963) Wild rodents as laboratory animals and their contribution to medical research in South Africa. *South African Journal of Medical Science* **28**, 53–69

Flynn RJ (1967) Notes on ringtail in rats. In *Husbandry of Laboratory Animals*, 3rd International Symposium of the International Committee on Laboratory Animals (ed. ML Conalty), pp. 285–288. London & New York: Academic Press

Gordon DH (1986) Extensive chromosomal variation in the pouched mouse, *Saccostomus campestris* (Rodentia, Cricetidae) from southern Africa: A preliminary investigation of evolutionary status. *Cimbebasia* **8**, 37–47

Nelson JB (1960) The problems of disease and quality in laboratory animals. *Journal of Medical Education* **35**, 34–43

Njaa LR, Utne F & Braeckkan OR (1957) Effect of relative humidity on rat breeding and ringtail. *Nature* **180**, 290–291

Pitchford RJ & Visser PS (1970) *Saccostomus campestris* Peters, 1846 as a laboratory animal. *Journal of the South African Veterinary Medical Association* **41**, 173–174

Stuhlmann RA & Wagner JE (1971) Ringtail in *Mystromys albicaudatus*. A case report. *Laboratory Animal Science* **21**, 585–587

Totton M (1958) Ringtail in newborn Norway rats—A study of the effect of environmental temperature and humidity on incidence. *Journal of Hygiene* **56**, 190–196

Journal of Zoology, London (*in press*).

Is the annual cycle in body weight of pouched mice (*Saccostomus campestris*) the result of seasonal changes in adult size or population structure?

G.T.H. ELLISON*, G.N. BRONNER**, P.J. TAYLOR***

*Mammal Research Institute, Pretoria University, Pretoria 0002,

**Department of Mammals, Transvaal Museum, Pretoria 0002, and

***Durban Natural Science Museum, Durban 4001,

Republic of South Africa

(With 4 figures in the text)

The annual cycle in body weight of pouched mice (*Saccostomus campestris*) was examined among 104 specimens which were collected throughout the year in the Transvaal province of South Africa. Each specimen was assigned to one of 5 age classes using toothwear characteristics although none of them belonged to the youngest age class. There was no significant effect of sex on body weight but older individuals were significantly heavier than younger ones. Pooled data from both sexes displayed seasonal variation in body weight with significantly heavier animals in the wet season (December-March) than during the dry season (June-September). However, there was no significant difference between the age structure of the population at these times. Instead, pouched mice in the two younger age classes were significantly lighter in the dry season compared to the wet season while older individuals maintained a relatively constant body weight throughout the year. These results suggest that the annual cycle in body weight of *S.campestris* is caused by a reduction in body weight of young animals which lose weight in winter as an adaptation to limit their energy requirements when food availability declines.

Contents

Introduction

Despite the increase in metabolic rate which accompanies a reduction in body size, small individuals are at an energetic advantage when compared to larger conspecifics because they have lower total energy requirements (Wunder *et al.* 1977). For this reason many small mammals inhabiting strongly seasonal environments display a reduction in mean body weight during winter and lower their energy requirements at a time when food availability declines (eg. Fuller *et al.* 1969; Iverson & Turner 1974; Wunder *et al.* 1977; Petterborg 1978; Heldmaier & Steinlechner 1981; Stewart & Barnett 1983; Murua *et al.* 1987; Korn 1989; Bozinovic *et al.* 1990). Although it is tempting to suggest that this reduction in body weight represents an adaptation to survive periods of food restriction, only a few of these studies have determined that individual animals lose weight during winter (Iverson & Turner 1974; Petterborg 1978; Heldmaier & Steinlechner 1981; Stewart & Barnett 1983; Murua et al 1987). In each of the other studies the winter decline in mean body weight might simply occur as a fortuitous result of seasonal variation in breeding activity because large, pregnant females would increase the average weight of the population during the breeding season, while an influx of small, young animals would lower the mean body weight at the end of the breeding season and during winter. The aim of the present study was to investigate the adaptive significance of seasonal changes in the body weight of pouched mice (*Saccostomus campestris*), a wet season breeder (Smithers 1983) which displays a significantly lower mean body weight during winter (Korn 1989).

Materials and methods

The annual cycle in body weight of pouched mice (*Saccostomus campestris*) was examined using specimens from the small mammal collection at the Transvaal Museum in Pretoria. The present study was restricted to individuals collected within the Transvaal in order to minimise any geographical effects on body weight (Korn 1989). At the same time, 12 specimens weighing less than 60% of the average weight given by Rautenbach (1982) were excluded so that the present study was directly comparable with Korn's (1989). Likewise, any females were excluded from the analysis if the catalogue indicated that they were pregnant when caught. This left a total of 104 (47 male and 57 female) pouched mice for which body weight and collection date were recorded. These were subsequently allocated to one of five tooth-wear classes (TW1-5) in order to provide a relative estimate of their ages (see Fig. 1).

All results are presented as mean $\pm$ one standard deviation. Two-way analyses of variance (ANOVA), together with Mann-Witney U (MWU) and Chi-squared (X^2) tests were used for the statistical analysis of results (Sokal and Rohlf 1981).

Results

There was no significant difference between the mean body weight (M_b) of male (52.0 $\pm$15.6g) and female (50.6 $\pm$14.0g) pouched mice examined in the present study (ANOVA: F= 0.0155, d.f.= 1 & 108, p>0.05). Although none of these were classified in the youngest age class (TW1) neither were any of the 12 excluded specimens. There was a significant effect of age on M_b (see Fig. 2) such that older pouched mice were significantly heavier than younger ones (ANOVA: F= 12.640, d.f.= 3 & 108, p<0.001). Pooled data for both sexes displayed a clear annual cycle in body weight (see Fig. 3) and individuals collected during the wet season (December-March: M_b= 55.7 $\pm$15.1g) were significantly heavier than those caught during the dry season (June-September: M_b= 49.5 $\pm$14.3g) (MWU: U= 785, n_1= 28, n_2= 44, p<0.05). This might have been the result of seasonal variation in the age structure of the population (see Fig. 4) because there was a peak in the number of young (TW2) animals in October and November followed by a progressive increase in the proportion of older individuals from December through to September when the two older age classes (TW4 & TW5) comprised 77% of the population. However, there was no significant difference in age structure between wet and dry seasons (X^2= 6.89, d.f.= 3, p>0.05). Instead, it appears that the annual cycle in body weight was caused by a reduction in the body weight of smaller individuals because the two younger age classes (TW2 & TW3) were significantly lighter in the dry season compared to the wet season (Table I).

TABLE I. *Variation in the mean body weight of different age classes of pouched mice between the wet season (December-March) and the dry season (June-September)*

Toothwear/ Age Class	Wet Season M_b (g)	Dry Season M_b (g)	Mann Witney U-test (U)	Significance level (p)
TW2	46.1± 6.8 (n=10)	36.0± 2.6 (n= 5)	45.5	<0.02
TW3	56.1±13.0 (n=17)	41.2± 9.2 (n= 5)	71.5	<0.05
TW4	61.7± 9.1 (n=10)	57.8±14.6 (n=12)	86.0	>0.05
TW5	59.9±24.7 (n= 7)	51.0± 8.1 (n= 6)	18.5	>0.05

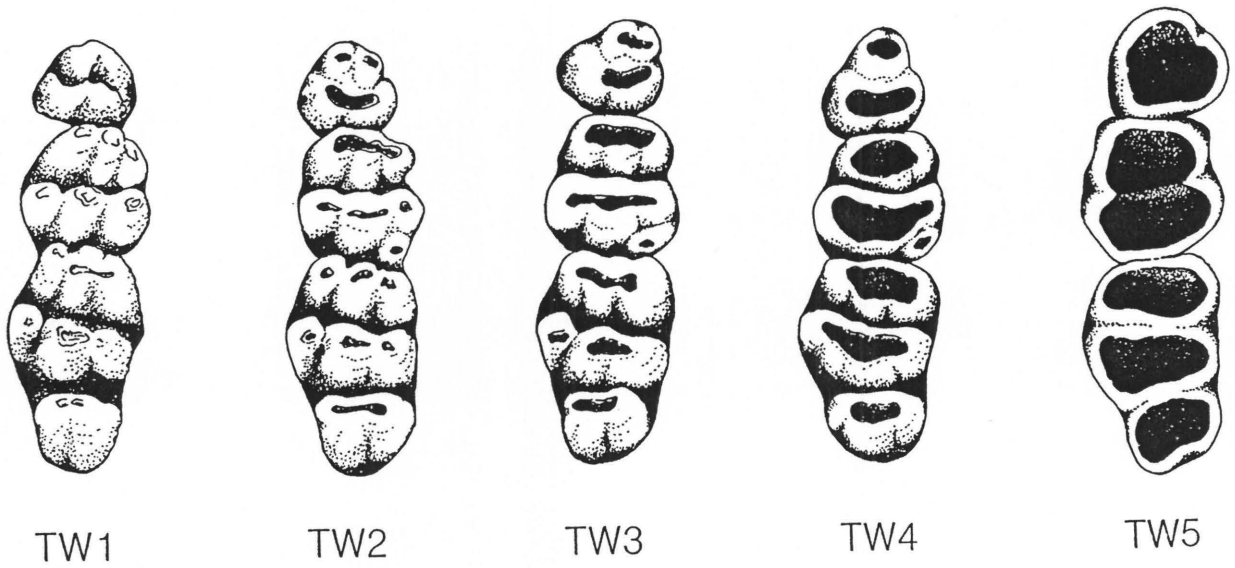

FIG. 1. Typical appearance of right upper toothrow in representative specimens from five toothwear/age classes collected at a single locality (northeast Zululand):

Toothwear Class 1 (TW1) - M^3 unerupted or not fully erupted.

Toothwear Class 2 (TW2) - all cheekteeth fully erupted; wear minimal and restricted to individual cusps.

Toothwear Class 3 (TW3) - cheekteeth moderately worn with cusps bridged by exposed dentine along laminar occlusal surfaces.

Toothwear Class 4 (TW4) - cheekteeth well worn with all cusps fused along continuous laminar surfaces; lamellae of M^3 pressed together; enamel borders between lamellae breaking down.

Toothwear Class 5 (TW5) - cheekteeth severely worn; lamellae closely pressed together in all cheekteeth; occlusal surfaces worn smooth due to complete removal of enamel ridges separating lamellae.

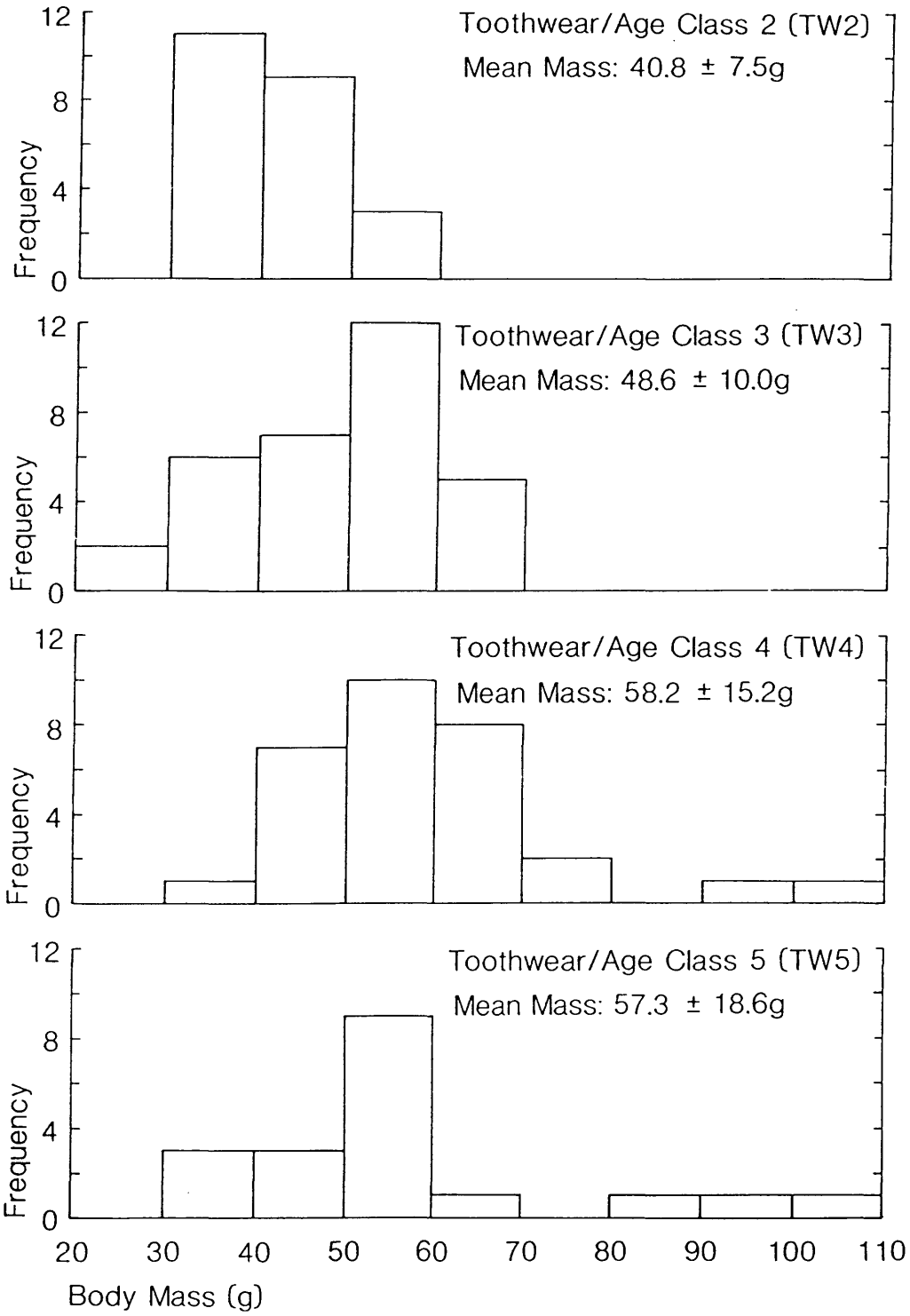

FIG. 2. The distribution of body weight among toothwear/age classes of *S.campestris* from the Transvaal.

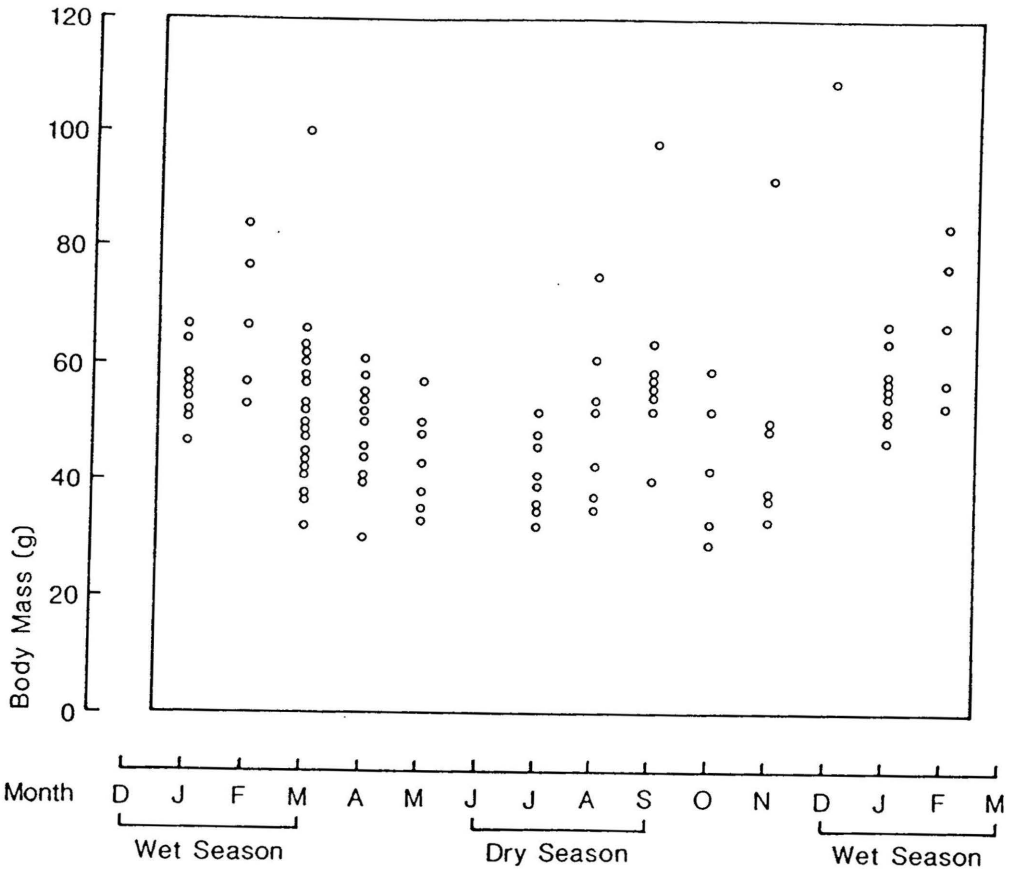

FIG. 3. The annual cycle of body weight in *S.campestris* from the Transvaal.

