

https://dspace.org/

Costs and benefits of solitary living in the bush Karoo rat (Otomys unisulcatus)

Makuya, Lindelani

2024-10

University of the Witwatersrand, Johannesburg

https://hdl.handle.net/10539/46865

Makuya, Lindelani. (2024). Costs and benefits of solitary living in the bush Karoo rat (Otomys unisulcatus). [PhD thesis, University of the Witwatersrand, Johannesburg]. WIReDSpace. https://hdl.handle.net/10539/46865

Downloaded from a DSpace repository. https://dspace.org/



Costs and benefits of solitary living in the bush Karoo rat (*Otomys unisulcatus*)

by

Lindelani Makuya

(607832)

Submitted in fulfilment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

in the Faculty of Science at the University of the Witwatersrand, Johannesburg

DECLARATION

I hereby declare that this thesis is my own unaided work, and that recognition has been given to the references used and assistance received. It is being submitted for the Degree of Doctor of Philosophy in the Faculty of Science at the University of the Witwatersrand. It has not been submitted for any degree or examination at any other University.



Lindelani Makuya
18 October 2024

ABSTRACT

Solitary living has traditionally been regarded as the ancestral as well as most common and most primitive form of social organisation in mammals. However, recent comparative studies indicate that solitary living is not ancestral for all mammalian orders, and that solitary living is an adaptation to the environment. I show that solitary living occurs in only 22% of studied mammalian species and argue that the main benefit of solitary living is to avoid the costs of group-living. Group-living has been explained using socio-ecological models, and I used the same factors of resource distribution and predation risk to develop a socio-ecological model explaining solitary living. To reach a better understating of solitary living, I studied the social system of free-living bush Karoo rats *(Otomys unisulcatus*) in the Succulent Karoo semi-desert of South Africa. I used trapping and marking, focal animal observations and mini-GPS dataloggers that I fitted simultaneously to neighbouring females. I found that 96% of female bush Karoo rats were solitary living, with social groups of two or three individuals occurring occasionally. Groups always consisted of close kin, typically females. The home ranges of kin neighbours overlapped more than those of non-kin. Neighbours were, however, attracted to the same foraging grounds, irrespective of relatedness. Females tolerated one another at shared foraging grounds, which could be interpreted as by-product mutualism, a simple form of cooperation. I recorded interactions between neighbouring bush Karoo rats both in a neutral test arena and in the field to investigate whether solitary living was due to aggression and social intolerance. Social interactions between neighbours were rare and aggression was rare in neutral arena tests. However, female bush Karoo rats were more aggressive towards non-kin intruders in the field tests. Finally, the relationship between mother and offspring remained amicable even after the offspring had dispersed from the lodges, indicating that maternal aggression was not the mechanism that led to offspring dispersal and solitary living. In conclusion, I showed that solitary living is not always characterised by aggression and avoidance, but rather that solitary species can have non-random and individualised social interactions that are influenced by kinship.

ACKNOWLEDGEMENTS

I am very grateful to Prof. Neville Pillay and Dr. Carsten Schradin for all their guidance, patients and supervision with the project and my thesis. I would not have made it so far if they were not so patient with me.

I appreciate the financial support I received from the WITS-CNRS Joint PhD programme and the National Research Foundation (Doctoral Grant Holder Bursary number 120857).

This thesis would also not have been possible without the assistance from the Succulent Karoo Research Station, by providing all the equipment, infrastructure and the animals to make the project possible. I further specially thank the cohort of research assistants I worked with over the years (Thandolwethu, Philisa, Emily, Tammy, Elena, Siya, Perfect, Nkululeko, Precious). I would also like to thank Kate Smorenburg, Lucie Lenoël and Johannes Meka for proof reading and editing parts of the thesis. This greatly helped to make the thesis read better. A special thanks to Siyabonga Sangweni and Adélie Destercke for helping with data analysis. I am thankful for comments by C.H. Fleming, M. Enstipp and C. Catoni which significantly improved the data chapters.

This study was made possible by the administrative and technical support of the Succulent Karoo Research Station (registered South African NPO 122-134). I am grateful for the support of Goegap Nature Reserve and its staff as well as the Northern Cape Department of Environment & Nature Conservation. This study was supported by the CNRS and the University of the Witwatersrand. All protocols were approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (AESC clearance numbers 2003/34/3, 2018-03-15B, 2021/05/02/B and 22-12-024B).

Finally, I would like to thank my family and friends for believing in me, helping me to believe in myself.

Table of Contents

DECLARATION ..... **i**

ABSTRACT ..... **ii**

ACKNOWLEDGEMENTS ..... **iii**

List of figures v

List of tables ix

Glossary xi

CHAPTER 1. General introduction 1

CHAPTER 2: Costs and benefits of solitary living 10

CHAPTER 3. Measuring range sizes in a 100 g rodent: mini-GPS are more reliable than transmitters, but the location error reduces reliability. 30

CHAPTER 4. Kin based spatial structure in a solitary small mammal as indicated by GPS dataloggers. . 53

CHAPTER 5. Tolerant mothers: aggression does not explain solitary living in the bush Karoo rat 78

CHAPTER 6. Shared feeding grounds in a solitary rodent: an indication for cooperation by kin selection and mutualism? 112

CHAPTER 7. General Discussion 127

APPENDIX 142

List of figures

Chapter 1

Figure 1. Socio-ecological model to explain group-living (adapted from Kappeler 2009, p. 493 Fig. 11.2). The model shows that the distribution of females is influenced by the net benefits of group living. In addition, the distribution of resources together with the predation risk and the risk of infanticide determines the distribution of the females. The males then map their distribution according to that of the females with the potential added benefit of parental care. 2

Chapter 2

Figure 1. Socio-Ecological Model of Solitary Living. Seven main factors that influence the social organisation of a species. + indicates which condition favours solitary living, - which favours group-living. Combined, these factors influence whether solitary or group-living leads to a net benefit for females. In mammals, social organisation is mainly determined by how females distribute themselves, while males follow female social organisation, being solitary living when females are solitary, joining groups when females form groups. 22

Chapter 3

Figure 1. A Bush Karoo rat carrying a mini-GPS datalogger. This female is marked with blond hair dye on the head and back, for individual recognition during behavioural observations..... 35
Figure 2. Track of a handheld extrex Venture GPS used during radio-tracking along a 11km hike. In addition, fixes taken every 5 minutes of 3 Gipsy 5 units (Gipsy units 24, 25 and 27) and one Gipsy 6 unit (Gipsy 35) are shown. At the start of the hike (left, blue arrow), fixes from Gipsy 25 were off the track, and during the middle of the hike (right, green arrow) in a narrow valley, fixes of Gipsy 24 were off the track. Otherwise, most Gipsy fixes were on the handheld GPS track..... 39
Figure 3 Estimated range sizes generated from mini-GPS (red bars) or radio-tracking (blue bars), calculated via different methods: autocorrelated kernel density estimation (AKDE); minimum convex polygons (MCP) 95%; Kernel density estimates (Kernel) 95%. 41
Figure 4. Daily ranges of four individuals (two males and two females) calculated using the MCP method for data obtained from mini-GPS (the large polygons) vs home range estimates from radio-tracking (the small polygons within). The coloured dots represent the different collection methods for the individuals and those outside the polygons represent the outliers excluded by the 95% method. 42

Figure 5. Daily ranges of four females calculated using the *ctmm* package method as autocorrelated Kernel density estimates correcting for the location error vs the stationary test units placed at the nests (the small lines within). This diagram shows that while location errors are relatively large for these small daily ranges, our method allowed to calculate daily ranges that are clearly larger than the location error 45

Chapter 4

Figure 1. Satellite image of the field site (star in insert map). The black polygons are the boundaries of the six trapping areas within the field-site that we demarcated for trapping purposes. In one trapping area we show an example of matrilines in part of the study site for August 2022. Symbols and colours represent lodges that were occupied by bush Karoo rats of three different matrilines (white: rats from other matrilines). In another trapping area the grey lines represent an example of the shortest distance between the occupied lodges calculated in qGIS. 58
Figure 2. Distribution of social units in bush Karoo rats in seven years. The top panel is from September 2017 – December 2020, and the bottom panel is from January 2021 – July 2023. The breaks in the lines are from periods when no data was collected, for example due to the Corona lockdown (March to December 2020). The definitions for the different social units are given in Table 1. The grey boxes indicate the wet, breeding season in winter / spring. The number next to the respective month refers to the year (i.e. Sept-17 = September 2017). 64
Figure 3. The number of female neighbours by kinship (a left; $P < 0.01$) and the mean distance to the nearest neighbour depending on kinship (b right; $P < 0.001$) in bush Karoo rats, for both seasons. points represent individual values (kin: $N = 556$, non-kin: $N = 444$). The black point is the model estimate and the error bars are the confidence intervals of the model. The half-violin plot shows the raw data distribution. 65
Figure 4. Seasonal comparison of daily range size (a left; non-breeding: N = 59, breeding: N = 81) and overlap (right; non-kin: $N = 54$, kin: $N = 72$) in bush Karoo rats. The black point is the model estimate and the error bars are the confidence intervals of the model. The half-violin plot shows the raw data distribution. 66

Chapter 5

Figure 1. The latency to first aggression by focal females to neighbouring cage-housed female bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values. 89

Figure 2. The number of charging events by focal females to neighbouring cage-housed bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values. 89
Figure 3. The time spent at the cage by focal females to neighbouring cage-housed bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values..... 90

Chapter 6

Figure 1. Example of lodges with runways. The hollow diamonds are the shrubs without lodges, black circles are lodges and brown lines are the runways between the lodges and shrubs. 117
Figure 2. Seasonal comparison of total food shared between bush Karoo rats at foraging grounds by kinship. Boxplots show 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values. 119
Figure 3. Mean fitted number of food plants within foraging grounds (solid line) and 95% confidence intervals (shaded area) against range overlap. Black dots are observed data. 120

Chapter 7

Figure 1. Components of social systems. Social systems consist of four components, indicated by the large white circles. Each component can have mutiple categories, for example there are 6 different categories of social organisation. Indicated in grey circles is for each component how many different categories are possible. For the social structure, the components are multiplied by two because they can each either occur or not (e.g., animals can have pair bonds or not, be territorial or not etc.), which provides the minimum number of possibilities. If the four components are seen categorically, then at least 1152 different forms of social systems are possible. In reality, most components are continous, not categorical (for example not dominance hierachy yes or no, but how hierarchical the dominance hierarchy is), leading to even a higher biodiversity of social systems. Apdapted from Kappeler (2019), published in Makuya & Schradin (2024b). 128
Figure 2. Components of social systems in solitary mammals. The social organisation is made up of three aspects which include the group composition (1 male or 1 female), the group size (1 adult), and the spatial structure (kinship structure or not) which can be continuos. The social structure is only made up of one asepect, the social interactions between neighbours which can be amicable or aggressive. The mating

system is made up of three aspects, polygyny, promiscuity or polygynandry. And finally, the care sytem is such that only maternal care is possible (adapted from Kappeler, 2019). 129

List of tables

Chapter 2

Table 1: Glossary of important terms for the categorisation of social systems in mammals used in our review (note that in other taxa, especially social insects, other terms and definitions might be used). Our categorisation is based on (Kappeler, 2019; Kappeler & Schaik, 2002). We include additional terms that are commonly used when writing about social systems, but that are not well defined and which might therefore lead to confusion, stating how we use these terms in the current review or why we try to avoid them. 11

Table 2. Solitary living in different mammalian taxa based on data from field studies. Included are all taxa for which detailed reviews are available (see "Reference"), but unpublished data indicates that similar patterns are found in all other mammalian orders (Olivier, 2023). Given are total number of species and in brackets the % of solitary species relative to all species studied. Obligate: all observed social units were solitary living; mainly: >50% of observed units were solitary living; facultative: <50% of observed units were solitary living. *: The published database does not allow to differentiate between mainly and facultatively solitary species. 13

Table 3. Environmental factors influencing the costs and benefits of group- versus solitary living. Left: Commonly stated benefits of group-living and how this induces costs of solitary living by missing these benefits. Right: Costs of group-living inducing benefits of solitary living, when being avoided. Whether a species evolves group- or solitary living depends on the net outcome of all the factors combined. 17

Chapter 3

Table 1 Categorising fixes of 12 bush Karoo rats (F: females, M: males) carrying Gipsy 6 units. Fixes were visualised depending on their horizontal accuracy category from > 100 m to < 10 m. When all fixes were close to each other and in the expected area where the individual had been observed, the accuracy was categorised as "Good". An accuracy where fixes with some potential outliers (fixes more than 15 m away from expected range) occurred that could be removed by using 95% contours was categorised as "Few outliers". Accuracy where many fixes were far away from the expected area (more than 50 m) was categorised as "Many outliers". "NA": no fixes were recorded in this accuracy category. The Gipsy unit of M111 had a broken antenna and was not used anymore. 39

Table 2. Results of the linear mixed models to test how range estimates were influenced by whether estimates were done using 3 different methods: minimum convex polygons (MCP), Kernel density

(Kernel), or autocorrelated Kernel density estimation; and whether data were from radio-tracking or mini-GPS. Factors in bold type indicate significant predictors. 40

Chapter 4

Table 1. Observed forms of social organization in bush Karoo rats. 60

Chapter 5

Table 1: Hypothesis and associated models tested in the dyadic and field intruder tests. 85

Glossary

Care system: Describes who cares for dependent young.

Conspecifics: A member of the same species.

Cooperation: An interaction between individuals that results in net benefits for everyone involved in the interaction.

Kin structure: A component of social organisation describing the genetic relatedness of individuals within the social unit and within the spatial structure.

Mating system: Describes who mates and reproduces with whom.

Philopatry: Remaining within or close to a natal territory.

Social organisation: The size and composition of a social unit. These social units include solitary, pair (male-female pairs), and group.

Social structure: The content, quality, and patterning of social relationships emerging from repeated interactions between individuals belonging to the same social unit.

Social system: The combination of four interrelated components: care system, mating system, social organisation, and social structure.

Socio-ecological model: A conceptual model that combines ecological factors to explain traits of social systems, thereby enabling prediction on relationships between resource distribution, competition and social systems.

Solitary living: When both male and female adults forage and sleep alone only meet for mating and during territorial encounters.

Spatial structure: A component of the social organisation that describes the distribution of social units across space.