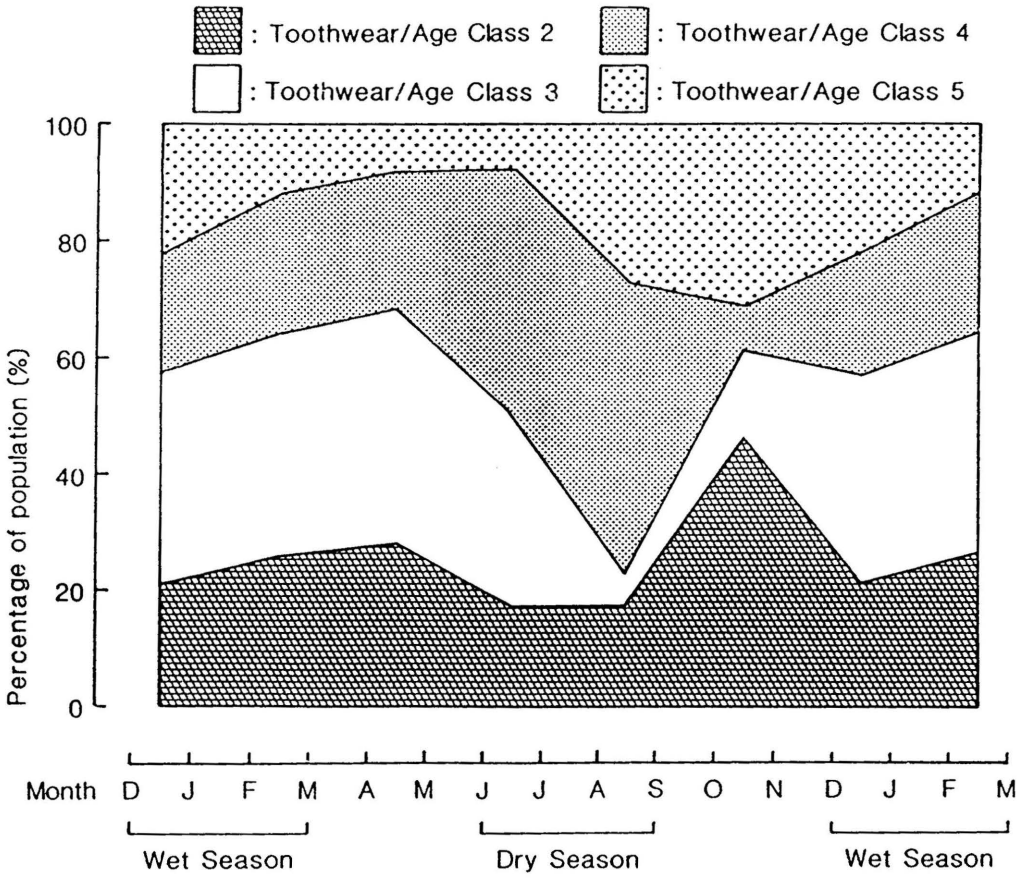

FIG. 4. Monthly variation in the age structure of *S.campestris* from the Transvaal.

Discussion

It is not clear why none of the pouched mice examined belonged to the youngest age class (TW1). Although it is possible that younger animals are not easily caught in the Sherman or Museum Special traps used by the Transvaal Museum (Korn 1987), both of these traps are capable of catching the smallest African rodent, *Mus minutoides*, which weighs between 2-12g (Smithers 1983). Alternatively, it is possible that young pouched mice are subjected to a higher rate of predation and are therefore scarcer than older age classes. Indeed, several studies have reported that *S.campestris* are commonly found in the diet of some predators such as barn owls, *Tyto alba* (eg. Coetzee 1963; Vernon 1972) which predominantly select younger rodents (Dean 1973). At the same time, Earl (1980) found that pouched mice suckle for up to 8 weeks in captivity, and attain a body weight of 38-56g which is equivalent to that of animals in age class TW2. It is therefore probable that young *S.campestris* reach this age before they leave the maternal nest, particularly as their mothers often hoard considerable quantities of seeds in their burrows (Ellison *in prep.*) which would enable the young to attain an even larger body size between weaning and dispersal.

Meanwhile, the results of the present study clearly indicate that the winter decline in body weight of *S.campestris* from the Transvaal reported by Korn (1989) was not simply the result of seasonal variation in the age structure of the population caused by a peak in breeding activity during the wet summer months. Instead, younger pouched mice displayed a significantly lower mean body weight during the dry season than during the wet season (Table I). It therefore appears that these individuals reduced their body weight as an adaptation to lower their energy requirements during the cool dry winter when food availability is low and thermoregulatory costs are high. Ellison & Skinner (1991) have shown that growing pouched mice stay small during winter in captivity and accrue considerable savings in total energy requirements without compromising their ability to produce heat. However, it remains unclear why adults in age classes TW4 and TW5 did not do the same. It is possible that older individuals might lose the capacity to modify their body size (Borer and Kaplan 1977), although Heldmaier & Steinlechner (1981) and Stewart & Barnett (1983) recorded a reduction in body weight of *Phodopus sungorus* and *Rattus fuscipes* over two consecutive winters which indicates that weight loss occurred in both subadults and adults. Alternatively, it may not be necessary for older pouched mice to reduce their body weight in winter if they have accumulated large stores of seeds in their burrows (Ellison *in prep.*). In contrast, younger animals may not have been able to establish a secure burrow or collect sufficient stores of seeds to see them through a decline in food availability during winter. This would explain why older pouched mice do not display a significant decline in body weight during winter while younger individuals do.

Summary

Although many small mammal populations display a reduction in body weight during winter it is not clear whether this represents a specific adaptation to reduce energy requirements when food availability declines, or whether it is simply a fortuitous result of a seasonal peak in breeding activity during summer. By sorting 104 pouched mice (*S.campestris*) into relative age classes based upon the degree of tooth wear the present study demonstrated that the winter decline in mean body weight described by Korn (1989) was due to a reduction in the weight of young individuals and was not the result of a change in the age structure of the population. However, there was no significant decline in the body weight of pouched mice from the two older age classes although these would have been less susceptible to food restriction during winter if they had been able to establish larger hoards of seeds in their burrows.

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REFERENCES

Borer, K.T. & Kaplan, L.R. (1977) Exercise-induced growth in golden hamsters: effects of age, weight and activity level. *Physiol. Behav.* **18:** 29-34.

Bozinovic, F., Novoa, F.F. & Veloso, C. (1990) Seasonal changes in energy expenditure and digestive tract of *Abrothrix andinus* (Cricetidae) in the Andes range. *Physiol. Zool.* **63:** 1216-1231.

Coetzee, C.G. (1963) The prey of owls in the Kruger National Park as indicated by owl pellets collected during 1960-61. *Koedoe* **6:** 115-125.

Dean, W.R.J. (1973) Age distribution of *Praomys natalensis* in *Tyto alba* pellets. *Zool. afr.* **8:** 140.

Earl, S.M.J.A. (1980) Development of behaviour in the pouched mouse *Saccostomus campestris*. Unpublished Dissertation, Faculty of Science, University of Pretoria.

Ellison, G.T.H. (*in prep.*) Group size, burrow morphology and hoarding activity of pouched mice (*Saccostomus campestris*: Cricetidae) in southern Africa.

Ellison, G.T.H. & Skinner, J.D. (1991) Seasonal energy requirements and thermoregulation of growing pouched mice, *Saccostomus campestris* (Cricetidae). *Int. J. Biometeorol.* **35:** 98-102.

Fuller, W.A., Stebbins, L.L. & Dyke, G.R. (1969) Overwintering of small mammals near Great Slave Lake, Northern Canada. *Arctic* **22:** 34-55.

Heldmaier, G. & Steinlechner, S. (1981) Seasonal control of energy requirements for thermoregulation in the Djungarian hamster (*Phodopus sungorus*), living in natural photoperiod. *J. Comp. Physiol. B* **142:** 429-437.

Iverson, S.L. & Turner, B.N. (1974) Winter weight dynamics in *Microtus pennsylvanicus*. *Ecology* **55:** 1030-1041.

Korn, H. (1987) Densities and biomasses of non-fossorial southern African savanna rodents during the dry season. *Oecologia* **72:** 410-413.

Korn, H. (1989) The annual cycle in body weight of small mammals from the Transvaal, South Africa, as an adaptation to a subtropical seasonal environment. *J. Zool. (Lond.)* **218:** 223-231.

Murua, R., Rojas M. & Gonzalez, L.A. (1987) Ciclo reproductivo anual de *Akodon olivaceus* y *Oryzomys longicaudatus* (Rodentia: Cricetidae). *Medio Ambiente* **8:** 44-51.

Petterborg, L.J. (1978) Effect of photoperiod on body weight in the vole, *Microtus montanus*. *Can. J. Zool.* **56:** 431-435.

Rautenbach, I.L. (1982) Mammals of the Transvaal. *Ecoplan Monogr.* **1:** 1-211.

Smithers, R.H.N. (1983) *The Mammals of the Southern African Subregion*. University of Pretoria, Pretoria.

Stewart, A.P. & Barnett, S.A. (1983) Seasonal changes in body weight and composition of Australian bush rats, *Rattus fuscipes*, and adaptation to winter. *Aust. J. Zool.* **31:** 29-37.

Vernon, C.J. (1972) An analysis of owl pellets collected in southern Africa. *Ostrich* **43:** 109-124.

Wunder, B.A., Dobkin, D.S. & Gettinger R.D. (1977) Shifts of thermogenesis in the prairie vole (*Microtus ochrogaster*). *Oecologia* **29:** 11-26.

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THERMOREGULATORY CIRCADIAN RHYTHMS IN THE POUCHED MOUSE (*SACCOSTOMUS CAMPESTRIS*)

A. HAIM,* G. T. H. ELLISON and J. D. SKINNER
Mammal Research Institute, University of Pretoria, Pretoria 0002, South Africa

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Abstract—1. Circadian rhythms of body temperature (T_b), oxygen consumption ($\dot{V}O_2$), and minimal thermal conductance (C) were studied in the pouched mouse, *Saccostomus campestris* under natural photoperiod during February at a constant ambient temperature of 28°C.

2. Circadian rhythms of body temperature were also studied under natural photoperiod and laboratory temperatures (Max: 28.1°C; Min: 23.2°C) during February.

3. The results of the present study suggest that changes in ambient temperature are not the main "zeitgeber" for body temperature rhythm, and it seems that photoperiod plays a major role in this species.

4. The relationship between the rhythms of T_b, $\dot{V}O_2$, and C are further discussed.

INTRODUCTION

Circadian rhythms are well known manifestations of internal, biological clocks, which provide "the framework for a temporal programming of biological activity to appropriate times of day" (Pittendrigh, 1981).

In their review of rhythmic variations in energy metabolism, Aschoff and Pohl (1970) concluded that there are genuine circadian rhythms of oxygen consumption, and they demonstrated that these rhythms are, to a certain extent, independent of both activity and food intake.

Circadian variation of thermoregulatory responses has been recorded in humans (Smith 1969, Aschoff and Heise 1972, Stephenson *et al.* 1984). Such circadian rhythms of temperature regulation have also been reported for the laboratory rat, *Rattus norvegicus*, by DeCastro (1978), Sugano (1980) and, more recently, by Shido *et al.* (1986). From the results of these studies, it is apparent that the thermoregulatory mechanisms of the rat change with circadian phase.

The pouched mouse, *Saccostomus campestris* (Rodentia:Cricetidae), is a sluggish, nocturnal species (DeGraaf, 1981) whose distribution extends from Uganda and Kenya in the tropics to the Cape Province in the south of Africa (Smithers, 1983).

Observations in our laboratory revealed that the mice are asleep during the day. Furthermore, in the winter, when they are exposed to low ambient temperatures ($T_a < 20$°C), they can become hypothermic. These characteristics make them interesting subjects for a study of circadian metabolic rhythms. In addition, although the circadian rhythm of activity has been extensively studied in the northern hemisphere cricetid, *Mesocricetus auratus* (for refs, see Elliot, 1981), no comparable investigation has yet been conducted in the southern hemisphere.

Therefore, the aim of the present study was to investigate the circadian rhythm of thermoregulation in a southern hemisphere cricetid, the pouched mouse, maintained under natural photoperiodic conditions.

MATERIALS AND METHODS

S. campestris individuals used in this study were collected at Vaalkop Dam Nature Reserve (27°30″E; 25°30″S) at the end of January 1987. They were brought to the Mammal Research Institute (Pretoria) and were housed separately in laboratory rat cages under natural photoperiodic conditions. Circadian rhythms were studied between 16 and 24 February 1987. The ambient temperature in the laboratory ranged from 23.2°C at 0600 hr and 28.1°C at 1800 hr. The studied animals were fed rat pellets (Epol), sunflower seeds and water *ad libitum* throughout the experimental period, whilst shredded paper was provided for bedding. Measurements during the dark period were made under dim red illumination.

Body temperature

Body (rectal) temperature (T_b) was measured in 12 animals on 16 November 1987 over 24 hr, at intervals of 4 hr and on 17 and 18 November 1987 every 6 hr, using a 52K/J Fluke thermometer, with a chromel-alumel thermocouple, inserted 3 cm into the rectum for 15 sec.

Oxygen consumption

Oxygen consumption ($\dot{V}O_2$) was measured in eight individuals, just below the thermoneutral zone at 28°C. An open circuit system was used as described by Depocas and Hart (1957) and Hili (1972). The subjects were placed in a clear perspex metabolic chamber (volume: 700 ml) through which dried air (Silica gel:Holpro) was passed at a rate of 600 ml/min. The metabolic chamber was submerged in a controlled temperature water bath (Labotec, Isando). Subjects were measured for at least 30 hr, during which time they were provided with shredded paper bedding, carrots and rat pellets *ad libitum*.

*Present Address: University of Haifa, ORANIM, PO Kiryat Tivon, 36910, Israel.

$\dot{V}O_2$ was recorded over a minimum of 30 min at 0600, 1200, 1800, and 2400 hr (two individuals were also measured at 2100 hr) using an Ametek S-3A/II (Applied Electrochemistry) Oxygen Analyzer. Five consecutive readings, which did not differ by more than 0.03%, were used for calculating resting metabolic rate (RMR). Following each $\dot{V}O_2$ measurement, the animal was removed from the metabolic chamber and its T_b (rectal) was measured. The apparatus was calibrated before and after each reading, and all $\dot{V}O_2$ measurements were corrected to standard temperature and pressure dry (STPD).

Minimal thermal conductance

Minimal thermal conductance (C) was calculated using the formula of Scholander *et al.* (1951) as given by Hart (1971), $C = \dot{V}O_2/(T_b - T_a)$. For justification of this method, see Bradley and Deavers (1980).

All results are given as mean ± standard error. The correlation coefficient, r, was used for establishing significance of results. Correlation coefficients with an associated probability of <0.05 were considered significant.

RESULTS

A clear circadian rhythm of T_b was recorded in the pouched mice (Fig. 1). This rhythm was out of phase with ambient temperature, T_b lagging behind T_a by approximately 6 hr. Maximal T_b was recorded between 2200 and 0200 hr, while minimal T_b was measured at 1400 hr (Fig. 1).

Figure 2 shows $\dot{V}O_2$, C and T_b of all individuals measured at different times of the circadian rhythm. From this figure, it appears that these three parameters are associated. An increase in $\dot{V}O_2$ is accompanied by an increase in T_b (between 1200 and 1800 hr), whilst in this period C does not change. A further increase in $\dot{V}O_2$ is observed between 1800 and 2400 hr. This increase, however, is not accompanied by an increase in T_b, while a significant increase in C occurs.

DISCUSSION

Many studies show that endotherms are characterized by a long term, stable, mean body temperature, with regular circadian (24 hr) variations (Aschoff, 1982).

The pouched mouse shows a 24 hr variation in body temperature which is clearly a true circadian rhythm in this species. Whereas minimal T_a was recorded at 0600 hr, minimal T_b was recorded at 1400 hr. On the other hand, maximal T_b was recorded between 1200 and 2200 hr, whilst maximal T_a was recorded at 1800 hr. Indeed, this result may raise the question of whether, in this species, ambient temperature can serve as a zeitgeber.