CHAPTER 1. General introduction

Understanding the factors shaping animal societies has been a hallmark of behavioural ecology for decades (Clutton-Brock, 2021; Kappeler, 2019). Socio-ecological models (SEM; Figure 1; Wrangham, 1980) were developed to explain sociality via ecological factors such as the distribution of resources and predation pressure, predicting the ecological conditions under which animals form groups (reviewed by Clutton-Brock & Janson, 2012; Kappeler, 2009). Such models consider the net benefits of group-living, depending on ecological factors, to predict whether individuals are distributed solitarily, in pairs or in groups. If the benefits of group-living outweigh the costs, the net benefits favour group-living. It is therefore obvious that solitary living should be an adaptive consequence of the same socio-ecological factors. However, socio-ecological models were traditionally made to explain group-living, with solitary living functioning as the default starting state that was not explained (Clutton-Brock, 2021; Wrangham, 1980). Therefore, there is a need to develop socio-ecological models for solitary living, as I have done in my thesis (Chapter 2). I regard solitary living as a special adaptation to the prevailing environmental conditions. Therefore, I expected the benefits of solitary living to outweigh the costs in species where all or most individuals are solitary living (Chapter 2).

Solitary living: an understudied phenomenon in mammals

The main benefit of solitary living is to avoid the costs of group-living (Makuya & Schradin, 2024). To study solitary living, we must define this concept. Animals are defined as solitary living when both male and female adults forage and sleep alone for more than 50% of the time[1], and meet only for mating (Makuya & Schradin, 2024a). This definition is based on the social organisation, one of four components of the social system that considers the composition of social units consisting of adult individuals that live together and share the same home range, including sleeping sites. The different social units described include solitary living, pair living (male-female pairs), and different forms of group-living (Kappeler & Schaik, 2002). Groups can comprise of a single male with multiple females, single female with multiple males, and multi-male and multi-female groups (Kappeler & Schaik, 2002). The remaining three components of the social system are the social structure (describing social interactions), the mating system (who mates with whom) and the care system (who takes care of the offspring; Kappeler, 2019; Kappeler & Schaik, 2002). In solitary mammals, the mating system is often polygynandry, while the care system is almost always that only mothers take care of the young. The social structure of solitary species has typically been assumed to be characterised by aggression and very few interactions. This view is changing (Makuya & Schradin, 2024b; Twining et al., 2024).

[1]Most solitary living species spend >95% of their time alone, which is >75% in solitary mammals studied. However, to have a definition that covers all possible (unstudied) species, the criteria to differentiate between solitary and pair / group-living was chosen to be 50%.

Socio-ecological model for group vs solitary living species

Group-living has been explained using socio-ecological models (Clutton-Brock & Janson, 2012; Kappeler & Schaik, 2002), linking ecological factors to social systems and predicting the social organisation and spatial structure of a species (Figure 1; Clutton-Brock & Janson, 2012; Kappeler, 2009; Kappeler & Schaik, 2002). In addition to these factors, the distribution of females is impacted by the risk of infanticide as well as predation risk. By understanding how all these factors affect female distribution, we can also understand how males are distributed because males tend to follow the decisions made by the females and their patterns of receptivity (Clutton-Brock, 1989; Emlen & Oring, 1977): When females form groups, males stay with them and attempt to monopolise reproductive opportunities, which is possible only for small groups, for example in gorillas. Large groups of females typically contain several adult males, for example in chimpanzees. If females are solitary, males will also be solitary, roaming over large areas to visit many females for mating, for example in leopards (*Panthra pardus*) (Clutton-Brock, 1989; Emlen & Oring, 1977).

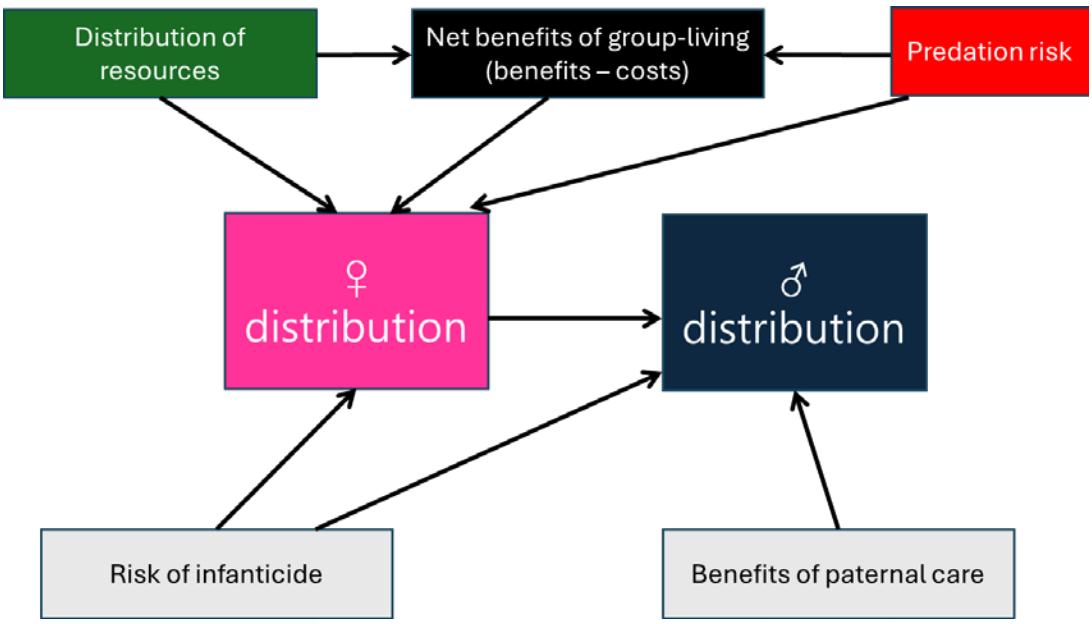

Figure 1. Socio-ecological model to explain group-living (adapted from Kappeler 2009, p. 493 Fig. 11.2). The model shows that the distribution of females is influenced by the net benefits of group living. In addition, the distribution of resources together with the predation risk and the risk of infanticide determines the distribution of the females. The males then map their distribution according to that of the females with the potential added benefit of parental care.

Spatial structure

Spatial structure indicates how individuals are distributed spatially with respect to each other, is characterized by who are neighbours and is an important aspect of a species´ social organisation (Boellstorff & Owings, 1995). One of the factors that determines the spatial structure is resource availability (Maher & Lott, 2000). In addition, the spatial structure can be characterized by kinship, whereby kin live closer to each other to exploit the same resources (Johnson et al., 2002) as in the Eurasian lynx *Lynx lynx* (Aronsson et al., 2020). Recent attempts have been made to apply the socio-ecological model to solitary species. For example, in mouse lemurs (*Microcebus sp.*), spatial ranging and sleeping associations of females differ between two species, the group-living *M. murinus* and the solitary living *M. berthae* (Dammhahn & Kappeler, 2009). *Microcebus murinus* relies on food resources distributed in large patches, thereby resulting in females occupying overlapping home ranges and forming sleeping groups. In contrast, small, dispersed food patches result in within group scramble competition in *M. berthae* and low associations between females (Dammhahn & Kappeler, 2009). The authors of the study highlighted how solitary living can be explained as an adaptation to the same ecological factors used to explain group-living.

Social structure in solitary living species

The social structure describes social interactions between individuals of the same species and has traditionally been used to describe social interactions within one pair or a group (Kappeler, 2019) but can also be used to describe interactions between neighbours in solitary species (Clutton-Brock, 2021). Do solitary living individuals interact with each other? Although the obvious answer to this question is yes, the quality of such interactions remains poorly studied. However, solitary species need to interact with conspecifics at some point in their lives, either for mating or at territory boundaries. To reach a better understanding of the social life of solitary species, we need to understand their social structure, how and when they interact with conspecifics.

Individuals of solitary species interact with conspecifics during territorial encounters, mating, during juvenile dispersal, and, in the case of female mammals, offspring care (Clutton-Brock, 2021). For example, amicable social interactions between neighbours occurs in solitary species such as pumas (*Puma concolor*; Elbroch & Quigley, 2016), Columbian ground squirrels (*Urocitellus columbianus*; Viblanc et al., 2016b), Brants´ whistling rats (*Parotomys brantsii*; Jackson, 1999), and nocturnal prosimians (Agnani et al., 2018; Müller & Thalmann, 2000). These studies showed that solitary mammals can have non-random social interactions that are not aggressive, and that the quality of such social interactions probably impacts individual fitness. Solitary living might thus only seem simple, with almost no social interactions, but can

in fact involve complex social structures. Solitary living in general is the most understudied of the social systems, with pair- and group-living instead having received the most attention (Clutton-Brock, 2021).

Costs and benefits of solitary living

Solitary living has been regarded as the ancestral form of social organisation in mammals (Lukas & Clutton-Brock, 2013b) which might explain why so little attention has been paid to understanding its costs and benefits. However, I predict that solitary animals benefit from their type of social organisation, despite missing out on the benefits of group-living. To understand the evolution of social organisation, it is important to understand the costs and benefits of all different forms of social organisation, including solitary living. Further, many mammal species are solitary living (Lukas & Clutton-Brock, 2013b; Makuya & Schradin, 2024a), so understanding how they respond to change is essential to assess the consequences of global change on mammalian biodiversity (Olivier et al 2022): how will environmental change influence to net benefit of solitary living?

PROBLEM IDENTIFICATION

Given the diversity of social systems in mammals, it is not a trivial question to ask why solitary living is so successful in many species. So far, it had been assumed that most mammals are solitary, and that solitary living is the primitive ancestral state needing no explanation for its evolution (Lukas & Clutton-Brock, 2013b). However, in my thesis, I will show that in recent comparative studies only 22% of mammals were recognised as solitary living and that solitary living is neither primitive nor ancestral (Chapter 2; Olivier et al., 2024). To better understand solitary living, I studied the social system of a small rodent, the bush Karoo rat (*Otomys unisulcatus*), focussing on its social organisation, spatial structure and social structure. I investigated how these social components depend on kinship, and whether there could be some simple form of cooperation in this solitary mammal. This information on the social system of the bush Karoo rat will help to better understand the costs and benefits of solitary living. My overarching question was what strategies do bush Karoo rats use for a successful solitary life. I expected to show that the bush Karoo rat is a good model to study the evolution of solitary living by focusing on its social organisation, spatial structure and social structure and finally review the costs and benefits of solitary living

STUDY SPECIES

Many solitary mammals, such as shrews, rodents and large predators are challenging to study because they are either small and nocturnal and occupy dense habitats, or are large and nocturnal and occupy large areas.

I studied the bush Karoo rat because it is an ideal model to study the costs and benefits of solitary living. It is diurnal, occupies an open habitat characterised by low growing annuals and dispersed shrubs, it is a central place forager that always returns to its main nest with food, and easily habituates to the presence of human observers. It weighs about 100g and a large sample of 20-50 social units can be studied simultaneously on a field site of 5ha. There are no peer-reviewed studies on the social system of bush Karoo rats. Unpublished student reports and PhD theses of populations in other parts of the distribution range of the species suggested that more than one adult rat can occupy a nest (Babu et al., 2011; Do Linh San et al., 2016; Hoepfl, 2013; Xalu et al., 2009). However, preliminary observations from our field site indicate that only one adult bush Karoo rat occupies a nest at a time (Thomson, 2018).

The bush Karoo rat builds stick-lodges inside shrubs and most published studies in this species focus on these lodges (Brown & Willan, 1991a; Jackson et al., 2004; Jackson et al., 2002; Vermeulen & Nel, 1988). The lodges offer a favourable microclimate with high humidity and mild temperatures and thus offer protection against the harsh outside environment, characterised by unpredictable winter rainfall coupled with a long dry season, to which the bush Karoo rat is physiologically poorly adapted (du Plessis & Kerley, 1991; du Plessis et al., 1992). These stick lodges start off small (20cm in height) at first but can reach 1.5 meters high and three metres wide over the years due to cumulative additions to continual work on them by multiple generations of bush Karoo rats (Schradin, 2005). Each occupant maintains the lodge during its tenure such that multiple generations maintain the lodges over time, resulting in a cumulative growth of the lodges (personal observations). These stick-lodges are thus expensive to build and represent a valuable resource.

AIM, OBJECTIVES AND PREDICTIONS

Aims

The main aim of my study was to reach a better understanding of the costs and benefits of solitary living. I aimed to archive this by studying the social organisation, spatial structure and social structure of a solitary living rodent, the bush Karoo rat. Furthermore, I investigated whether cooperation existed between female kin, compared to interactions with non-kin neighbours. To be able to address these aims, I validated the method of using mini-GPS data loggers to assess space use in this small rodent species.

Objectives and predictions

I had four objectives.

1. *Review evidence about costs and benefits of solitary living in mammals (Chapter 2)*

To reach a better understanding of what is known about solitary living in mammals, I reviewed the literature to assess how common solitary living is in mammals, when in phylogenetic history it might have evolved, what we know about the social structure of solitary mammals, and what empirical evidence exists about the costs and benefits of solitary living. I also developed a socio-ecological model for solitary living.

2. *Investigate the social organisation in bush Karoo rats (Chapters 3 and 4)*

I investigated the social organisation of bush Karoo rats using trapping and observation data. I also assessed whether the spatial organisation represents kinship, i.e. whether close kin live close to each other. For this, first I validated the use of mini-GPS dataloggers (Chapter 3) and then studied the daily range sizes and overlap of daily range sizes to test three predictions regarding social organisation in (Chapter 4).

- The bush Karoo rat has a solitary social organisation.
- Daily ranges will be larger during the breeding season, when females search for food than during the dry non-breeding season.
- The spatial organisation will comprise of a female kin structure, meaning that 1) female neighbours were close kin, and 2) daily ranges of female kin neighbours overlapped more than that of non-kin.

3. *Investigate the social structure of bush Karoo rats (Chapter 5)*

Social interactions, including close proximity, amicability and aggression, were recorded during behavioural observations. I experimentally measured aggression / amicability in dyad encounters in a neutral presentation arena and in field intruder tests. I compared the reactions towards kin versus non-kin neighbours. Using all three approaches, I investigated how mother-infant relationships changed over time. Based on the above objectives, two predictions were tested.

- More amicable behaviour, closer proximity and less aggression will occur between kin neighbours compared to non-kin neighbours.
- Mother-infant relationships will change over time with a decrease in amicable behaviour and being in close proximity, and an increase in aggression as juvenile offspring become adult, explaining solitary living.

4. *Test for evidence of cooperation between closely related neighbours (Chapter 6)*

Here, I combined results about the spatial structure (part of social organisation) and social structure (social interactions) and related these to food availability using linear and generalized models. To assess the quality

of home ranges, I conducted runway surveys where I counted food plants along the runways. I further calculated the proximity of bush rats to their neighbours' using data from mini-GPS dataloggers that were carried by the neighbouring rats simultaneously. Based on the above objectives, two predictions were tested.