The higher T_b values at night are in agreement with its nocturnal behaviour pattern (Earl, 1980). Mean T_b can be up to 2.3°C lower during the inactive part of the day, when the mice are asleep. Such results were also obtained from other nocturnal species such as

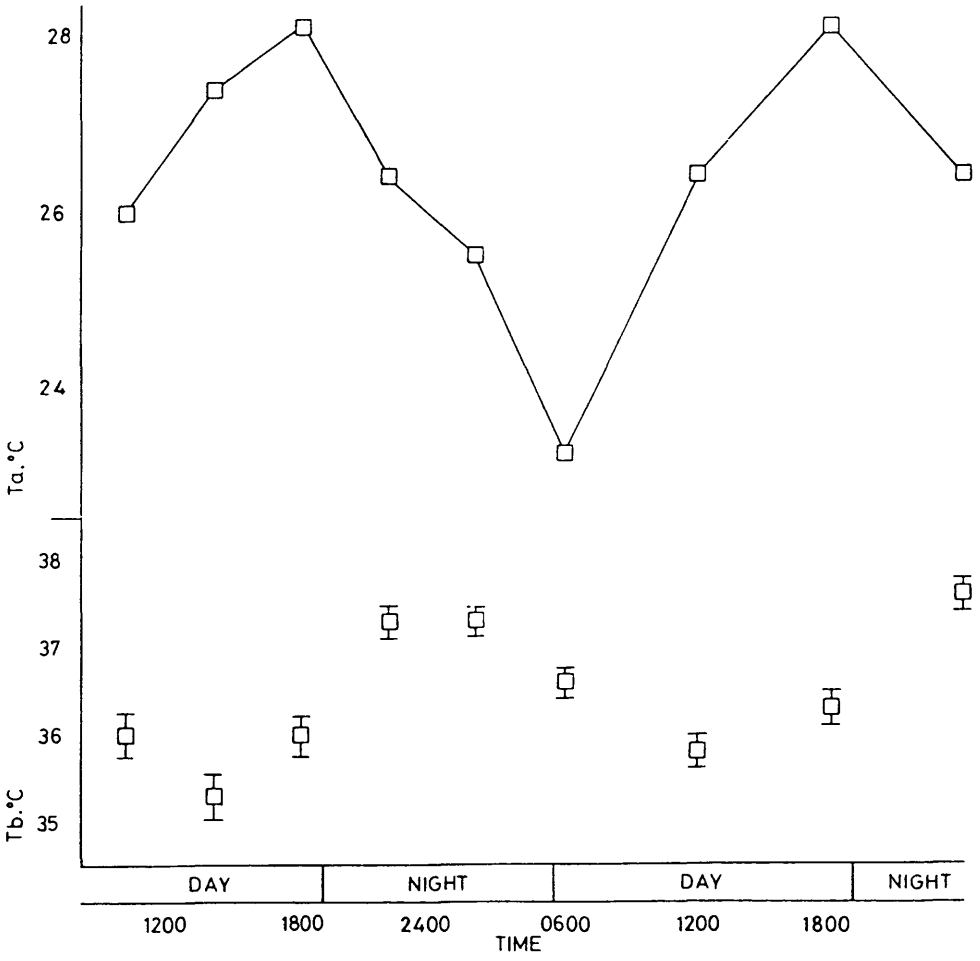

Fig. 1. Body temperature (T_b,°C) of 12 pouched mice against time of day.

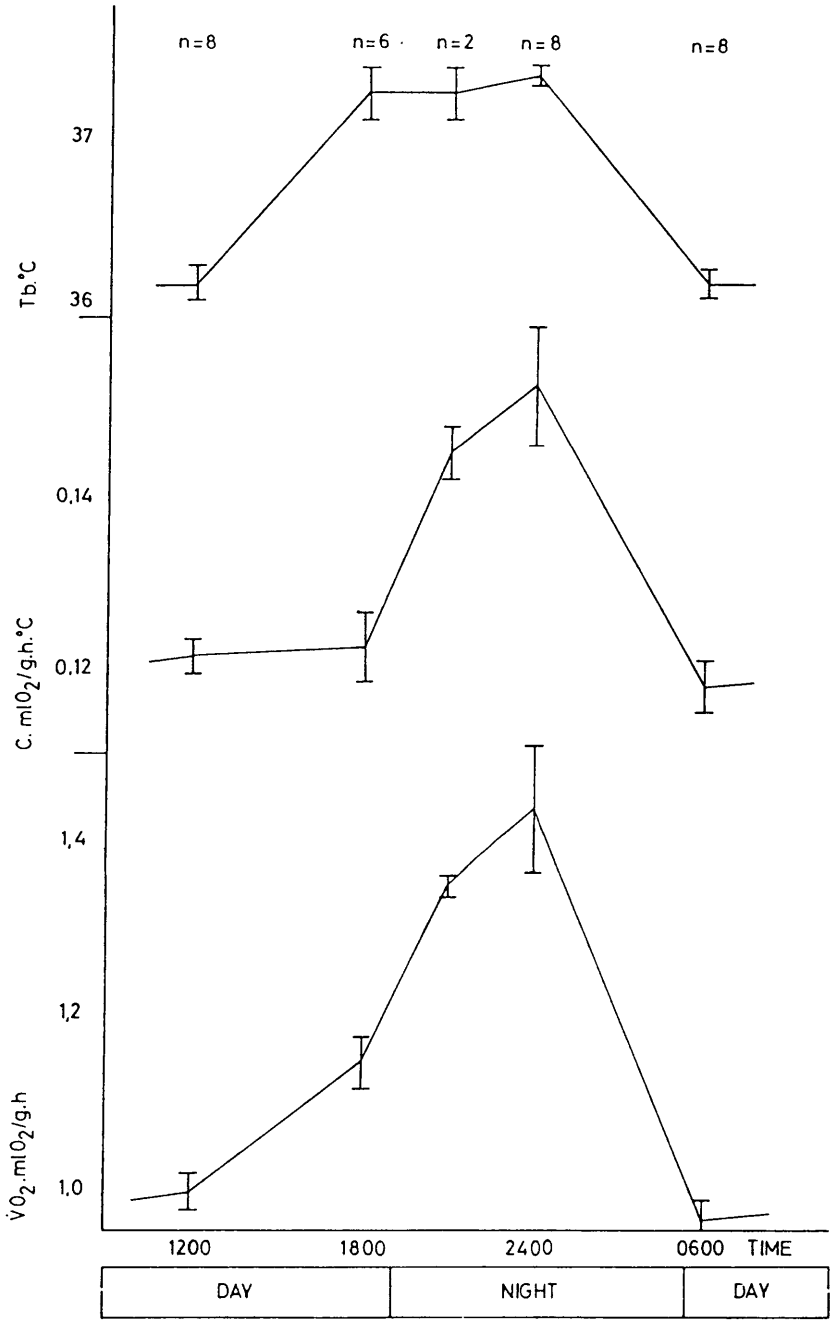

Fig. 2. Oxygen consumption ($\dot{V}O_2$ · ml O_2/g-hr), minimal conductance (C · ml O_2/g-hr, °C) and body temperature (T_b, °C) against time of day for all individuals measured at 28°C.

the laboratory rat (Shido *et al.*, 1986). In contrast, the domestic pig, *Sus scrofa*, a diurnal species, shows an increase in body temperature during daytime and a fall at night (Bond *et al.*, 1967). Ingram and Dauncey (1985) discuss the relationship between rhythms of body temperature and activity. In their opinion, physical activity and feeding contribute to the circadian variations in body temperature. From observations on the pouched mouse, it seems as if the rhythms of $\dot{V}O_2$ and T_b drive that of activity, as T_b and $\dot{V}O_2$ increased while the mice were still asleep at 1800 hr.

A circadian rhythm in oxygen consumption has been documented in many homeothermic species (Aschoff, 1982). In this study, $\dot{V}O_2$ and T_b were measured in pouched mice kept at a constant T_a 28 ± 0.5°C in a submerged metabolic chamber. Under such conditions a circadian rhythm of T_b and $\dot{V}O_2$ is still evident. The mean difference between maximal and minimal T_b at this constant temperature is reduced to 1.6°C, but there remains a clear oscillation. This result supports the idea that ambient temperature is not the most important zeitgeber in this species.

$\dot{V}O_2$ increased by 49% between the minimal value recorded at 0600 hr and the maximal recorded at 2400 hr. This increase may result from food intake and specific dynamic action (SDA) in addition to locomotor activity. Throughout our experiments, food was provided *ad libitum* (foraging is confined to scotophase in this species; Earl 1980). Because food is consumed at this time, one may argue that SDA will also increase $\dot{V}O_2$. Of the total increase in $\dot{V}O_2$, 26% was obtained between 1800 and 2400 hr. This

increase may thus derive from feeding and its associated activity.

From the results of the present study it is difficult to draw a clear conclusion about the role of feeding and activity rhythms in metabolism. The experiments were carried out in a metabolic chamber in which food was provided *ad libitum*. Although $\dot{V}O_2$ measurements were carried out only when the pouched mice were resting (not active), the nocturnal increase in $\dot{V}O_2$ could still be attributed to SDA.

The circadian rhythm of T_b may be regulated by the combination of heat production and heat dissipation. The dependence of thermal conductance on body size and circadian phase was reviewed by Aschoff (1981). In his opinion, conductance of mammals may be 50% greater if measured during the activity period. Circadian rhythms of heat dissipation by sweating and subcutaneous blood flow have been studied in the human by Stephenson *et al.* (1984). They suggest that the synchrony of these rhythms with that of body temperature, may be the result of a relative increase in noradrenaline secretion, occuring during the latter part of the day. The maximal increase of C is only 30% of the minimal values measured in the inactive phase. This value is in agreement with those mentioned by Aschoff (1981).

To better understand the relationships between T_b and $\dot{V}O_2$ the data were plotted in Fig. 3. From this figure it is apparent that both $\dot{V}O_2$ and T_b show a significant, positive correlation between 1200 and 1800 hr ($r = 0.566$; $P < 0.05$). It may be concluded that during this period minimum thermal conductance does not change, and this result is in agreement with the calculated values for conductance as represented in Fig. 2. Furthermore, it is apparent that $\dot{V}O_2$ and T_b do not show significant correlation between consecutive measurements during other parts of the circadian rhythm. However, the relationship between $\dot{V}O_2$ and T_b remains positive, which suggests that C changes at these times in a similar direction to that of $\dot{V}O_2$ and therefore T_b remains constant between 1800 and 2400 hr, and 0600 and 1200 hr, but decreases between 2400 and 0600 hr.

The increase in T_b at 1800 hr may indicate heat storage. This phenomenon has been reported elsewhere (Abrams and Hammel, 1965; Sugano, 1983; Shido *et al.*, 1986) and has been explained by a delay in the conduction of heat from body core to the outside (Sugano, 1983), or by its conversion into the "work of increased activity" (Shido *et al.*, 1986). The outphase between T_b and C at 1800 hr in the present study tends to support the former explanation. Indeed, it is tempting to suggest that $\dot{V}O_2$ or T_b must exceed a threshold value before C increases.

Both body temperature and $\dot{V}O_2$ are manifestations of a combination of the behaviour and metabolism of an animal at the time of measurement, and possibly for some hours/minutes beforehand. In general, heat production and heat dissipation (positive and negative behaviour) are balanced in endotherms so that body temperature is maintained at a level for optimum performance of bodily functions with the least expenditure of energy. However, optimum body temperature and $\dot{V}O_2$ each assume different values according to the function being performed.

Most species perform particular functions at a similar time each day and the observation presented in this paper of a circadian rhythm in body temperature and $\dot{V}O_2$ in the pouched mouse strongly suggest that it has a daily rhythm that involves it in most activity during the hours of darkness and least during daylight which is in accordance with observed behaviour (Earl, 1980).

In conclusion, the results of the present study suggest that photoperiod rather than ambient temperature is the most important zeitgeber in this species and, furthermore, that the circadian rhythm of $\dot{V}O_2$ is the major metabolic and thermoregulatory rhythm in the pouched mouse.

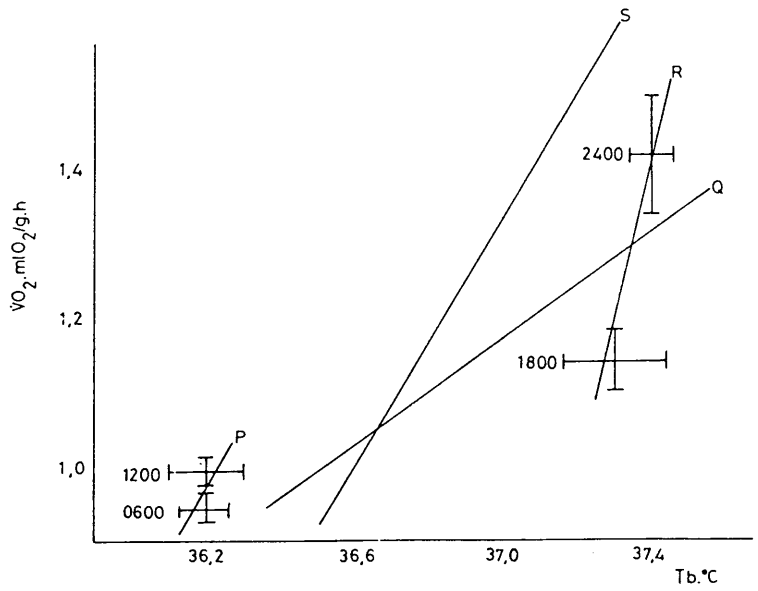

Fig. 3. Oxygen consumption ($\dot{V}O_2$·ml O_2/g-hr) against body temperature (T_b, °C) for 0600, 1200, 1800, and 2400 hr. Regression lines of $\dot{V}O_2$ against T_b are drawn for consecutive readings and have the following correlation coefficients (r): P: 0600–1200 hr, $r = 0.278$; $P < 0.2$. Q: 1200–1800 hr, $r = 0.566$; $P < 0.05$. R: 1800–2400 hr, $r = 0.222$; $P < 0.2$. S: 2400–0600 hr, $r = 0.459$; $P < 0.1$.

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REFERENCES

Abrams R. and Hammel H. T. (1965) Cyclic variations in hypothalamic temperature in unanaesthetized rats. *Am. J. Physiol.* **208,** 698–702.

Aschoff J. (1981) Thermal conductance in mammals and birds: its dependence on body size and circadian phase. *Comp. Biochem. Physiol.* **69A,** 611–619.

Aschoff J. (1982) The circadian rhythm of body temperature as a function of body size. In *A Companion To Animal Physiology* (Edited by Taylor C. R., Johanson K. and Bolis L.), pp. 173–188. Cambridge University Press, Cambridge.

Aschoff J. and Pohl H. (1970) Rhythmic variations in energy metabolism. *Fed. Proc.* **29,** 1541–1552.

Aschoff J. and Heise A. (1972) Thermal conductance in man: its dependence on time of day and ambient temperature. In *Advances in Climatic Physiology* (Edited by Itoh S., Ogata K. and Yoshimura H.), pp. 334–348. Igaku Shoin, Tokyo.

Bond T. E., Kelly C. F. and Heitman H. (1967) Physiological response of swine to cycling environmental conditions. *Anim. Prod.* **9,** 453–462.

Bradley S. R. and Deavers D. R. (1980) A re-examination of the relationship between thermal conductance and body weight in mammals. *Comp. Biochem. Physiol.* **65A,** 465–476.

DeCastro J. M. (1978) Diurnal rhythms of behavioural effects on core temperature. *Physiol. Behav.* **21,** 883–886.

DeGraaf G. (1981) *The Rodents of Southern Africa.* Butterworths, London.

Depocas F. and Hart J. S. (1957) The use of the Pauling Oxygen Analyzer for measurements of oxygen consumption of animals in open-circuit systems, and in short-lag, closed-circuit apparatus. *J. appl. Physiol.* **10,** 388–392.

Earl S. M. J. A. (1980) Development of behaviour in the pouched mouse *Saccostomus campestris.* Unpublished MSc Thesis, University of Pretoria, South Africa.

Elliott J. A. (1981) Circadian rhythms, entrainament and photoperiodism in the Syrian hamster. In *Biological Clocks in Seasonal Reproductive Cycles* (Edited by Follett B. K. and Follett D. E.), pp. 203–216. John Wright, Bristol.

Hart J. S. (1971) Rodents. In *Comparative Physiology of Thermoregulation* (Edited by Whittow G. C.), pp. 1–148. Academic Press, New York.

Hill R. W. (1972) Determination of oxygen consumption by use of the paramagnetic oxygen analyzer. *J. appl. Physiol.* **33,** 261–263.

Ingram D. L. and Dauncey M. J. (1985) Circadian rhythms in the pig. *Comp. Biochem. Physiol.* **82A,** 1–5.

Pittendrigh C. S. (1981) Circadian organization and the photoperiodic phenomena. In *Biological Clocks in Seasonal Reproductive Cycles* (Edited by Follett B. K. and Follett D. E.), pp. 1–35. John Wright, Bristol.

Shido O., Sugano Y. and Nagasaka T. (1986) Circadian change of heat loss in response to change in core temperature in rats. *J. Therm. Biol.* **11,** 199–202.

Smith R. E. (1969) Circadian variations in human thermoregulatory responses. *J. appl. Physiol.* **26,** 554–560.

Smithers R. H. N. (1983) *The Mammals of the Southern African Subregion.* University of Pretoria Press, Pretoria.

Stephenson L. A., Wenger C. B., Donovan B. H. and Nadel E. R. (1984) Circadian rhythm of sweating and cutaneous blood flow. *Am. J. Physiol.* **246,** R321–R324.

Sugano Y. (1983) Heat balance of rats acclimated to diurnal 2 hour feeding. *Physiol. Behav.* **30,** 289–293.