- Related females will share more foraging grounds (as measured by runway surveys) without aggressive interactions than unrelated females.
- Related females will have higher temporal overlap in their ranges than non-related females.

THESIS OUTLINE

This thesis comprises of 7 chapters, the general introduction (chapter 1), a review on the costs and benefits of solitary living (chapter 2), four empirical chapters (3-6), and a general discussion (chapter 7). Chapter 2 is published in the *Journal of Zoology*, Chapter 3 in *Mammalian Biology*, Chapter 4 in *Animal Behaviour*, Chapter 5 and 6 are submitted and currently under review. For the review chapter and the empirical chapters, I was responsible for the main investigation, data collection, statistical analyses, writing of the original draft and final edits. Additional publications that I authored or co-authored during my thesis are listed in the Appendix and include a commentary on the importance of studying solitary species published in *Proceedings of the National Academy of Science (PNAS)* (Makuya & Schradin, 2024b).

A separate reference list is available at the end of each chapter, and thus there is overlap in referencing between chapters. All tables and figures are numbered sequentially per chapter, while page numbers are numbered throughout the thesis.

REFERENCES

Agnani, P., Kauffmann, C., Hayes, L. D., & Schradin, C. (2018). Intra-specific variation in social organization of Strepsirrhines. *American Journal of Primatology, 80*(5), e22758. doi: doi:10.1002/ajp.22758

Aronsson, M., Åkesson, M., Low, M., Persson, J., & Andrén, H. (2020). Resource dispersion and relatedness interact to explain space use in a solitary predator. *Oikos, 129*(8), 1174-1184.

Babu, N., Xalu, M., & San, E. D. L. (2011). Socio-spatial organization of bush Karoo rats (Otomys unisulcatus) in the Albany Thicket Biome: preliminary results.

Boellstorff, D. E., & Owings, D. H. (1995). Home range, population structure, and spatial organization of California ground squirrels. *Journal of Mammalogy, 76*(2), 551-561.

Brown, E., & Willan, K. (1991a). Microhabitat selection and use by the bush Karoo rat *Otomys unisulcatus* in the Eastern Cape Province. *S Afr J Wildl Res, 21*, 69-75.

Clutton-Brock, T. (2021). Social evolution in mammals. *Science, 373*(6561), eabc9699.

Clutton-Brock, T. H. (1989). Mammalian mating systems. *Proceedings of the Royal Society B: Biological Sciences, 236*, 339-372.

Clutton-Brock, T., & Janson, C. (2012). Primate socioecology at the crossroads: past, present, and future. *Evolutionary Anthropology: Issues, News, and Reviews, 21*(4), 136-150.

Dammhahn, M., & Kappeler, P. M. (2009). Females go where the food is: does the socio-ecological model explain variation in social organisation of solitary foragers? *Behavioral Ecology and Sociobiology, 63*, 939–952. doi: 10.1007/s00265-009-0737-2

Do Linh San, E., Babu, N., Xalu, M., Le Gars, S., Perquin, J., & Baxter, R. (2016). A conservation assessment of Otomys unisulcatus. *The red list of mammals of South Africa, Swaziland and Lesotho*.

du Plessis, A., & Kerley, G. I. H. (1991). Refuge strategies and habitat segregation in two sympatric rodents *Otomys unisulcatus* and *Parotomys brantsii*. *Journal of Zoology, London, 224*, 1-10.

du Plessis, A., Kerley, G. I. H., & Winter, P. E. D. (1992). Refuge microclimates of rodents: a surface nesting *Otomys unisulcatus* and a burrowing *Parotomys brantsii*. *Acta Theriologica, 37*, 351-358.

Elbroch, L. M., & Quigley, H. (2016). Social interactions in a solitary carnivore. *Current Zoology, 63*, zow080. doi: 10.1093/cz/zow080

Emlen, S. T., & Oring, L. W. (1977). Ecology, secual selection, and the evolution of mating systems. *Science, 197*, 215-223.

Hoepfl, J. (2013). Aspects of the biology and behaviour of the Bush Karoo Rat (Otomys unisulcatus) in the Central Karoo, South Africa.

Jackson, T. P. (1999). The social organisation and breeding system of Brants´ whistling rat (*Parotomys brantsii*). *Journal of Zoology (London), 247*, 323-331.

Jackson, T. P., Bennett, N. C., & Spinks, A. C. (2004). Is the distribution of the arid-occuring otomyine rodents of southern Africa related to physiological adaptation or refuge type? *J Zool Lond, 264*, 1-10.

Jackson, T. P., Roper, T. J., Conradt, L., Jackson, M. J., & Bennett, N. C. (2002). Alternative refuge strategies and their relation to thermophysiology in two sympatric rodents, *Parotomys brantsii* and *Otomys unisulcatus*. *J Arid Environ, 51*, 21-34.

Kappeler, P. (2009). Verhaltensbiologie. *Verhaltensbiologie:*.

Kappeler, P. M. (2019). A framework for studying social complexity. [journal article]. *Behavioral Ecology and Sociobiology, 73*(1), 13. doi: 10.1007/s00265-018-2601-8

Kappeler, P. M., & Schaik, C. P. v. (2002). Evolution of primate social systems. *International Journal of Primatology, 23*, 707-740.

Lukas, D., & Clutton-Brock, T. H. (2013b). The evolution of social monogamy in mammals. *Science, 341*(6145), 526-530. doi: 10.1126/science.1238677

Maher, C. R., & Lott, D. F. (2000). A review of ecological determinants of territoriality within vertebrate species. *The American Midland Naturalist, 143*(1), 1-29.

Makuya, L., & Schradin, C. (2024a). Costs and benefits of solitary living in mammals. *Journal of Zoology, 323*, 9-18. doi: https://doi.org/10.1111/jzo.13145

Makuya, L., & Schradin, C. (2024b). The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences, 121*(13). doi: https://doi.org/10.1073/pnas.2402871121

Müller, A. E., & Thalmann, U. (2000). Origin and evolution of primate social organisation: a reconstruction. *Biological Reviews, 77*, 405-435.

Olivier, C.-A., Martin, J. S., Pilisi, C., Agnani, P., Kauffmann, C., Hayes, L., Jaeggi, A. V., & Schradin, C. (2024). Primate social organization evolved from a flexible pair-living ancestor. *Proceedings of the National Academy of Sciences, 121*(1), e2215401120.

Schradin, C. (2005). Nest-Site Competition in Two Diurnal Rodents from the Succulent Karoo of South Africa. *Journal of Mammalogy, 86*(4), 757-762. doi: 10.1644/1545-1542(2005)086[0757:Ncitdr]2.0.Co;2

Thomson, J. (2018). *Describing the social organisation of the bush Karoo rat Otomys unisulcatus.* BSc (Hons) Zoology with Industrial Experience, The University of Manchester.

Twining, J. P., Sutherland, C., Zalewski, A., Cove, M. V., Birks, J., Wearn, O. R., Haysom, J., Wereszczuk, A., Manzo, E., & Bartolommei, P. (2024). Using global remote camera data of a solitary species complex to evaluate the drivers of group formation. *Proceedings of the National Academy of Sciences, 121*(12), e2312252121.

Vermeulen, H., & Nel, J. (1988). The bush Karoo rat *Otomys unisulcatus* on the Cape West coast. *African Zoology, 23*(2), 103-111.

Viblanc, V. A., Saraux, C., Murie, J. O., & Dobson, F. S. (2016b). Kin effects on energy allocation in group-living ground squirrels. *Journal of Animal Ecology*, n/a-n/a. doi: 10.1111/1365-2656.12541

Wrangham, R. W. (1980). An ecological model of female-bonded primate groups. *Behaviour*, 262-300.

Xalu, M., Babu, N., & San, E. D. L. (2009). Activity patterns of bush Karoo rats (Otomys unisulcatus) in the Thicket Biome of South Africa: preliminary results.

CHAPTER 2: Costs and benefits of solitary living

Makuya, L[1]., & Schradin, C[1,2]. (2024). Costs and benefits of solitary living in mammals. *Journal of Zoology*, *323*, 9-18. doi: https://doi.org/10.1111/jzo.13145.

Authors' affiliations:

[1] School of Animal, Plant & Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS 2050, Johannesburg, South Africa

[2] IPHC, UNISTRA, CNRS, 23 rue du Loess, 67200 Strasbourg, France

Abstract

While for decades behavioural ecologists have studied the costs and benefits of group-living, solitary living has received little attention. Instead, it was assumed to be the default stage from which sociality evolved. Mammals underwent around 200 million years of social evolution, with a few species evolving communal or cooperative breeding in harsh environments. Other mammal species are successful with solitary living in exactly the same and many other environments, indicating that solitary living is beneficial under many environmental conditions. Comparative studies on mammals indicate that solitary living might not be the ancestral but a derived state. Solitary living in mammals is less common than previously believed, occurring in 22% of the studied species. Here we review costs and benefits of solitary living in mammals. We found very few studies that considered solitary living and show important future avenues of research based on the factors that are important for the evolution of group-living. We also emphasize that a solitary form of social organisation does not imply an unsocial lifestyle: Solitary mammals typically have non-random but individualised social interactions with their neighbours, indicating important social structure.

Keywords: solitary living; group-living; social system; socio-ecology; thermoregulation; mammal

Introduction

Mammals show a fascinating diversity of social systems, including solitary and pair-living species, extended families and cooperative or communal breeding (Burda et al., 2000; Clutton-Brock, 1989).

Understanding the evolution of this diversity of mammalian social systems received significant attention (Kleiman & Malcolm, 1981; Lukas & Clutton-Brock, 2013b; Thierry et al., 2000). Comparative studies have made the arguments that the first placental mammal was solitary and that the more complex forms of social systems such as monogamy and cooperative breeding evolved later (Gebo, 2004; Lukas & Clutton-Brock, 2013b). Here we question the assumption that solitary living in mammals is primitive. Instead, we argue that solitary living in mammals is a specialized form of social organization which we need to study if we want to comprehensively understand mammalian social evolution and how to protect solitary living mammals.

What is solitary living?

We follow the concept of Kappeler & Schaik (2002) (updated by Kappeler (2019)) which identifies four components for each social system including the (i) social organisation, (ii) social structure (iii) mating system, and (iv) the care system (see Table 1 for definitions). It is now widely accepted that we have to clearly differentiate between these components, as for example not all pair-living species mate monogamously, display pair-bonds and biparental care (Fernandez-Duque et al., 2020; Huck et al., 2020; Kvarnemo, 2018). Previous studies defined solitary living based on a mixture of the mating system (only one breeding female) and care-system (maternal care only) (Lukas & Clutton-Brock, 2013b), which lead to the problem that several species having social units consisting of several adults were categorized to be solitary rather than group-living (Schradin, 2017).

We define solitary living based on the social organisation of social units (see Table 1 for definitions): both adult males and adult females sleep and forage alone for more than 50% of the time and mainly meet for courtship and mating. While for our definition the criteria of 50% applies (allowing all species on the continuum from 0 to 100% to be categorised), from our experience social units typically are either >75% of the time with the same conspecifics (then called group living) or >75% of the time alone (then called solitary living). Social units that forage alone but consistently (>50% of the time) share the same nesting site and territory with the same conspecific(s) are not regarded as solitary living, but as pair- or group-living solitary foragers. A social unit (a solitary individual, a pair, or a group) can have only one form of social organisation, while species consisting of many social units can have several forms of social organisation, leading to intra-specific variation in social organisation.

Table 1: Glossary of important terms for the categorisation of social systems in mammals used in our review (note that in other taxa, especially social insects, other terms and definitions might be used). Our categorisation is based on (Kappeler, 2019; Kappeler & Schaik, 2002). We include additional terms that are commonly used when writing about social systems, but that are not well defined and which might therefore lead to confusion, stating how we use these terms in the current review or why we try to avoid them.

Term	Definition	Examples / comments
Terms from the classification by (Kappeler, 2019; Kappeler & Schaik, 2002)		
Care system	Describes who shows infant care.	Parental and allo-parental care; helpers at the nest.
Mating system	Describes who mates with whom and who reproduces.	Monogamy; singular or plural breeding; communal breeding.
Social organisation	Composition and size of a social unit.	Solitary; pair living; group living.
Social structure	Social interactions including communication.	Social bonding; dominance hierarchy. Includes all social behaviours, amicable and aggressive ones.
Social system	The combination of the four components care system, mating system, social organisation and social structure.	A biparental monogamous pair living species with pair bonds. A maternal, polygynous, solitary species where female kin tolerate each other but are aggressive towards non-kin.
Social unit	Individuals that live together and that share the same home range including sleeping sites.	A solitary male leopard. A pair of nocturnal prosimians that share the same home range and sleeping site. A group of baboons.
Additional terms		
Colony	Multiple social units (social organisation) that live so close to each other that communication and visual contact between social units is possible (social structure). Interactions between social units are typically aggressive, within social units amicable.	Colonies of bats where (in some species) the social units forming a colony can consist of one breeding male and multiple females with offspring. In whistling rats (*Parotomys spec.*), social units are solitary females that defend their burrow and territory against other solitary females in the colony.
Social behaviour	Any behaviour shown in interactions between conspecifics. Includes amicable and aggressive behaviours (social structure) and behaviours of the care system.	Grooming, fighting, warming offspring.
Social	This term is not well defined. It is typically used to classify individuals / species with amicable interactions (social structure), but also those living in groups (social organisation) or showing parental care (care system).	This term should be avoided and replaced by specific terms from the four components of social systems. Its not useful to rank species as “very” social, “highly” social, when no measure for comparison is provided.
Unsocial	This term is not well defined. It is typically used to classify individuals / species where nearly all interactions (social structure) between conspecifics are aggressive and individuals live solitarily (social organisation).	This term should be avoided and replaced by specific terms from the four components of social systems.

A species is said to be mainly solitary when most males and females follow this definition. This excludes dispersers, as most mammal species have a solitary dispersal phase, often biased but not restricted towards males (Lawson Handley & Perrin, 2007b). Including dispersers into the classification of a species` social organisation would inflate the number of species being partially solitary living to nearly 100% and make most species showing intra-specific variation in social organisation. While such a classification would be useless, understanding costs and benefits of solitary living will help us to better understand the life history of the many mammalian species that have a solitary disperser stage.

Many species show intra-specific variation in social organisation (Schradin et al., 2018a) and this includes species where most individuals live solitarily, though some individuals form pairs or even groups (Makuya et al., 2022; Valomy et al., 2015b). Therefore, one can differentiate between species that are obligately solitary living (all individuals are solitary), mainly solitary living (most individuals are solitary living) and facultatively solitary living (most individuals live in pairs or groups and less than 50% solitary). Considering such variation is important in comparative studies about social evolution and can challenge previous studies that ignored variation (Dalerum, 2007; Griesser & Suzuki, 2016; Olivier et al., 2024; Qiu et al., 2022).