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SEASONAL ACCLIMATIZATION AND THERMOREGULATION IN THE POUCHED MOUSE *SACCOSTOMUS CAMPESTRIS*

A. HAIM,* P. A. RACEY,† J. R. SPEAKMAN,† G. T. H. ELLISON and J. D. SKINNER
Mammal Research Institute, University of Pretoria, Pretoria 0002, R.S.A.

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Abstract—The pouched mouse *Saccostomus campestris*, a nocturnal, fossorial rodent, is widely distributed in the Southern-African subregion.

Seasonal acclimatization of mechanisms of thermoregulation and body mass were studied in this species under the natural photoperiod conditions prevailing in Pretoria during January–February and July–August at room temperatures of 23.2–28.1 and 9.8–18.2°C, respectively.

The following parameters were measured or calculated: body mass, oxygen consumption and body temperature at various ambient temperatures, minimal overall thermal conductance and capacity for non-shivering thermogenesis.

In winter-acclimatized pouched mice, body mass, resting metabolic rates and non-shivering thermogenesis capacity are higher than in summer-acclimatized mice, while lower critical point and minimal thermal conductance are higher in summer-acclimatized mice than in winter-acclimatized.

The seasonal changes of the studied parameters, may help explain the ability of this species to cope with its habitat in the two different seasons.

Key Word Index: Non shivering thermogenesis; thermal conductance; seasonal acclimatization; fossorial; photoperiod; metabolic rate

INTRODUCTION

Seasonal acclimatization of thermoregulation and metabolism has been studied in several rodent species from the northern hemisphere (Rosenmann *et al.*, 1975; Cygan, 1985; Heldmaier *et al.*, 1986; Feist and Feist, 1986). Intensive studies have been carried out on the Djungarian hamster *Phodopus sungorus* (Heldmaier *et al.*, 1982, 1986; Steinlechner and Heldmaier, 1982).

The pouched mouse *Saccostomus campestris*, which is both nocturnal and fossorial, is one of the two species of Cricetomyinae rodents known from the Southern-African subregion. It is widely distributed in this area with the exception of arid habitats (De Graaff, 1981; Smithers, 1983). According to Rautenbach (1982) the distribution of this species in the Transvaal is largely confined to woodland. The aim of this study is to investigate seasonal changes in energy metabolism, heat production and thermoregulation in the pouched mouse and to compare the results with those known from rodents of the northern hemisphere.

MATERIALS AND METHODS

The pouched mice used in this study were trapped at Vaalkop Dam Nature Reserve (27°39′E, 25°30′S), R.S.A. The mice studied during summer, summer-acclimatized (SA) were captured at the end of January 1987 and those which were studied during winter, winter-acclimatized (WA), were captured at the end of April 1987. Each group consisted of four males and two-non-pregnant females. In both seasons they were kept for at least 2 weeks in individual rat cages under natural photoperiod and room temperature. In January–February room temperature varied between a maximum of 28.1°C in the early evening and a minimum of 23.2°C in the early morning. During July–August room temperature varied between 18.2 and 9.8°C. The mice were fed with rat pellets and sunflower seeds. Water was offered *ad lib*, and shredded paper was provided for bedding.

Oxygen consumption

Oxygen consumption ($\dot{V}O_2$) was measured over a range of ambient temperatures (T_a), between 5–40°C in summer-acclimatized and 5–34°C in winter-acclimatized animals. Individuals were measured separately in an open flow system (Depocas and Hart, 1957; Hill, 1972). $\dot{V}O_2$ was monitored with an Ametek S-3A/II (Applied Electrochemistry) oxygen analyser. In order to assess minimal observed metabolic rates (MOMR) and minimal resting metabolic rates (RMR), $\dot{V}O_2$ measurements were carried out during day-time (photophase) in a clear perspex metabolic chamber (volume of 700 ml). A flow of dried air (silica gel, Holpro) was passed through the metabolic chamber at a rate of 600 ml/min. After a 2–3h stabilizing period, $\dot{V}O_2$ was recorded for 30 min. Five consecutive readings, at intervals of 3 min (which did not differ by more than 0.03%), were used for calculating the resting metabolic rate (RMR). The oxygen

*,†Permanent addresses: *University of Haifa, Oranim P. O. Kiryat Tivon 36910, Israel and author to whom all correspondence should be addressed; and †Department of Zoology, University of Aberdeen, Aberdeen AB9 2TN, Scotland.

analyser was calibrated before and after each measurement. Body mass (W_b) of measured pouched mice was recorded before the $\dot{V}O_2$ measurements were carried out. All $\dot{V}O_2$ measurements were corrected to standard temperature and pressure (STPD).

Temperature regulation

Body (rectal) temperature (T_b) was measured at the end of each 30 min period of $\dot{V}O_2$ measurement. A chromel–alumel thermocouple connected to a 52 K/J Fluke thermometer, was inserted 3 cm into the rectum for a period of 15 s. T_a was also measured using a chromel–alumel thermocouple placed inside the metabolic chamber and connected to the same thermometer.

Minimal overall thermal conductance

Conductance was calculated below the lower critical point at $T_a = 28$ and 24°C for SA and WA pouched mice, respectively, using the formula of Scholander *et al.* (1950), as given by Hart (1971), assuming that the main avenue for heat loss under experimental conditions is through "dry" physical parameters.

Non-shivering thermogenesis

Non-shivering thermogenesis was measured at lower critical temperature; $T_a = 30$°C in SA mice and at $T_a = 27$°C in WA mice, in unanaesthetized individuals, using the method of Heldmaier *et al.* (1982). Noradrenaline (L-noradrenaline bitartarate Sigma; NA), 1.5 mg/kg, was injected subcutaneously (Heldmaier, 1972) into mice after at least five successive O_2 readings with intervals of 3 min that did not differ by more than 0.015%. This value represents minimal $\dot{V}O_2$, ($\dot{V}O_2$min). Recordings went on for at least 75 min after NA injection. NST capacity was calculated as the ratio between oxygen consumption measured as a response to a noradrenaline injection $\dot{V}O_2$NA and $\dot{V}O_2$min. T_b was measured before injecting NA ($T_b$min) and at the end of $\dot{V}O_2$ measurement (T_bNA).

All results are given as mean and standard deviations. Student's t-test was used to test significance of differences. Comparison between the slopes of regression lines was done as in Marascuilo and Serlin (1988).

RESULTS

The thermoneutral point for SA pouched mice is at 32 ± 1°C with a resting metabolic rate of 0.770 ± 0.10 ml O_2/g/h. In WA mice, the thermoneutral zone is between 28 and 30 ± 1°C with a resting metabolic rate of 0.932 ± 0.05 ml O_2/g/h, which is significantly ($P < 0.05$) higher (Figs 1 and 2).

The equations of the regression lines calculated for $\dot{V}O_2$ between $T_a = 5$°C and the lower critical point are: $\dot{V}O_2 = -0.123 \times T_a + 4.482$ for SA mice and $\dot{V}O_2 = -0.103 \times T_a + 3.891$ for WA mice. The slopes of the two equations do not differ significantly.

The body temperature at the thermoneutral point for SA mice was 35.2 ± 0.9°C while for WA mice it was 35.3 ± 0.2°C at an ambient temperature of 28°C and 37.2 ± 1.0°C at an ambient temperature of 32°C. SA mice were hyperthermic at an ambient temperature of 36°C and above. Two individual mice died on reaching the upper lethal temperature of 40°C (Fig. 1). In the SA mice, salivation was observed in all mice at an ambient temperature of 36°C while in WA mice, salivation was observed at $T_a = 34$°C and body temperature was regulated at 38.1 ± 0.7°C.

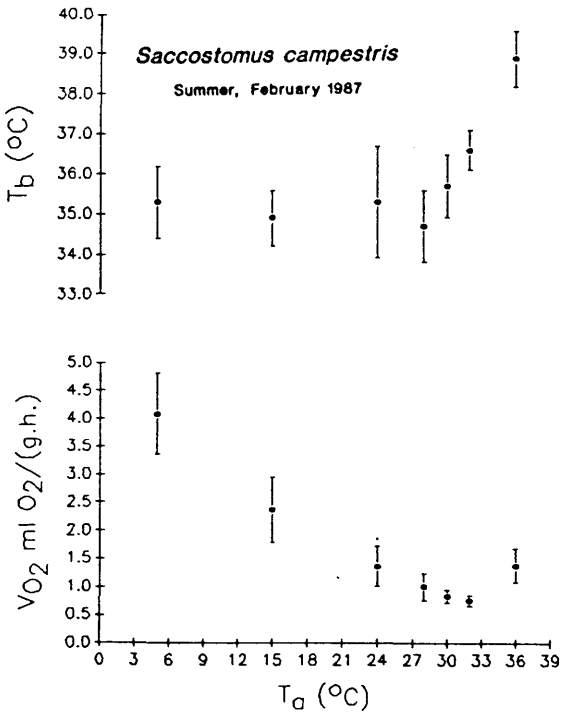

Fig. 1. Oxygen consumption $\dot{V}O_2$ and body temperature T_b. of SA—summer acclimatized—pouched mice *Saccostomus campestris* at different ambient temperatures (T_a). Each point is a mean $\pm$ SD of $n = 6$.

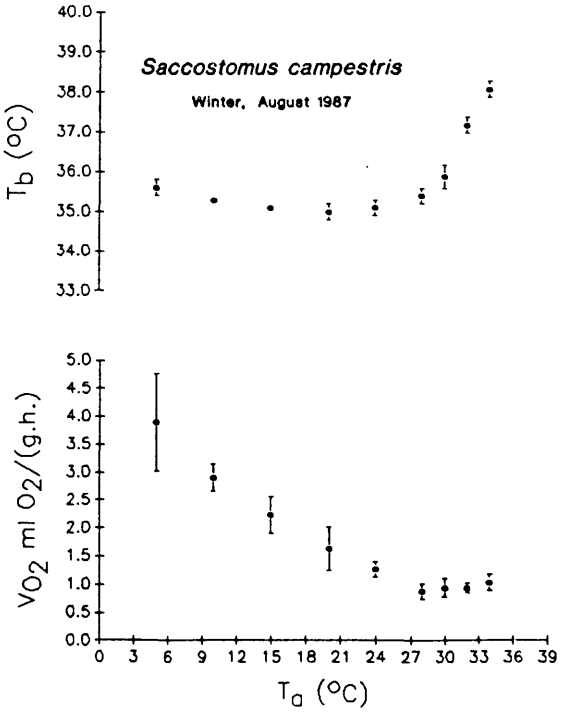

Fig. 2. Oxygen consumption $\dot{V}O_2$ and body temperature T_b. of WA—winter acclimatized—pouched mice *Saccostomus campestris* at different ambient temperatures (T_a). Each point is mean $\pm$ SD of $n = 6$.

Table 1. Overall thermal conductance (C) for the two groups of pouched mice (SA—summer acclimatized and WA—winter acclimatized) *Saccostomus campestris*. All results are mean ± SD of $n = 6$, T_a = ambient temperature, W_b = body mass

	C (ml O_2/g/h/°C)		W_b (g)	
T_a (°C)	24	28	24	28
SA mice	0.132 ± 0.01	0.152 ± 0.02	60.2 ± 8.3	61.4 ± 9.0
WA mice	0.114 ± 0.01*	0.125 ± 0.01*	83.4 ± 9.2**	85.2 ± 9.7**

*Significantly ($P < 0.05$) lower when compared with SA mice.
**Significantly ($P < 0.01$) higher when compared with SA mice.

Table 2. Non-shivering thermogenesis (NST) in SA (summer acclimatized) and WA (winter acclimatized) pouched mice *Saccostomus campestris*, expressed as maximal $\dot{V}O_2$ ($\dot{V}O_2$NA) and T_b (T_bNA) response to an injection of noradrenaline—NA, (1.5 m g/kg s.c.). Minimal values of $\dot{V}O_2$ ($\dot{V}O_2$ min) and T_b (T_b min) were measured close to lower critical temperature (T_a); W_b = body mass, NST capacity $\dot{V}O_2$NA/$\dot{V}O_2$ min. All results are mean ± SD of $n = 6$

	SA mice	WA mice
T_a (°C)	30	28
W_b (g)	61.3 ± 8.5	85.2 ± 9.4*
$\dot{V}O_2$ min (ml O_2/g/h)	0.840 ± 0.10	0.980 ± 0.06*
T_b min (°C)	35.8 ± 0.9	35.4 ± 0.2
$\dot{V}O_2$ NA (ml O_2/g/h)	3.286 ± 0.36	4.460 ± 0.48**
T_b NA (°C)	38.8 ± 0.5	39.8 ± 0.4*
$\dot{V}O_2$ NA/$\dot{V}O_2$ min	3.9 ± 0.3	4.5 ± 0.4*

*Significantly ($P < 0.05$) higher when compared with SA mice.
**Significantly ($P < 0.01$) higher when compared with SA mice.

The minimal overall thermal conductance of WA mice measured at 24°C was significantly ($P < 0.05$) lower than that of SA mice measured at 28°C (Table 1). Body mass of WA mice was significantly ($P < 0.01$) higher than in SA mice (Tables 1 and 2). A higher body mass was also observed in mice freshly captured in winter when compared to those captured in summer.

$\dot{V}O_2$ values obtained as a response to noradrenaline injection were significantly ($P < 0.01$) higher in WA mice when compared with the values for SA mice (Table 2). Maximum body temperature and capacity for NST were significantly ($P < 0.05$) higher in the WA mice when compared with the SA mice (Table 2).

DISCUSSION

The pouched mouse *Saccostomus campestris* in the Transvaal experiences a cold dry winter alternating with a warm and wet summer. Being a nocturnal species (De Graaff, 1981) it avoids the heat of the habitat during summer, yet can be active in ambient temperatures which exceed those of winter by more than 25°C. As in other small rodents insulation does not play a major role in improving thermoregulation (Hart, 1956). Therefore, seasonal changes in mechanisms of heat production may be expected (Heldmaier *et al.*, 1986).

The results of this study show that the pouched mouse responds to experimental conditions by seasonal acclimatization of heat production and thermoregulatory mechanisms. Mean body mass in the WA mice increased significantly ($P < 0.01$) relative to the SA mice (Table 1). This increase contrasts with what is found in the Djungarian hamster *Phodopus sungorus* (Heldmaier *et al.*, 1986) and in the Red-backed vole *Clethrionomys rutilus* (Sealander, 1967), In both these species SA individuals have a higher body mass relative to WA individuals. An increase in body mass of WA rodents was reported in the Golden spiny mouse *Acomys ressatus* (Haim and Borut, 1975) and in the Yellow-necked field mouse *Apodemus flavicollis* (Cygan, 1985).

Oxygen consumption is related to body mass by an allometric relationship: $\dot{V}O_2$ ml O_2/g/h = 3.8 $W^{-0.23}$ (W = body mass) (Brody, 1945). Resting metabolic rate in the thermoneutral zone is significantly ($P < 0.05$) higher in the WA mice. The deviation from the expected $\dot{V}O_2$ according to body mass (85.2 g) in WA mice is only 12% while for the SA mice ($W_b = 61.3$ g) it is 38%. Low values of $\dot{V}O_2$ in the thermoneutral zone are found in rodents from desert habitats on the one hand (Yousef and Johnson, 1975; Haim, 1984, 1987; Haim and Borut, 1986) but also in subterranean and fossorial rodents on the other (McNab, 1966; Haim and Fairal, 1986; Lovegrove, 1986). As arid habitats are excluded from the distribution of *Saccostomus campestris*, it would seem as if the low resting metabolic rate is an adaptation to the fossorial habits of this species. However, although RMR is significantly different the slopes of the regression lines of the two groups do not differ significantly.

The seasonal changes in body mass play a role in minimal overall thermal conductance as this parameter is related to body mass by the allometric equation $C = 0.760\ W^{-0.426}$ (Bradley and Deavers, 1980). One would expect higher conductance values for the SA mice as their body mass is lower. Indeed at $T_a = 24$°C as well as at $T_a = 28$°C (Table 1) conductance is significantly ($P < 0.05$) lower in the WA mice. The difference in conductance values between these two groups may contribute to a better thermoregulatory ability, which is an adaptation enabling the pouched mice to cope with low ambient temperatures during activity in winter. In the SA mice the calculated values of conductance at $T_a = 28$°C are only 12% higher than those expected for the related body mass. This deviation of 12% in conductance from the predicted values according to body mass is much lower than the deviation of almost 40% found in subterranean mole-rats *Spalax ehrenbergi* ($2n = 60$) (Nevo and Shkolnik, 1974) and *Cryptomus hottentotus* (Haim and Fairall, 1986). The 12% higher value of conductance (than that predicted from body mass) may contribute to their low T_b (35°C) at the lower critical temperature (Figs 1 and 2). De Graaff (1981) describes the pouched mouse as a sluggish animal which can easily be picked up by hand.