Table 2. Solitary living in different mammalian taxa based on data from field studies. Included are all taxa for which detailed reviews are available (see "Reference"), but unpublished data indicates that similar patterns are found in all other mammalian orders (Olivier, 2023). Given are total number of species and in brackets the % of solitary species relative to all species studied. Obligate: all observed social units were solitary living; mainly: >50% of observed units were solitary living; facultative: <50% of observed units were solitary living. *: The published database does not allow to differentiate between mainly and facultatively solitary species.

Taxa	Number of species (IUCN)	Number species with field data	Species obligate solitary living	Species mainly solitary living	Species facultatively solitary living	Reference
Marsupials	345	65	20 (31%)	6 (9%)	8 (12%)	(Qiu et al., 2022)
Xenarthra (slots and ant eaters)	30	9	7 (78%)	1 (11%)	1 (11%)	(Makuya et al., 2022)
Macroscelidae (elephant shrews)	19	8	1 (13%)	0	5 (63%)	(Olivier et al., 2022a)
Eulipotyphla (insectivores)	445	16	7 (44%)	0	5 (31%)	(Valomy et al., 2015b)
Artiodactyla (even-toed ungulates)	226	100	5 (5%)	0*	55* (55%)	(Jaeggi et al., 2020)
Carnivora (carnivores)	271	171	71 (42%)	0*	<46 (27%)	(Dalerum, 2007)
Strepsirrhini (prosimians)	132	43	3 (7%)	5 (12%)	7 (16%)	(Agnani et al., 2018)

Haplorrhini (apes and monkeys)	450	180	0 (0%)	5 (3%)	0 (0%)	(Olivier et al., 2024)

Solitary living in mammals

It is often believed that most mammals are solitary living. For example, Lukas & Clutton-Brock (2013b) classified 1741 out of 2545 (68%) of mammalian species as solitary living. This included several hundreds of mammal species that have not been studied in the wild but that were assumed to be solitary living due to their nocturnal and cryptic lifestyle (Agnani et al., 2018; Schradin, 2017). Table 2 summarises reviewed empirical data from field studies on mammals from a broad range of taxa. It becomes evident that solitary living is rather rare in mammals, being the obligate or main form of social organisation in 22% (131 of 592) of the studied species (Table 2).

Phylogenetic heritage of solitary living? Evidence from palaeontology

Cynodonts living 250 million years ago gave rise to mammaliaforms, the ancestors of true mammals. Fossil evidence of parental care in cynodonts was reported already more than 60 years ago (Brink, 1956) and is available for two cynodont genera (Jasinoski & Abdala, 2017). Cynodonts were burrowing animals (Groenewald et al., 2001) and associations of both juveniles with adults as well as of several adults found in fossilized burrows are indicative of group-living (Jasinoski & Abdala, 2017). The absence of sexual size dimorphism in the cynodont *Thrinaxodon* and the small number of adults found together (Jasinoski & Abdala, 2017) indicates that this species might have been pair-living.

Some cynodonts were already pair or group-living, and the same has been found for some early mammals. For example *Filikomys primaevus*, which lived 75 million years ago and belonged to the then dominant mammalian order of Multituberculata, was group living (Weaver et al., 2020). Fossils found at a dinosaur nesting site in the USA indicate that this burrowing species lived in groups of multiple generations that shared one burrow system and nesting site. The stem-metatherian *Pucadelphys andinus* that lived in Bolivia 63 million years ago is a relative of extant marsupials (Ladeveze et al., 2011). Fossils of multiple adults of both sexes were found together with sub-adults and juveniles, indicating a group-living social organisation (Ladeveze et al., 2011). The only major mammalian taxa for which no fossil evidence for group-living exists are monotremes. Monotremes are regarded as the most primitive living mammals and typically regarded to be solitary (Nowak & Wilson, 1999). However, there have been numerous reports of den sharing in platypus (Makuya et al., 2022). In sum, fossil evidence indicates that ancestors of mammals and some early mammals were social, questioning a solitary ancestry for mammals.

Evidence from recent comparative studies

Multiple comparative studies on different mammalian taxa aimed at estimating the social organisation of the common ancestor. In Xenarthra, most species were described to be solitary living, but too few studies were available for a comparative analysis (Makuya et al. 2022). In Eulipotyphla it was suggested that the ancestor might have been pair-living (Valomy et al., 2015b). Comparative studies using modern Bayesian statistics and databases based on field studies found pair-living to be the most likely ancestral form of social organisation in marsupials (Qiu et al., 2022), artiodactyla (Jaeggi et al., 2020), primates (Olivier et al., 2024), and even all mammals (Olivier, 2023).

Solitary living as a special adaptation

Both evidence from paleontology as well as from comparative studies indicates that solitary living is not ancestral in mammals. If solitary living is not ancestral, then it must have evolved as a specific adaptation, and this probably repeatedly in several lineages as indicated by the comparative studies on different mammalian taxa.

Solitary mammals are the product of millions of years of evolution. Today, they live in very diverse habitats, and many are exposed to environmental variation and harshness (Makuya et al., 2023; Schradin et al., 2023), factors associated with the evolution of a few complex mammalian societies (Lukas & Clutton-Brock, 2017). For example, in the harsh Kalahari, only 2 out of 90 mammal species listed by Anderson (1998) are cooperative breeders (meerkats and Damaraland mole-rats), while 30 species are solitary living. Solitary living should be considered as a specialized adaptation to specific environments, in the same way as has been done for cooperative breeding. The problem is that so far solitary living has not been studied as an evolved trait.

There is some evidence that solitary living evolved as an adaptation to specific environments. Extant monotremes are highly specialized invertebrate feeders, and their solitary social organization might be an adaptation to this non-sharable and sparsely distributed food resource (Nowak & Wilson, 1999). Aardvarks (Taylor & Skinner, 2003), ant bears and armadillos are other specialized insect feeders that are solitary (Makuya et al., 2022). Being insectivorous means feeding competition doesn't allow large groups and might thus favour pair- or solitary living, which is common in Eulipotyphla (Valomy et al., 2015b). For some insectivorous species, being pair- or group-living solitary foragers can be a solution, for example for insectivorous bats (Kerth, 2022). Large predators such a many bears and most large cats are solitary, maybe

as an adaptation to the distribution and size of their prey (Nowak & Wilson, 1999). Thus, wide distribution of food sources might favour solitary living. Solitary living also occurs in folivorous sloths (Makuya et al., 2022) which are well camouflaged as a protection against predation, an advantage that would disappear if groups of sloths would occupy the same tree.

Solitary species need not be asocical

Solitary living in mammals is assumed to develop due to offspring dispersing when reaching adulthood either because they themselves develop the urge to leave, or because the mother rejects them. Maternal aggression towards offspring reaching puberty has been observed in European hamsters (*Cricetus cricetus;* (Eibl-Eibesfeldt, 1953) and in some, but not all species of elephant shrews (Rathbun, 1979 Schubert et al., 2012). More studies are needed to know how common maternal aggression is in inducing dispersal, or whether its internal motivation of the offspring when / after reaching puberty driving them to disperse and become solitary living.