NST is the main avenue for heat production in cold-acclimated small mammals (Jansky, 1973; Webster, 1974) as well as in those acclimated to long scotophase (Lynch, 1970; Haim and Fourie, 1980), and in WA mice (Heldmaier *et al.*, 1981; Haim and

Yahav, 1982). The results of this study show that in the WA pouched mice there is a significant ($P < 0.01$ and $P < 0.05$) increase in $\dot{V}O_2$ NA and T_b NA, as well as a significant ($P < 0.05$) increase in NST capacity—$\dot{V}O_2$ NA/$\dot{V}O_2$min (Table 2). Such seasonal changes in NST were reported for the Djungarian hamster (Heldmaier *et al.*, 1982) as well as for the Red-backed vole (Feist and Feist, 1986) and *Peromyscus leucopus* and *P. maniculatus* (Zegers and Merritt, 1988). In the pouched mouse the increase in NST may be a response to the change in photoperiod as well as the change in T_a. Further experiments using the same methods and protocol as in Haim (1982) and Heldmaier *et al.* (1982) may distinguish between the contribution of low ambient temperatures and long scotophase to the increased NST.

Heldmaier (1972) showed that NST, measured as $\dot{V}O_2$ NA, is a function of body mass. The allometric relation is described by the equation NST = $\dot{V}O_2$ ml O_2/g/h = 30 $W^{-0.454}$ for cold-acclimated ($T_a = 5°C$) rodents. The expected maximal $\dot{V}O_2$ due to noradrenaline injection for a cold-acclimated rodent weighing 85.2 g is 4.0 ml O_2/g/h. The measured $\dot{V}O_2$ in this study for the WA mice is 4.46 = 0.48 ml O_2/g/h which is above the expected value, although the mice were exposed to $T_a > 5°C$. The expected $\dot{V}O_2$ value for cold-acclimated mice weighing 61.3 g is 4.6 ml O_2/g/h. The results of this study show that in the SA mice $\dot{V}O_2$ NA is only 3.29 ± 0.36 ml O_2/g/h. Yet this is a relatively high value, as the mice are not cold-acclimated.

The increase in body mass on the one hand and the increase in NST capacity on the other, may appear as adaptations enabling the pouched mice to cope with the cold winter. The NST values measured in the SA mice are in agreement with those obtained from other subterranean rodents (Haim *et al.*, 1984, Haim and Fairall, 1986).

From the results of this study it may be concluded that seasonal acclimatization in the pouched mouse involves mechanisms of heat production and dissipation as well as changes in body mass. Acclimatization to winter conditions includes: an increase in body mass and RMR as well as in NST capacity and a decrease in lower critical point and overall thermal conductance.

SUMMARY

Seasonal acclimatization of body mass and mechanisms of thermoregulation were studied in the pouched mouse *Saccostomus campestris*.

The results of this study reveal that under winter conditions the studied individuals increased body mass, resting metabolic rate and non-shivering thermogenesis capacity when compared with summer-acclimatized individuals on the one hand, while on the other, such individuals decrease their lower critical point and overall thermal conductance when compared with summer-acclimatized individuals.

It may be concluded that seasonal acclimatization in the pouched mouse involves changes in body mass as well as changes in thermoregulatory mechanisms.

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REFERENCES

Bradley S. R. and Deavers D. R. (1980) A re-examination of the relationship between thermal conductance and body weight in mammals. *Comp. Biochem. Physiol.* **65A,** 465–476.

Brody S. (1945) *Bioenergetics and Growth*, p. 1023. Reinhold, New York.

Cygan T. (1985) Seasonal changes in thermoregulation and maximum metabolism in the yellow-necked field mouse. *Acta Theriol.* **30,** 115–130.

De Graaff G. (1981) *The Rodents of Southern Africa*, pp. 115–119. Butterworths, Durban.

Depocas F. and Hart J. S. (1957) The use of the Pauling Oxygen Analyzer for measurements of oxygen consumption of animals in open-circuit system, and in short-lag, closed-circuit apparatus. *J. appl. Physiol*, **10,** 388–392.

Feist D. D. and Feist C. F. (1986) Effects of cold, short day and melatonin on thermogenesis, body weight and reproductive organs in Alaskan red-backed voles. *J. comp. Physiol. B* **156,** 741–746.

Haim A. (1982) Effects of long scotophase and cold acclimation on heat production in two diurnal rodents. *J. comp. Physiol. B* **148,** 77–81.

Haim A. (1984) Adaptive variations in heat production within gerbils (genus Gerbillus) from different habitats. *Oecologia, Berl.* **61,** 49–52.

Haim A. (1987) Thermoregulation and metabolism of Wagner's gerbil (Gerbillus dasyurus): a rock dwelling rodent adapted to arid and mesic environments. *J. therm. Biol.* **12,** 45–48.

Haim A. and Borut A. (1975) Size and activity of a cold resistant population of the golden spiny mouse (*Acomys russatus*: Muridae). *Mammalia* **39,** 605–612.

Haim A. and Borut A. (1986) Reduced heat production in the bushy tailed gerbil *Sekeetamys calurus* (Rodentia) as an adaptation to arid environments. *Mammalia* **50,** 27–33.

Haim A. and Fairall N. (1986) Physiological adaptations to the subterranean environment by the mole rat *Cryptomys hottentotus*. *Cimbebasia* **8**(6), 49–53.

Haim A. and Fourie Fle R. (1980) Heat production in cold and long scotophase acclimated and winter acclimatized rodents. *Int. J. Biomet.* **24,** 231–235.

Haim A. and Yahav S. (1982) Non shivering thermogenesis in winter acclimatized and in long scotophase and cold acclimated *Apodemus mystacinus* (Rodentia). *J. therm. Biol.* **7,** 193–195.

Haim A., Heth G., Avnon Z. and Nevo E. (1984) Adaptive physiological variation in nonshivering thermogenesis and its significance in speciation. *J. comp. Physiol. B* **154,** 145–147.

Hart J. S. (1956) Seasonal changes in insulation of the fur. *Can. J. Zool.* **24,** 53–57.

Hart J. S. (1971) Rodents. In *Comparative Physiology of Thermoregulation* (Edited by Whittow C. G.), Vol. II, pp. 1–149. Academic Press, New York.

Heldemaier G. (1972) Cold-adaptive changes of heat production in mammals. In *Proceedings of Environmental Physiology* (Edited by Smith R. E.), pp. 79–82. FASEB.

Heldmaier G., Steinlechner S., Rafael J. and Latteier B. (1982) Photoperiod and ambient temperature as environmental cues for seasonal thermogenic adaptation in the djungarian hamster, *Phodopus sungorus*. *Int. J. Biomet.* **26,** 339–345.

Heldmaier G., Steinlechner S., Rafael J. and Vsiansky P. (1981) Photoperiodic control and effect of melatonin on nonshivering thermogenesis and brown adipose tissue. *Science* **212,** 917–919.

Heldmaier G., Boekler H., Buchberger A., Klaus S., Puchalski S., Steinlechner S. and Wiesinger H. (1986) Seasonal

variation of thermogenesis. In *Living in the Cold: Physiological and Biochemical Adaptations* (Edited by Heller H. C. *et al.*), pp. 361–372. Elsevier Science, Amsterdam.

Hill R. W. (1972) Determination of oxygen consumption by use of the paramagnetic oxygen analyzer. *J. appl. Physiol.* **33,** 261–263.

Jansky L. (1973) Non-shivering thermogenesis and its thermoregulatory significance. *Biol. Rev.* **48,** 85–132.

Lovegrove B. G. (1986) The metabolism of social subterranean rodents: adaptation to aridity. *Oecologia, Berl.* **69,** 551–555.

Lynch G. R. (1970) Effect of photoperiod and cold acclimation on non-shivering thermogenesis in *Peromyscus leucopus*. *Am. Zool.* **10,** 308.

Marascuilo L. A. and Serlin R. C. (1988) *Statistical Methods for Social and Behavioral Sciences*. Freeman, New York.

McNab B. K. (1966) The metabolism of fossorial rodents: a study of convergence. *Ecology* **47,** 712–733.

Nevo E. and Shkolnik A. (1974) Adaptive metabolic variation of chromosome forms in mole rats, Spalax. *Experientia* **30,** 724–726.

Rautenbach I. L. (1982) *Mammals of the Transvaal*, pp. 92–93. Ecoplan Monograph, Pretoria.

Rosenmann M., Morrison P. and Feist D. D. (1975) Seasonal changes in the metabolic capacity of red-backed voles. *Physiol. Zool.* **48,** 303–310.

Scholander P. F., Hock R., Walters V. and Irving L. (1950) Adaptations to cold in arctic and tropic mammals and birds in relation to body temperature, insulation, and basal metabolic rate. *Biol. Bull.* **99,** 259–271.

Sealander J. A. (1967) Reproduction status and adrenal size in the northern red-backed vole in relation to season. *Int. J. Biomet.* **11,** 213–220.

Smithers R. H. N. (1983) *The Mammals of the South African Subregion*, pp. 283–284. University of Pretoria, Pretoria.

Steinlechner S. and Heldmaier G. (1982) Role of photoperiod and melatonin in seasonal acclimatization in the djungarian hamster *Phodopus sungorus*. *Int. J. Biomet.* **26,** 329–337.

Webster A. J. F. (1974) Adaptation to cold. In *MIP International Review of Science, Physiology, Ser. 1, Vol. 7 (Environmental Physiology)* (Edited by Robertshaw D.), pp. 71–107. Butterworths, London.

Yousef M. K. and Johnson H. D. (1975) Thyroid activity in desert rodents. A mechanism for lower metabolic rate. *Am. J. Physiol.* **229,** 727–731.

Zegers D. A. and Merritt J. F. (1988) Adaptations of *Peromyscus* for winter survival in an Appalachian montane forst. *J. Mammal.* **69,** 516–523.

APPENDIX II: UNPUBLISHED MANUSCRIPTS CITED IN THE THESIS

ELLISON, G.T.H., SKINNER, J.D. & FERGUSON, J.W.H. *in prep.* The effect of temperature and photoperiod on daily metabolism and activity of pouched mice (*Saccostomus campestris*) from contrasting habitats in southern Africa.

WESTLIN, L.M. & ELLISON, G.T.H. (*in prep.*) Reproductive flexibility in female pouched mice (*Saccostomus campestris*: Cricetomyinae).

The effect of temperature and photoperiod on daily metabolism and activity of pouched mice (*Saccostomus campestris*: Cricetidae) from contrasting habitats in southern Africa.

G.T.H. ELLISON, J.D. SKINNER AND J.W.H. FERGUSON

Mammal Research Institute and Department of Zoology, Pretoria University, Pretoria 0002, Republic of South Africa

ELLISON, G.T.H., J.D. SKINNER AND J.W.H. FERGUSON. *The effect of temperature and photoperiod on daily metabolism and activity of pouched mice* (Saccostomus campestris*: Cricetidae) from southern Africa.* - Daily variation in metabolism (oxygen consumption) and activity was recorded in two populations of pouched mice (*Saccostomus campestris)* from contrasting habitats, following warm (25°C) and cold (10°C) acclimation under long (LD 14:10) and short (LD 10:14) photoperiod. 7 cold acclimated pouched mice displayed bouts of hypometabolism which were characteristic of spontaneous torpor. All but one of these occurred following acclimation to short photoperiod which indicates that photoperiod had an effect on torpor. However, these reductions in energy expenditure were masked by a significant increase in daily metabolism which occurred at 10°C and there was no significant effect of photoperiod or locality on metabolic rate. Meanwhile, there was a significant interactive effect of photoperiod and temperature on the percentage of time spent active such that pouched mice became more active under short photoperiod at 25°C but displayed a reduction in activity under short photoperiod at 10°C when the incidence of torpor was highest. Although pouched mice from the cooler locality maintained a similar level of activity at 25°C and 10°C, pouched mice from the warmer area displayed a reduction in daily activity following cold acclimation. However there was no indication that photoperiod had a differential effect on the activity of pouched mice from contrasting habitats.

Circadian rhythms Metabolism Activity Ambient temperature Photoperiod Pouched mice *Saccostomus campestris*

WHEN confronted with a decline in ambient temperature small mammals must increase metabolic heat production if they are to maintain homeothermy (Wunder 1984). Alternatively they can avoid or reduce the energetic costs of cold exposure on a daily basis by limiting activity in the cold and/or by relaxing homeothery and entering bouts torpor (Stebbins 1977, Brahnak and Kramm 1977, Hudson 1978, Wunder 1984). Whilst ambient temperature is the ultimate factor responsible for these changes several studies have shown that photoperiod can also play an important role, firstly as a zeitgeber for daily rhythms of activity (Doucet and Bider 1974, Pratt and Goldman 1986, Figala and Tester 1986) and metabolism (Chew et al. 1965, Sheffield and Andrews 1980, Haim et al. 1988), and secondly as an anticipatory cue for seasonal changes in environmental temperature (Heldmaier et al. 1982, Heath and Lynch 1983, Wunder 1984). In this respect photoperiod is not subject to random fluctuations on the scale exhibited by other environmental factors and is therefore the "most reliable environmental variable which announces the approach of seasonal changes" (Steinlechner and Heldmaier 1982).

However, a series of studies have revealed that different populations of white footed mice (*Peromyscus leucopus*) from high and low latitudes differ in their ability to respond to changes in photoperiod (Lynch et al. 1981, Heath and Lynch 1982, 1983). These studies suggest that the extent to which each population uses photoperiod depends upon the relative severity of seasonal changes in temperature and/or food availability (Lynch et al. 1981). However, because seasonal variation in photoperiod also changes with latitude it remains unclear whether geographical differences in photoresponsivity are a result of differences in the extent of seasonal climatic or photoperiodic changes.

The present study therefore aims to investigate the relative importance of temperature and photoperiod on daily metabolism and activity of two populations of pouched mice (*Saccostomus campestris*) from different localities at similar latitude with contrasting climatic conditions. Pouched mice are nocturnal cricetid rodents (Perrin 1981, Smithers 1983) which display a fundamental circadian rhythm of oxygen consumption entrained to photoperiod (Haim et al. 1988).

METHOD

Animals and Housing

Two populations of pouched mice (*Saccostomus campestris*) from contrasting habitats were examined in the present study. The first comprised animals live-trapped at Vaalkop Dam Nature Reserve in the central Transvaal highveld (25°23'S 27°28'E: 2n=44), the second being two individuals and their offspring caught at Nwanedzi in the eastern Transvaal lowveld (24°23'S 31°40'E: 2n=46). Climatic data for each locality were obtained from the nearest meteorological station and these are summarised in Table 1.

Both populations were held in captivity at Pretoria University (25°45'S 28°12'E) for at least four months prior to experimentation. During this period and throughout the present study the pouched mice were housed separately in standard laboratory rat cages and provided with rat pellets (Epol, Vereeniging) and water ad lib., a diet occasionally supplemented with sunflower seeds, apple and carrots. Shredded paper and sawdust were provided for bedding and their cages were cleaned weekly, and dusted with insecticide to eradicate ectoparasites (KARBADUST: Sentrachem, Silverton. Containing 50g/kg Carbamate). In addition, relative humidity was maintained above 45% to prevent ringtail (Ellison and Westlin-Van Aarde 1990).

Acclimation

At the start of the present study the pouched mice were transferred to a windowless climate chamber (Specht Scientific, Johannesburg) in which ambient temperature (T_a) and photoperiod could be accurately controlled. They were then subjected to a series of four consecutive conditions comprising warm (25°C) and cold (10°C) acclimation under long (14L) and short (10L) photoperiod:
1. Long day - warm: LD 14:10 and 25°C, 2. Short day - warm: LD 10:14 and 25°C
3. Short day - cold: LD 10:14 and 10°C, 4. Long day - cold: LD 14:10 and 10°C.

Following four weeks acclimation to each regime the daily metabolism and activity of six pouched mice from each locality (males:females - Vaalkop 3:3, Nwanedzi 4:2) were recorded as follows:

Daily Metabolism

Oxygen consumption (VO_2) was measured as an indication of metabolic rate (MR) using an open-circuit system (as described by Depocas & Hart 1957, Hill 1972, and Withers 1977: Equation 3a) and assuming an RQ of 0.85. The mice were placed in large (13 l), perspex metabolic chambers, through which a flow of dried air (silica gel) was passed at a rate of 600ml·min^{-1}. The chambers were immersed in a constant temperature water bath (Labotec, Isando), and a thermocouple (J-Type: Grant Instruments, Cambridge) within the chamber was used for monitoring T_a. The water bath was covered by a light-tight hood and long or short photoperiod was maintained using an electric light connected to an automatic time switch.

The pouched mice were transferred to these chambers together with their bedding, food and water (fresh apple), at least 24h prior to the start of measurement to allow them to become habituated to their new surroundings. VO_2 was subsequently recorded every 30 min for a further 24h.

Following full equilibration of the system, the air leaving the chambers was dried and its oxygen content determined using an Ametek S-3A/I oxygen analyser (Applied Electrochemistry, Pittsburgh) connected to a multi-channel data logger (1200 Series Squirrel: Grant Instruments, Cambridge). An in-line, three way valve (Air/Water 350 KPA: Ascoreg, Johannesburg) and time switch alternated the input to the oxygen analyser so that air from two chambers, containing separate pouched mice, could be recorded twice an hour using a single oxygen analyser. The oxygen analyser was calibrated before and after each measurement and all readings were corrected to standard temperature and pressure, dry (STPD). Gross VO_2 was converted to mass specific VO_2 using the mean of body mass measurements taken at the beginning and end of each experiment.

Average daily metabolic rate (ADMR) was calculated as the mean VO_2 measured over a full 24h. Likewise, the mean VO_2 recorded during periods of light and dark was defined as MR_{light} and MR_{dark} respectively.

Daily Activity

The daily activity of each pouched mouse was recorded in the climate chamber using an electronic activity monitor (No.31400: Stoelting, Chicago) connected to an event recorder (A620X: Esterline Angus, Indianapolis). Each movement within the cage was represented by a spike on the recording chart and constant activity produced 120 spikes every 30 min. In this way activity was measured as the number of spikes per 30 min whilst the percentage of time
spent active (%ACT) was calculated as the percentage of 30 min periods in which 1 or more spike occurred. To exclude the effect of disturbance, no one entered the climate chamber for 48h while activity was being measured and only records taken during the second 24h were used for estimating daily activity (Packer 1980).

Statistical Analyses.

All results are presented as mean $\pm$ one standard deviation. Three way analyses of variance (3-ANOVA) were used to statistically compare the independent and interactive effects of ambient temperature, photoperiod and locality on metabolism and activity (Sokal and Rohlf 1981).

RESULTS

Body Mass

The mean body mass (M_b) of pouched mice examined in the present study declined from 91.1 $\pm$13.9g at T_a=25°C to 83.2 $\pm$13.7g at T_a=10°C (see Table 2). However, there was no significant effect of either T_a ($F(1,40)=3.569$, $p>0.05$) or photoperiod on M_b ($F(1,40)=0.000$, $p>0.05$), and there was no significant difference in M_b between the two populations ($F(1,40)=0.825$, $p>0.05$).

Daily Metabolism

The mean metabolic rates of pouched mice from Vaalkop and Nwanedzi recorded over 24h under each acclimatory regime have been summarised in Table 2 and are displayed in Fig.s 1 and 2 respectively. MR varied in phase with photoperiod so that MR_{light}, which approximated resting metabolism, was consistently lower than MR_{dark} recorded during the active period. ADMR, MR_{light} and MR_{dark} were all significantly higher following cold acclimation ($F(1,40)>124.338$, $p<0.001$) whilst there was no significant effect of photoperiod or locality on MR ($F(1,40)<0.749$, $p>0.05$).

However, variation in MR also increased substantially at T_a=10°C compared to T_a=25°C (Table 2), partly because 7 cold acclimated pouched mice (4 Vaalkop and 3 Nwanedzi) displayed daily fluctuations in MR together with periods of hypometabolism which are characteristic of spontaneous torpor (Ellison and Skinner 1991). Only one individual (Vaalkop #1012, see Fig.3) entered torpor under long photoperiod while the remaining 6 animals did so following acclimation to short days.

As seen in the examples displayed in Fig.3, the MR of torpid pouched mice was characterised by a distinct decline in VO_2 during the early morning which fell below 50% of the mean resting metabolism (MR_{light}) of non-torpid animals for more than 2 hours before ending in a sharp increase in MR which overshot resting levels briefly during arousal. Despite the high levels of MR associated with arousal, these short bouts of torpor resulted in a substantial decline in daily energy requirements. Indeed, when compared to the mean ADMR of non-torpid individuals the four torpid pouched mice from Vaalkop saved 36.2%, 28.8%, 22.6% and 21.7% of ADMR while those from Nwanedzi saved 33.1%, 26.9% and 16.4% of ADMR.

TABLE 1. Climatic conditions for pouched mice localities (Weather Bureau, Pretoria)

Locality:	Nwanedzi	Vaalkop Dam
Weather Station:	Satara (24°24'S 31°47'E)	Brits (25°35'S 27°49'E)
Altitude:	275m	1158m
Mean Annual Rainfall:	437mm	621mm
	Air Temperatures (Summer: November - February)	
Mean daily maximum	32.3°C	29.8°C
Mean daily minimum	19.8°C	16.2°C
Mean daily range	12.5°C	13.5°C
Mean lowest monthly	12.6°C	6.6°C
	Air temperatures (Winter: May - August)	
Mean daily maximum	26.2°C	22.3°C
Mean daily minimum	9.2°C	3.0°C
Mean daily range	17.0°C	19.3°C
Mean lowest monthly	1.2°C	-7.9°C

TABLE 2. A summary of metabolic measurements recorded for pouched mice from Vaalkop and Nwanedzi following acclimation to the four different photoperiodic and temperature regimes.

Experimental Condition :	14L:10D & 25°C		10L:14D & 25°C		10L:14D & 10°C		14L:10D & 10°C	
Locality :	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi
Body Mass (g) :	86.1±11.8	91.5±15.7	92.0±18.2	94.8±10.9	79.9±18.0	81.9±11.3	83.2±15.8	88.1±10.6
MR_{light} ($ml O_2 \cdot g^{-1} \cdot h^{-1}$) :	0.862±0.072	0.946±0.077	0.944±0.094	0.908±0.103	1.680±0.365	1.793±0.331	1.824±0.225	1.816±0.257
MR_{dark} ($ml O_2 \cdot g^{-1} \cdot h^{-1}$) :	1.173±0.165	1.189±0.126	1.247±0.151	1.144±0.123	2.067±0.337	2.284±0.434	2.108±0.440	2.288±0.439
ADMR ($ml O_2 \cdot g^{-1} \cdot h^{-1}$) :	0.991±0.111	1.048±0.077	1.123±0.120	1.048±0.112	1.906±0.346	2.080±0.388	1.944±0.310	2.014±0.320

TABLE 3. A summary of activity measurements recorded for pouched mice from Vaalkop and Nwanedzi following acclimation to the four different photoperiodic and temperature regimes.

Experimental Condition :	14L:10D & 25°C		10L:14D & 25°C		10L:14D & 10°C		14L:10D & 10°C	
Locality :	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi	Vaalkop	Nwanedzi
Mean Daily Spike Count (n) :	09.9±02.8	16.4±10.9	22.5±15.4	22.5±08.7	08.6±03.3	11.4±09.5	20.4±18.4	09.5±03.7
Mean Daily %ACT (Arcsin) :	47.4±10.8	57.9±15.8	59.7±17.0	76.4±11.6	49.9±09.0	47.0±15.1	64.9±23.9	53.8±08.2

Daily Activity

The mean number of activity spikes recorded over 24h under each acclimatory regime are also displayed in Fig.s 1 and 2 for pouched mice from Vaalkop and Nwanedzi respectively. From these figures it is clear that *S.campestris* from both localities were predominantly nocturnal, becoming active gradually at the beginning of the dark phase and terminating activity abruptly when the lights went on. However, the activity monitor used in the present study also detected a substantial amount of movement during the light period which indicates that pouched mice also display a considerable amount of diurnal activity presumably conducted exclusively within their nests.

In common with other small rodents (Hamann 1987) there was a high degree of variation in the number of activity spikes displayed by different individuals (see Table 3) which made these data unsuitable for statistical analysis using ANOVA. Instead the percentage of time spent active (%ACT) was subjected to an arcsin transformation (Sokal & Rohlf 1981) and used for analysing the effect of temperature, photoperiod and locality on the activity of pouched mice (see Table 3).

These data revealed a significant interactive effect of temperature and photoperiod on activity ($F(1,40)=9.581$, $p<0.01$) such that %ACT at $T_a=25^oC$ was higher under short photoperiod than under long photoperiod yet the reverse was true at $T_a=10^oC$. Likewise, there was a joint effect of temperature and locality on %ACT ($F(1,40)=5.881$, $p<0.05$) because pouched mice from Vaalkop spent a similar percentage of their time active at both temperatures whilst those from Nwanedzi spent less time active at $T_a=10^oC$ than at $T_a=25^oC$. Meanwhile, there were no independent effects of temperature, photoperiod or locality on %ACT ($F(1,1)<0.243$, $p>0.05$).

DISCUSSION

The results of the present study clearly indicate that, irrespective of photoperiod, *S.campestris* display an increase in metabolic heat production in response to cold exposure. However, some cold acclimated pouched mice achieved substantial reductions in daily energy requirements during bouts of spontaneous torpor, although these savings were effectively masked by the overall increase in metabolism which occurred at $T_a=10^oC$. Nevertheless, because torpor was more common in short day - cold than long day - cold it is evident that photoperiod did exert some, albeit subtle, effect on daily metabolic rate.

By definition, spontaneous torpor occurs as a result of cold stress (Hill 1975, Ellison and Skinner 1991), yet for many species, including *S.campestris*, acclimation to short winter photoperiod is a prerequisite for the full expression of torpor (Heldmaier and Steinlechner 1981, Heath and Lynch 1983, Tannenbaum and Pivorun 1988). Linking the occurrence of torpor to short photoperiod in this way not only ensures that individuals anticipate seasonal declines in temperature, but also guarantees a return to full activity in spring-time, regardless of ambient temperature (Heldmaier and Steinlechner 1981).

Although active metabolism can substitute for thermoregulatory heat production (Hart and Jansky 1963, Mount and Willmott 1967) there was no increase in activity at $T_a=10^oC$ which might have accounted for the higher levels of metabolism observed in the present study. Indeed, activity is an expensive form of heat production because movement alone can cause an increase in heat loss (Hart and Jansky 1963, Mount and Willmott 1967, Wunder 1975) whilst additional thermogenesis is required if animals are active at colder temperatures outside their nests (Pauls 1981, Tannenbaum and Pivorun 1984). For this reason many small mammals actually exhibit a reduction in activity during cold exposure and in this way avoid the thermoregulatory expense of foraging at lower temperatures (Stebbins 1971, Hamann 1987, Doucet and Bider 1974, Marten 1973, Sheffield and Andrews 1980). However, there was also no uniform decline in activity following cold acclimation in the present study. Instead there was an interactive effect of temperature and photoperiod such that pouched mice spent more time active under short photoperiod at $T_a=25^oC$ but less time active under short photoperiod at $T_a=10^oC$. As a nocturnal species one might expect *S.campestris* to become more active during short days (Daan and Aschoff 1975) and this was the case at $T_a=25^oC$. However, at cold temperatures the influence of photoperiod is often overridden by thermoregulatory responses and these can sever the coupling between photoperiod and activity (Figala and Tester 1986, Tester 1987).

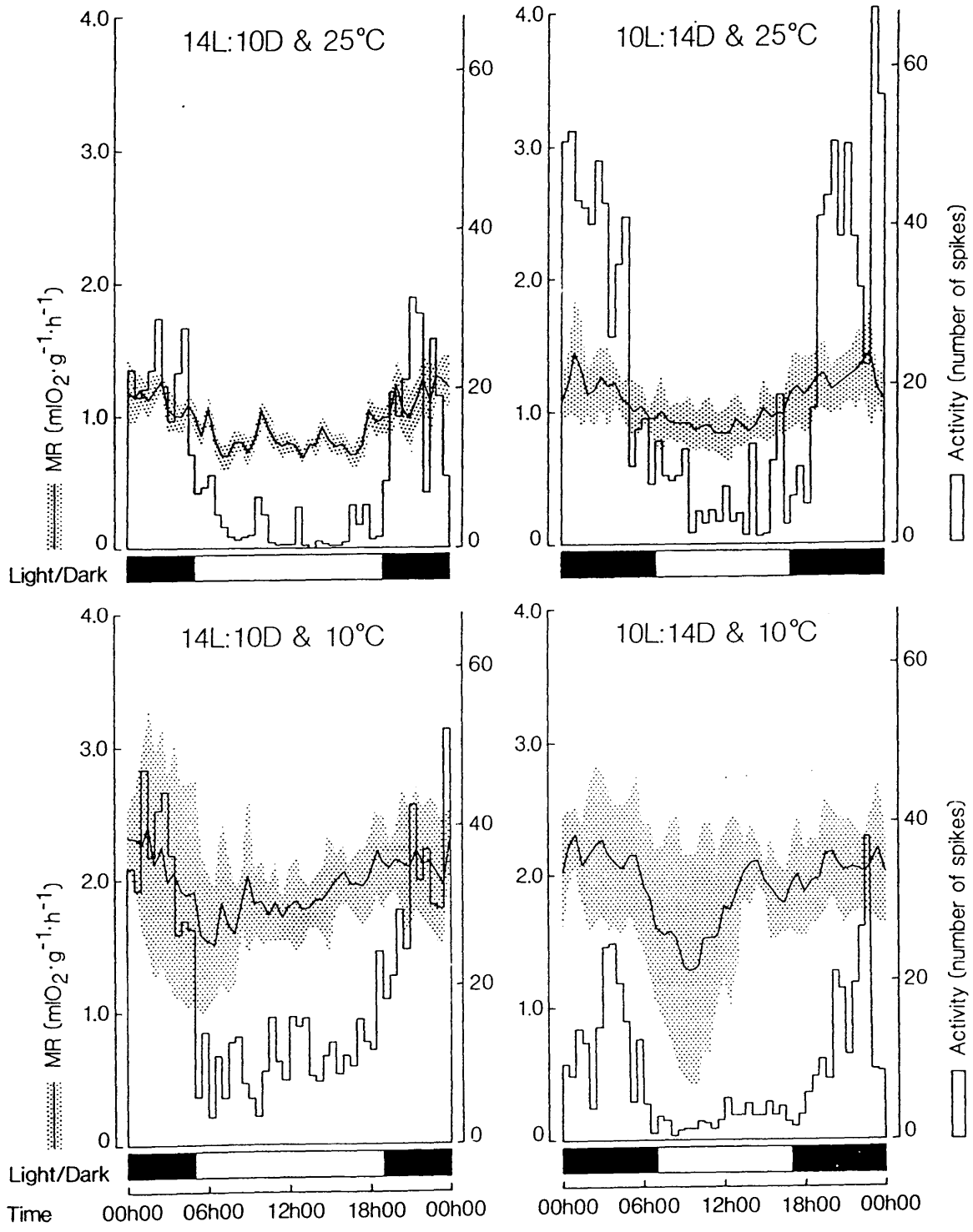

FIG. 1. Mean metabolic rate (---) and activity ([_]) of pouched mice from Vaalkop recorded every 30 min. over 24 hours following acclimation to each of the four different photoperiodic and temperature regimes. The shaded area represents one standard deviation of mean metabolic rate.

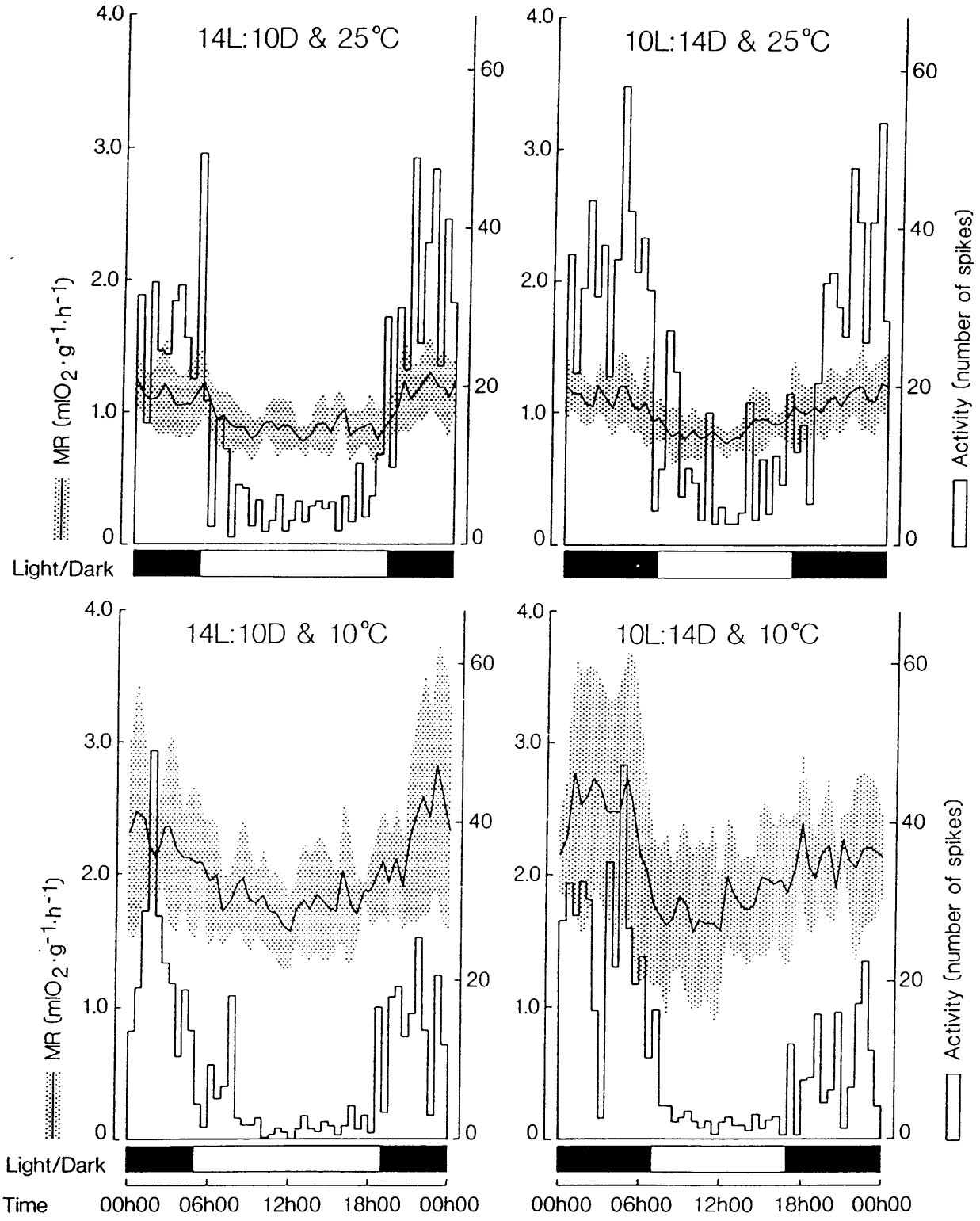

FIG. 2. Mean metabolic rate (---) and activity ([_]) of pouched mice from Nwanedzi recorded every 30 min. over 24 hours following acclimation to each of the four different photoperiodic and temperature regimes. The shaded area represents one standard deviation of mean metabolic rate.

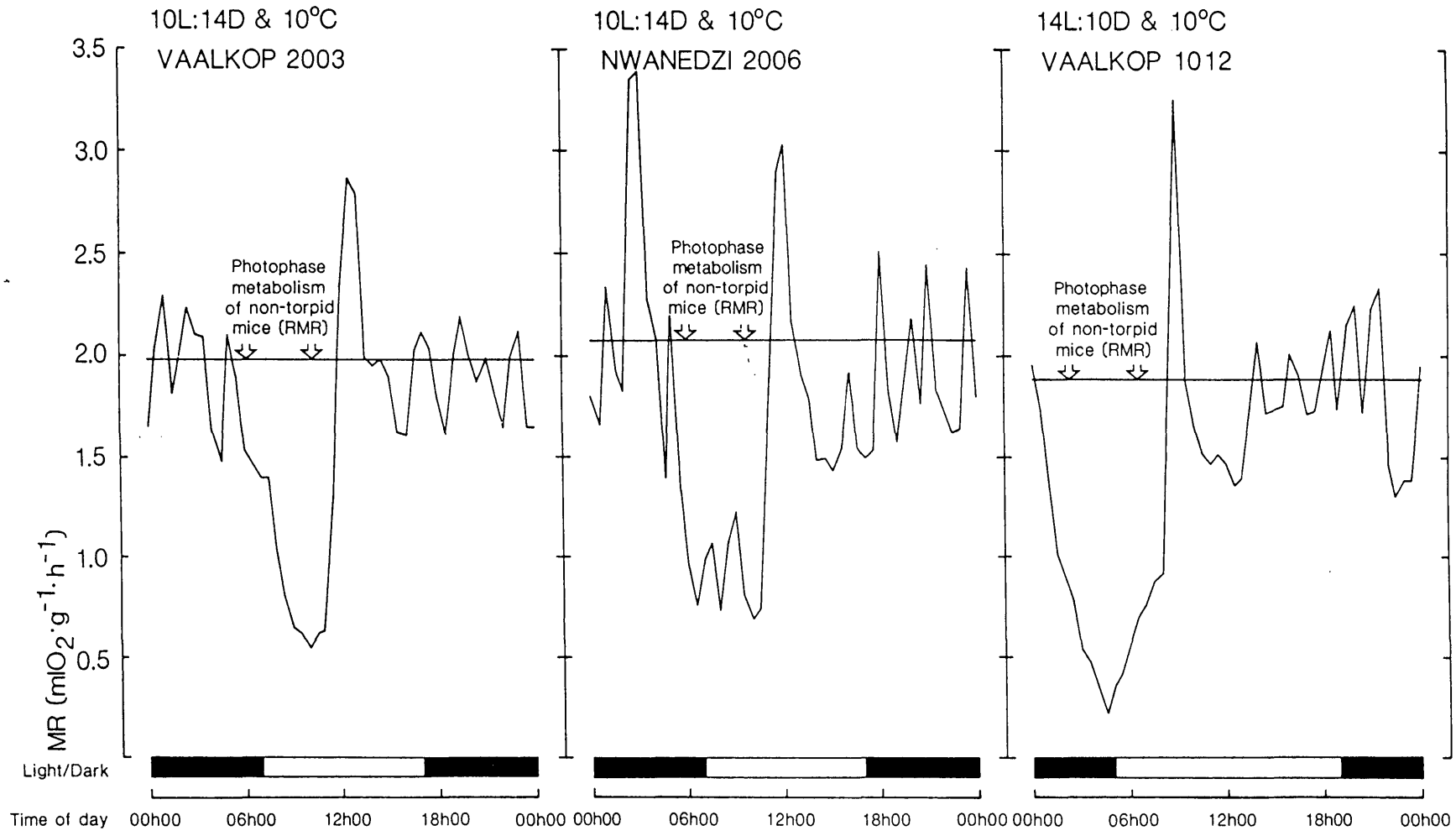

FIG. 3. Daily variation in metabolic rate of 3 representative pouched mice which displayed periods of hypometabolism characteristic of torpor at T_a=10°C. Vaalkop 2003 and Nwanedzi 2006 were 2 of the 6 pouched mice which entered torpor following acclimation to LD 10:14, whilst Vaalkop 1012 was the only individual which exhibited torpor at LD 14:10.

In this way spontaneous torpor might have obscured the effect of photoperiod on the activity of pouched mice at T_a=10°C because torpid animals are generally less active than non-torpid individuals (Lyman 1954, Ruf et al. 1991). This would explain the differential effect of photoperiod on the activity of cold acclimated pouched mice, as (with one exception) both populations only displayed torpor following acclimation to short days.

Meanwhile there was also a joint effect of T_a and locality on the %ACT of *S.campestris* although this wasn't caused by differences in the incidence of torpor because the expression of torpor was similar for both populations. However, inadequate physiological thermoregulation is often compensated by behaviour (Turk and Arnold 1988) and therefore the lower activity of cold acclimated pouched mice from Nwanedzi suggests that these animals avoided cold exposure because they were unable to cope with thermoregulation under these conditions. In fact, when acclimated to 25°C pouched mice from cooler areas, such as Vaalkop, exhibit a greater capacity for nonshivering thermogenesis (NST) (Ellison and Skinner in prep.a) which might permit them to remain active at colder temperatures than those from warmer areas like Nwanedzi. However, pouched mice from both localities display a similar increase in NST capacity following cold acclimation (Ellison and Skinner in prep.b) which indicates that the differences in activity observed in the present study weren't the result of differences in thermogenetic ability, but simply reflect intraspecific variation in their behavioural response to cold. Indeed, similar differences in activity have been reported for closely related *Phodopus* species (Hamann 1987). Presumably these differences are innate because they were still present under controlled laboratory conditions. However, they may have been the result of prior exposure to the different climatic conditions prevailing in each locality as early cold exposure can produce permanent effects on behavioural thermoregulation (Lynch et al. 1976).

In conclusion, the present study indicates that ambient temperature has an overriding effect upon the daily metabolism of pouched mice, although both photoperiod and locality can, to some extent, modify their physiological and behavioural responses to cold. However, there was no indication that photoperiod had a differential effect on the daily metabolism or activity of pouched mice from contrasting localities in southern Africa.

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REFERENCES

Bahnak, B. R.; Kramm, K. R. The influence of environmental temperature and photoperiod on activity in the red squirrel, *Tamiascuirus hudsonicus*. Int. J. Biometeorol. 21:348-356; 1977.

Chew, R. M.; Lindberg, R. G.; Hayden, P. Circadian rhythm of metabolic rate in pocket mice. J. Mamm. 46:477-494; 1965.

Daan, S.; Aschoff, J. Circadian rhythms of locomotor activity in captive birds and mammals: their variations with season and latitude. Oecologia 18:269-316; 1975.

Depocas, F.; Hart, J. S. Use of the Pauling oxygen analyzer for measurements of oxygen consumption of animals in open-cicuit system and in short-lag, closed-circuit apparatus. J. Appl. Physiol. 10:388-392; 1957.

Doucet, G. J.; Bider, J. R. The effects of weather on the activity of the masked shrew. J. Mamm. 55:348-363; 1974.

Ellison, G. T. H.; Skinner, J. D. The influence of ambient temperature on spontaneous daily torpor of pouched mice (*Saccostomus campestris*: Rodentia - Cricetidae) from southern Africa. J. therm. Biol. in press 1991.

Ellison, G. T. H.; Skinner, J. D. Intraspecific variation in thermoregulation and energetics of pouched mice (*Saccostomus campestris*: Cricetidae) from southern Africa. in prep. a.

Ellison, G. T. H.; Skinner, J. D. The relative importance of photoperiod and temperature as seasonal thermoregulatory cues for pouched mice (*Saccostomus campestris*: Cricetidae) from southern Africa. in prep. b.

Ellison, G. T. H.; Westlin-van Aarde, L. M. Ringtail in the pouched mouse (*Saccostomus campestris*). Lab. Anim. 24:174-175; 1990.

Figala, J.; Tester, J. R. Comparison of seasonal rhythms of grey squirrels (*Scuirus caroliensis*, Rodentia) in captivity and in the wild. Vest. cs. Spolec. zool. 50:33-48; 1986.

Haim, A.; Ellison, G. T. H.; Skinner, J. D. Thermoregulatory circadian rhythms in the pouched mouse (*Saccostomus campestris*). Comp. Biochem. Physiol. A 91:123-127; 1988.

Hamann, Von U. Zu Aktivität und Verhalten von drei Taxa der Zwerghamster der Gattung *Phodopus* Miller, 1910. Z. Säugetierkunde 52:65-76; 1987.

Hart, J. S.; Jansky, L. Thermogenesis due to exercise and cold in warm- and cold-acclimated rats. Can. J. Biochem. Physiol. 41:629-634; 1963.

Heath, H. W.; Lynch. G. R. Intraspecific differences for melatonin-induced reproductive regression and the seasonal molt in *Peromyscus leucopus*. Gen. Comp. Endocrinol. 48:289-295; 1982.

Heath, H. W.; Lynch, G. R. Intraspecific differences in the use of photoperiod and temperature as environmental cues in white-footed mice *Peromyscus leucopus*. Physiol. Zool. 56:506-512; 1983.

Heldmaier, G.; Steinlechner, S. Seasonal pattern and energetics of short daily torpor in the Djungarian hamster, *Phodopus sungorus*. Oecologia 48, 265-270; 1981.

Heldmaier, G.; Steinlechner, S.; Raphael, J.; Latteier, B. Photoperiod and ambient temperature as environmental cues for seasonal thermogenic adaptation in the Djungarian hamster, *Phodopus sungorus*. Int. J. Biometeorol. 26:339-345; 1982.

Hill, R. W. Determination of oxygen consumption by use of the paramagnetic oxygen analyzer. J. Appl. Physiol. 33:261-263; 1972.

Hill, R. W. Daily torpor in *Peromyscus leucopus* on an adequate diet. Comp. Biochem. Physiol. 51A:413-423; 1975.

Hudson, J. W. Shallow daily torpor: a thermoregulatory adaptation. In: Wang, L. C. H.; Hudson, J. W., eds. Strategies in cold. London: Academic Press; 1978:67-108.

Lyman, C. P. Activity, food consumption and hoarding in hibernators. J. Mamm. 35:545-552; 1954.

Lynch, G. R.; Lynch, C. B.; Dube, M.; Allen, C. Early cold exposure: Effects on behavioural and physiological thermoregulation in the house mouse, *Mus musculus*. Physiol. Zool. 49:191-199; 1976.

Lynch, G. R., Heath, H. W.; Johnston, C. M. Effect of geographic origin on the photoperiodic control of reproduction in the white-footed mouse, *Peromyscus leucopus*. Biol. Reprod. 25:475-480; 1981.

Marten, G. G. Time patterns of *Peromyscus* activity and their correlations with weather. J. Mamm. 54:169-188; 1973.

Mount, L. E.; Willmott, J. V. The relation between spontaneous activity, metabolic rate and the 24 hour cycle in mice at different environmental temperatures. J. Physiol. 190:371-380; 1967.

Packer, W. C. Nest building and activity patterns in four sympatric rodent species. S. Afr. J. Zool. 15:50-55; 1980.

Pauls, R. W. Energetics of the red squirrel; a laboratory study of the effects of temperature, seasonal acclimatization, use of nest and exercise. J. therm. Biol. 6:79-86; 1981.

Perrin, M. R. Notes on the activity patterns of 12 species of southern African rodents and a new design of activity monitor. S. Afr. J. Zool. 16:248-258; 1981.

Pratt, B. L.; Goldman, B. D. Activity rhythms and photoperiodism of Syrian hamsters in a simulated burrow system. Physiol. Behav. 36:83-89; 1986.

Ruf, T.; Klingenspor, M.; Preis, H.; Heldmaier, G. Daily torpor in the Djungarian hamster (*Phodopus sungorus*): interactions with food intake, activity and social behaviour. J. Comp. Physiol. B 160:609-615; 1991.

Sheffield, M. V.; Andrews, R. V. Interactions of ambient temperature and photoperiod on deer mouse energetics. Comp. Biochem. Physiol. 67A:103-115; 1980.

Smithers, R. H. N. The Mammals of the Southern African Subregion. Pretoria: University of Pretoria Press; 1983:283-284.

Sokal, R. R.; Rohlf, F. J. Biometry. San Francisco: W. H. Freeman and Co.; 1981:1-859.

Stebbins, L. L. Seasonal variation in circadian rhythm of deermice in northwestern Canada. Arctic 24:124-131; 1971.

Stebbins, L. L. Some aspects of over-wintering in *Peromyscus maniculatus*. Can. J. Zool. 56:386-390; 1977.

Steinlechner, S.; Heldmaier, G. Role of photoperiod and melatonin in seasonal acclimatization of the Djungarian hamster, *Phodopus sungorus*. Int. J. Biometeor. 26:329-337; 1982.

Tannenbaum, M. G.; Pivorun, E. B. Differences in daily torpor patterns among three southwestern species of *Peromyscus*. J. Comp. Physiol. B 154:233-236; 1984.

Tannenbaum, M. G.; Pivorun, E. B. Seasonal study of daily torpor in southeastern *Peromyscus maniculatus* and *Peromyscus leucopus* from mountains and foothills. Physiol. Zool. 61:10-16; 1988.

Tester, J. R. Changes in daily rhythms of some free-ranging animals in Minnesota. Can. Fld. Nat. 101:13-21; 1987.

Turk, A.; Arnold, W. Thermoregulation as a limit to habitat use in alpine marmots (*Marmota marmota*). Oecologia 76:544-548; 1988.

Withers, P. C. Measurement of VO_2, VCO_2, and evapourative water loss with a flow-through mask. J. Appl. Physiol. 42:120-123; 1977.

Wunder, B. A. Strategies for, and environmental cueing mechanisms of, seasonal changes in thermoregulatory parameters of small mammals. In: Merritt, J. F., ed. Winter ecology of small mammals. Carnegie Mus. Nat. Hist. Spec. Publ. 10:165-172; 1984.

Wunder, B. A. A model for estimating metabolic rate of active or resting mammals. J. Theor. Biol. 49:345-354; 1975.

REPRODUCTIVE FLEXIBILITY IN FEMALE POUCHED MICE (*SACCOSTOMUS CAMPESTRIS*: CRICETOMYINAE)

By

L. M. WESTLIN* AND G.T.H. ELLISON

Mammal Research Institute, Pretoria University, Pretoria 0002, South Africa

* Current address: Department of Theriogenology, Faculty of Veterinary Medicine, Pretoria University, Pretoria 0002, South Africa

ABSTRACT

The present review examined variation in: 1. the onset of puberty, 2. the oestrous cycle, 3. the litter interval, and 4. the litter size of pouched mice (*Saccostomus campestris*) housed under "optimal" conditions (14h photoperiod at 22±2°C with *ad libitum* food and water) in order to assess whether this species can manipulate these critical components of reproduction. Neither puberty nor the oestrous cycle displayed any flexibility while the litter interval appeared to be minimal under these conditions. Although litter size was variable among wild individuals a similar degree of variability was displayed by captive individuals under optimal conditions which suggests that litter size is random and cannot be controlled. It therefore appears that reproductive flexibility in female pouched mice is negligible and that the reproductive rate of wild individuals would depend primarily upon the duration of suitable, "optimal" conditions while behavioural factors associated with finding a mate could also play an important role.

INTRODUCTION

Ultimately, an individual's fitness is determined by the number of successfully reproducing offspring it leaves in the next generation (Parker 1984) and therefore the quality and quantity of reproductive output must be maximal if an individual is to optimise its fitness (Krebs & McCleery 1984). However, for female mammals each reproductive event demands a large increase in energy expenditure which can adversely affect future reproductive potential (Thompson & Nicoll 1986). For this reason, one would expect female mammals to display mechanisms which enable them to modify the timing and extent of their investment in reproduction. Such reproductive flexibility might be provided by manipulating one or more of four components of reproduction, ie: 1. the onset of puberty, 2. the oestrous cycle, 3. the interval between successive litters, and 4. litter size.

The pouched mouse (*Saccostomus campestris*) is a solitary cricetid rodent which is widely distributed in sub-saharan Africa (Smithers 1983). In southern Africa this species is strongly associated with arid and semi-arid woodland savannas (Rautenbach 1978) which are seasonal environments where suitable conditions for reproduction can be unpredictable (Korn 1989). Although pouched mice are seasonal breeders in the wild (Smithers 1983), they breed all year round under "optimal" laboratory conditions (14h photoperiod at 22±2°C with food and water *ad libitum*; Westlin-van Aarde 1988). Under these conditions animals lived for at least 3 years but only bred successfully during their first two years when they produced an average of 6.9 litters per year. With a maximum litter size of 13 (Westlin-van Aarde 1988) each female has the potential to produce 179 offspring under "optimal" conditions (2 years x 6.9 litters x 13 young).

The aim of the present review was to determine the scope for reproductive flexibility in female pouched mice by examining their capacity to modify reproductive activity under "optimal" conditions in captivity. The evidence for manipulation by *S.campestris* of critical components of reproduction will now be reviewed.

1. MODIFICATION OF THE AGE AT PUBERTY

In several rodent species the onset of puberty is known to be accelerated by stimuli from an adult male, whilst the presence of an adult female has been shown to suppress reproductive maturity in some group-living species (Vandenbergh 1983). However, among juvenile female *S.campestris* raised on their own, in groups of 5 or singly with an adult male, there were no significant differences in the age at which their vagina opened, age at first oestrus or age at first conception (Westlin-van Aarde 1989b). Likewise there was no delay in the age at first conception of juvenile females housed singly with an adult female despite intense aggression and frequent attacks from their older conspecifics (Westlin-van Aarde 1989b). It therefore appears that the onset of puberty in female pouched mice is fixed under optimal conditions.

2. ALTERATION OF THE OESTROUS CYCLE

The onset and duration of oestrus can also be modified by the presence of conspecifics and many studies have shown that the oestrous cycle of different rodent species can be either lengthened or shortened following exposure to males or females (Lombardo & Terman 1980, Breed 1976). However, olfactory signals from conspecifics might also be able to stimulate or defer the onset of oestrus, particularly in seasonal breeding species like pouched mice (Bronson & Dezell 1968, Aron 1979, Dominic & Pandey 1979, Sànchez-Criado 1982, McClintock 1983). To investigate this possibility the onset of oestrus was determined in 21 wild-caught, anoestrous females after they were housed under "optimal" conditions and placed in cages containing either clean bedding or bedding soiled by unrelated males or females (unpublished data). Of those females which exhibited oestrus 4 out of 21 did so when housed in cages containing clean bedding, while 2 of 8 and 4 of 9 did so during exposure to soiled bedding from female and male cages respectively. However, there was no significant effect of soiled bedding (X^2=0.578, d.f.=1, p>0.20) or sex of previous occupant (X^2=1.811, d.f.=1, p>0.10) on the proportion of females entering oestrus. Similarly, Westlin-van Aarde (1989b) found no effect of males or females on the oestrous cycle pattern in pouched mice which suggests that the onset and duration of oestrus is also inflexible in this species.

3. ADJUSTMENT OF THE LITTER INTERVAL

Variation in the gestation period of *S.campestris* is minimal in captivity (20-22 days) and of 36 pregnant females examined only one gave birth after 20 days and 7 after 22 days, the majority (n=28, 78%) giving birth after 21 days (Westlin-van Aarde 1988). Thus variability in gestation period provides little scope for adjusting the litter interval in this species. At the same time, pouched mice enter a lactational anoestrous after parturition and although lactation typically lasts only 25 days (Westlin-van Aarde 1988) Earl (1978) reported that pups would suckle for up to 55 days when kept with their mothers and this would undoubtedly delay subsequent pregnancies. However, pouched mice start eating solid food at 13-17 days old (Earl 1977, Westlin-van Aarde 1989a) and wild individuals probably disperse from the maternal burrow when they are 20-30 days old because individuals weighing less than 20g (ca. 25 days old: Westlin-van Aarde 1989a) have been collected by a number of Museums in southern Africa (Transvaal Museum, Pretoria, South Africa; State Museum, Windhoek, Namibia; Natural History Museum, Bulawayo, Zimbabwe). For this reason there is no reason to suggest that lactation is extended under natural conditions. Meanwhile, female pouched mice exhibit their first oestrus after 28 $\pm$1.4 days of lactational anoestrus when their pups are weaned at 25 days old, although of 12 individuals examined, only 5 became pregnant at this time, and the remaining 7 females underwent one or two cycles without mating, or mated once or twice without conceiving (Westlin-van Aarde 1988). Therefore, some females exhibit a delay of 2-7 days between the onset of oestrus and pregnancy, but this is insignificant when compared to the duration of gestation ($\pm$21 days) or lactational anoestrus ($\pm$25 days). However, there is evidence that this period can be extended even after the pups are weaned because Westlin-van Aarde (1989b) found that pregnancy was blocked in 7 out of 8 females when the stud male was replaced with a strange male, whilst pregnancy was blocked up to 4 times when females were exposed to a series of different males. It therefore appears that the litter interval can be extended by some social factors although there is very little scope to shorten the litter interval under "optimal" conditions.

4. MANIPULATION OF LITTER SIZE

Undoubtedly the most direct avenue for modifying reproductive output is the manipulation of litter size, and circumstantial evidence from museum archives and published reports (summarised in Table 1) certainly indicates that wild pouched mice display considerable variation in litter size. This variation may be due to any one of 4 possible causes: the number of oocytes fertilised, pre-implantation loss, post-implantation loss and peri-natal death. In captivity pre-implantation loss (as indicated by the higher number of corpora lutea than embryos) was estimated at only 7.91% (Westlin-van Aarde 1988).

Table 1. Variation in the number of foetuses reported for *Saccostomus campestris*

LOCALITY	MEAN NUMBER OF FOETUSES	RANGE	SAMPLE SIZE	SOURCE
East Africa	5	4 - 6	2	Heller in Shortridge 1934
Amudat, Uganda	7	-	1	Delany 1964
Kenya	5	4 - 6	2	Hollister 1919 in Delany 1975
Malawi	5.1	2 - 9	10	Hanney 1965
Kabompo, Zambia	7	-	1	Ansell 1960
Chipata, Zambia	6	-	1	Ansell 1960
Ngoma, Zambia	5	-	1	Sheppe 1973
Zimbabwe	6.7	1 - 10	7	Smithers and Wilson 1979
Botswana	7.4	5 - 10	8	Smithers 1971
Bushmanland, Namibia	9.5	8 - 11	2	Windhoek State Museum
Okahandja, Namibia	8	6 - 10	2	Windhoek State Museum
Gobabis, Namibia	9	7 - 11	2	Windhoek State Museum
Gobabis, Namibia	8	-	1	Shortridge 1934
Levuhvu, RSA	5.5	4 - 7	2	Transvaal Museum
Steilloopbrug, RSA	7.5	6 - 9	2	Transvaal Museum
Potgietersrus, RSA	5.8	3 - 8	4	Transvaal Museum
Vaalwater, RSA	5	-	1	Transvaal Museum
Roossenekal, RSA	6	-	1	Transvaal Museum
Lake Sibaya, RSA	5.4	3 - 8	5	Transvaal Museum
Shihangwane, RSA	6	-	1	Transvaal Museum
Mean:	6.4	1 - 11	56	

However, the same study showed that post-implantation loss was negligible in *S.campestris* and this is confirmed by the similarity between the number of foetuses found in wild individuals and the litter size of females in captivity (see Table 2 below). At the same time, peri-natal death (0-2 days after birth) was 10.6% and declined rapidly after 2 days of age (Westlin-van Aarde 1988). It therefore appears that variability in litter size was due to pre-implantation loss, although the higher number of corpora lutea than embryos reported by Westlin-van Aarde (1988) might also be the result of the accumulation of corpora lutea from previous ovulations.

Table 2. Variation of litter size among captive *Saccostomus campestris*

LOCALITY	MEAN LITTER SIZE	RANGE	SAMPLE SIZE	SOURCE
?	4.3	-	-	Keogh pers.comm. in Earl 1980
?	6	3 - 10	-	Pitchford and Visser 1970
Ngoma, Zambia	4	-	1	Wrangham 1969
Gam, Namibia	5	-	1	Present study
Letaba, RSA	4.7	1 - 7	7	Present study
Satara, RSA	8.3	5 - 10	4	Present study
Derdepoort, RSA	4.9	2 - 8	44	Earl 1980
Skukuza, RSA	9	-	1	Present study
Brits, RSA	6	-	1	Present study
Komatipoort, RSA	7.6	3 - 13	45	Westlin-van Aarde 1988
Sishen, RSA	5.5	5 - 6	4	Present study
Barkly West, RSA	7	-	1	Present study
Pofadder, RSA	6	5 - 7	2	Present study
Beaufort West, RSA	6	-	1	Present study
Mean:	6.2	1 - 13	114	

Notes - 1. Litter sizes from Skukuza and Barkly West are based upon the number of pups found within excavated burrow systems.
2. Pouched mice from Komatipoort had been in captivity since 1959.

For this reason it remains unclear what mechanism(s) is(are) responsible for the variability in litter size. At the same time it is also not clear whether this variation can be controlled by female pouched mice particularly as substantial variability was still evident under "optimal" laboratory conditions when one might expect females to produce the maximum number of offspring (Westlin-van Aarde 1988). Although other rodents display an increase in litter size with parity (Cowan and Arsenault 1954, Frank 1956, Kalela 1957, Negus and Pinter 1965, Richmond and Conaway 1969, Hasler and Banks 1975, Gustafsson, Andersson and Westlin 1980) neither age nor parity influences litter size in *S.campestris* (Westlin-van Aarde 1988) and consequently, the variation in litter size observed appears random in this species.

CONCLUSION

This review indicates that the onset of puberty and the expression of oestrus are fixed in female pouched mice housed under "optimal" conditions. Likewise, litter size appears to be random in *S.campestris* because similar variability in litter size is observed in wild and captive individuals. Meanwhile, litter interval was minimal under controlled conditions although the presence of suckling pups can prolong the period of lactational anoestrus while unfamiliarmales are able to repeatedly block pregnancy. It therefore appears that reproductive flexibility in female pouched mice is negligible and that the reproductive rate of wild individuals would depend primarily upon the duration of suitable, "optimal" conditions while behavioural factors associated with finding a mate could also play an important role.

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REFERENCES

ANSELL, W.F.H. 1960. Mammals of northern Rhodesia. Government Printer, Lusaka.

ARON, C. 1979 Mechanisms of control of the reproductive function of olfactory stimuli in female mammals. *Physiol. Rev.* **59**: 229-282.

BREED, W.G. 1976. Effect of environment on ovarian activity of wild hopping mice (*Notomys alexis*). *J. Reprod. Fert.* **47**: 395-397.

BRONSON, F.H. & DEZELL, H.E. 1968. Studies on the estrus-inducing (pheromonal) action of male deer mouse urine. *Gen. comp. Endocrinol.* **10**: 339-343.

DELANY, M.J. 1964. A study of the ecology and breeding of small mammals in Uganda. *Proc. zool. Soc. Lond.* **142**: 347-370.

DELANY, M.J. 1975. The rodents of Uganda. Trustees of the British Museum (Natural History), London.

DOMINIC, C.J. & PANDEY, S.D. 1979. Suppression of oestrus-inducing pheromone production in the Indian field mouse, *Mus booduga* Gray, by treatment with antiandrogen. *Endokrinologie* **75**: 1-5.

COWAN, I.McT. & ARSENAULT, M.G. 1954. Reproduction and growth in the creeping vole, *Microtus oregoni serpens* Merriam. *Can. J. Zool.* **32**: 198-208.

EARL, S.M.J.A. 1980. Development of behaviour in the pouched mouse *Saccostomus campestris.* M.Sc. Thesis, University of Pretoria, Pretoria, South Africa.

EARL, Z. 1978. Postnatal development of *Saccostomus campestris*. *African Small Mammal Newsletter* **2**: 10-12.

FRANK, F. 1956. Beiträge zur Biologie der Feldmaus, *Microtus arvalis* (Pallas). *Lab. Zool. Jahrb. (Syst.)* **84**: 32-74.

GUSTAFSSON, T., Andersson, B. & Westlin, L. 1980. Reproduction in a laboratory colony of bank vole, *Clethrionomys glareolus*. *Can. J. Zool.* **58**: 1016-1021.

HANNEY, P. 1965. The Muridae of Malawi. *J. Zool. Lond.* **146**: 577-633.

HASLER, J.F. & BANKS, E.M. 1975. Reproductive performance and growth in captive collared lemmings (*Dichrostonyx groenlandicus*). *Can. J. Zool.* **53**:777-787.

HOLLISTER, N. 1919. East African mammals in the US National Museum II. Rodentia, Lagomorpha and Tubulidentata. *Bull. U.S. natn. Mus.* **99**: 1-184.

KALELA, O. 1957. Regulation of reproductive rate in subarctic populations of the vole *Clethrionomys rufocanus* (Sund.). *Ann. Acad. Sci. Fenn. Ser. A4* **34**: 1-60.

KORN, H. 1989. The annual cycle in body weight of small mammals from the Transvaal, South Africa, as an adaptation to a subtropical seasonal environment. *J. Zool. Lond.* **218**: 223-231.

KREBS, J.R. & McCleery, R.H. 1984. Optimization in Behavioural Ecology. *In:* Behavioural ecology - An evolutionary approach. Eds. Krebs, J.R. & Davies, N.B. Blackwell Scientific, Oxford.

LOMBARDO, D.L. & TERMAN, C.R. 1980. The influence of the social environment on sexual maturation of female deer mice (*Peromyscus maniculatus bairdi*). *Res. Popul. Ecol. Kyoto* **22**: 93-100.

McCLINTOCK, M.K. 1983. Pheromonal regulation of the ovarian cycle: enhancement, suppression and synchrony. *In:* Pheromones and reproduction in mammals. Ed. Vandenbergh, J.G. Academic Press, New York.

NEGUS, N.C. & PINTER, A.J. 1965. Litter sizes of *Microtus montanus* in the laboratory. *J. Mammal.* **46**: 434-437.

PARKER, G.A. 1984. Evolutionary stable strategies. *In:* Behavioural ecology - An evolutionary approach. Eds. Krebs, J.R. & Davies, N.B. Blackwell Scientific, Oxford.

PITCHFORD, R.J. & VISSER, P.S. 1970. *Saccostomus campestris* Peters 1846 as a laboratory animal. *J. S. Afr. Vet. med. Assn.* **41**: 173-174.

RAUTENBACH, I.L. 1978. A numerical re-appraisal of the southern African biotic zones. *Bull. Carnegie. Mus. Nat. Hist.* **6**:175-187.

RICHMOND, M.E. & CONAWAY, C.H. 1969. Management, breeding and reproductive performance of the vole, *Microtus ochrogaster*, in the laboratory. *Lab. Anim. Care* **19**: 80-87.

SANCHEZ-CRIADO, J.E. 1982. Involvement of the vomero-nasal system in the reproductive physiology of the rat. In: Olfaction and endocrine regulation. Ed. Breipohl, W. IRL Press, London.

SHEPPE, W.A. 1973. Notes on Zambian rodents and shrews. Puku 7: 167-190.

SHORTRIDGE, G.C. 1934. The mammals of South West Africa. Heinemann, London.

SMITHERS, R.H.N. 1971. Mammals of Botswana. *Nat. Mus. Rhodesia Mem.* **4**: 1-340.

SMITHERS, R.H.N. 1983 The mammals of the southern African subregion. Pretoria University, Pretoria.

SMITHERS, R.H.N. & WILSON, V.J. 1979. Checklist and atlas of the mammals of Zimbabwe Rhodesia. *Nat. Mus. Rhodesia Mem.* **8**: 1-147.

THOMPSON, S.D. & NICOLL, M.E. 1986. Basal metabolic rate and energetics of reproduction in therian mammals. *Nature (London)* **321**:690-693.

VANDENBERGH, J.G. 1983. Pheromones and Reproduction in Mammals. Academic Press, New York.

WESTLIN-VAN AARDE, L.M. 1988. Reproduction in a laboratory colony of the pouched mouse, *Saccostomus campestris*. *J. Reprod. Fert.* **83**: 773-778.

WESTLIN-VAN AARDE, L.M. 1989a. Pre- and post-natal development of pouched mice, *Saccostomus campestris*. *J. Zool. Lond.* **218**: 497-501.

WESTLIN-VAN AARDE, L.M. 1989b. Social environment and reproduction in female pouched mice, *Saccostomus campestris*. *J. Reprod. Fert.* **86**:367-372.

WRANGHAM, R.W. 1969. Captivity behaviour and post-natal development of the Cape pouched mouse, *Saccostomus campestris* Peters. *Puku* **5**: 207-210.