Solitary species don't have to be asocial, actively repelling each other. Instead, species with a solitary social organization often have a complex social structure in which individuals do interact in a non-random way. For example the puma (*Puma concolor*) displays a complex social structure characterized by hierarchical reciprocal tolerance of adult females centred around dominant adult males (Elbroch et al., 2017b). Similarly, female leopards (*Panthera pardus*) share space according to kinship (Roex et al., 2022). Complex social structures influencing fitness also occur in nocturnal solitary prosimians (Agnani et al., 2018; Müller & Thalmann, 2000) as well as in solitary living but colony forming rodents such as Columbian ground squirrels (*Urocitellus columbianus*, (Viblanc et al., 2016a)) and Brants´ whistling rat (*Parotomys brantsii*, (Jackson, 1999)). The spatial distributions of solitary species can affect social interactions and fitness (Siracusa et al., 2021). Solitary species can gain fitness benefits by living near familiar neighbours (Walmsley et al., 2023). In sum, social interactions often play an important role in solitary species.

Costs and benefits of solitary living

While for decades studying the benefits of group-living has been a main focus of behavioural ecology, little is known about the benefits of solitary living. To understand why a species is group- or solitary living, we must know the costs and benefits. Many studies found good evidence that group-living provides benefits, but we still lack good measures of the costs of group-living (Clutton-Brock, 2021; Krause & Ruxton, 2002). Benefits of solitary living arise when costs of group-living can be avoided (Table 3). Thus, benefits of

solitary living are the costs of group-living and missing benefits of group-living represents costs of solitary living (Table 3). Whether a species benefits from solitary or from group-living living depends on the net cost-benefit calculation of all factors summarised in Table 3, and it is the environment that influences the outcome for each factor. For example, while for a species under its environmental condition one factor such as foraging might favour group-living, high costs of group-living due to increased predation risk might nevertheless lead to the evolution of solitary living. Here we discuss how solitary living animals might compensate or avoid these costs. Understanding benefits of solitary living will not only allow us to understand why so many species have this form of social organisation but will also lead to a better understanding of the evolution of sociality.

Table 3. Environmental factors influencing the costs and benefits of group- versus solitary living. Left: Commonly stated benefits of group-living and how this induces costs of solitary living by missing these benefits. Right: Costs of group-living inducing benefits of solitary living, when being avoided. Whether a species evolves group- or solitary living depends on the net outcome of all the factors combined.

Factor	**Benefits of group living**	**Costs of solitary living**	**Costs of group-living**	**Benefits of solitary living**
Parasites	Removal of ectoparasites by grooming.	Reduced ectoparasites removal rate.	Increased infection risk.	Decreased infection risk.
Disease	None (possibly taking care of sick in humans and a few other species).	None.	Increased infection risk.	Decreased infection risk.
Predation risk	Reduced predation due to (i) dilution effect, (ii) confusion effect, (iii) overall increased vigilance but decreased individual vigilance, (iv) group defence against predators.	Overall increased risk to be predated if detected by predator. Increased individual vigilance.	Increased risk of being detected by predator as groups are more conspicuous than single individuals.	Reduced risk to be detected.
Foraging and competition for food	Finding good food-sites. Catching prey. Shared foraging grounds.	No information transfer regarding good food sites. Decreased success in catching prey. Smaller foraging grounds.	Competition for food, reduced food availability.	Monopolised access to food.
Territoriality	Large territory due to group defence.	Smaller territory.	Larger territory needed to have sufficient resources.	Smaller territories that are less costly to defend.

Reproduction	Easy access to potential mates.	Increased investment to find mates.	Direct competition for access to mates. Reproductive suppression. High reproductive skew.	All individuals can reproduce. No mate monopolisation. Low reproductive skew.
Infant care	Help by allo-parents.	Infants alone for longer, higher individual costs of infant care.	Costs of showing allo-parental care.	All parental investment goes into own offspring. Males invest time and energy into mate searching instead of paternal care.
Thermoregulation	Reduced costs of physiological thermoregulation due to huddling.	High costs of thermoregulation.	Increased risk of infection with disease and parasites due to body contact between individuals.	-

Parasites and disease

Decreased risk of disease and parasite transmission has been discussed to be a main benefit of solitary living (Krause & Ruxton, 2002). Indeed, a positive correlation between parasite load and colony size has been reported in both birds (Brown & Brown, 1986) and mammals (Lopez et al., 2013), though empirical evidence is mixed, as sociality can also lead to decreased parasite load (Lutermann et al., 2013). Thus, removal of ecto-parasites would be a benefit for group-living (Table 2). Meta-analyses focus on gregarious species living in groups of different sizes, having very little data from solitary species, finding that parasite intensity and prevalence but not parasite species richness increase with group size (Patterson & Ruckstuhl, 2013). Surprisingly few studies have measured parasite load in solitary species. In rodents, no effect of sociality on endoparasite richness was found, but solitary rodents have a higher ectoparasite richness (Bordes et al., 2007). The weak evidence of increased risk of disease with increased sociality could be due to evolved mechanisms to reduce infection risk / parasite load. This includes allo-grooming to remove parasites, sickness behaviour (sick individuals being inactive and thus reducing spread of disease (Hart & Hart, 2019)), and possibly a stronger immune system to cope with increased disease load (Bordes et al., 2007). In sum, while it is a plausible hypothesis that solitary species have a lower disease transmission rate, empirical data supporting this hypothesis are so far missing, maybe because so few solitary species have been studied. Another problem is that research has focussed on ecto-parasites, which can be removed by conspecifics in social species, while few data are available regarding the spread of infectious disease caused by bacteria and viruses.

Predation risk

While living in groups can be of great benefit to avoid predation due to the dilution and confusion effect as well as group defence (Krause & Ruxton, 2002), this is mainly the case for large mammals. For small mammals (and most mammals are small) such as rodents, insectivores and small carnivores, it is less clear whether moving around in a group would bring benefits of predator avoidance, or rather attract predators (Krause & Ruxton, 2002). Little research has been done on this topic in mammals, but from birds it is known that larger flocks can attract more predator attacks (Cresswell, 1994) and that nests in colonies suffer higher predation rate than solitary nests (Andersson & Wiklund, 1978). Small mammals that are central place foragers can benefit from high densities when warning each other with warning calls, whether they are truly social (marmots; (Blumstein et al., 2006)) or whether colonies consist of many solitary individuals (whistling rats; LeRoux et al., 2002). Similarly, solitary Formosan squirrels *Callosciurus erythraeus* have alarm calls to inform the neighbourhood and they mob together against predators (Tamura, 1989; Tamura et al., 1989). Thus, solitary small mammals can benefit from conspecifics by forming colonies, while solitary small mammals not living in colonies reduce predation risk by being less conspicuous than a group.

Foraging and competition for food

Solitary individuals cannot benefit from following knowledgeable conspecifics to good food sources, though it has been often observed that solitary predators, such as cougars (Elbroch et al., 2017b) and Tasmanian devils (Andersen et al., 2020), meet at carcasses, leading to severe competition.

Conspecifics compete for food, inducing costs, especially when food is scarce and patchily distributed. A group will deplete a resource faster than a single individual. Thus, amount and distribution of food sources will determine to what extent it is costly to forage in groups.

Territorial solitary individuals can benefit from monopolising access to food sources. It has been argued that species feeding on dispersed insects as well as those predating on large widely distributed prey (leopards, tigers, bears) are solitary to avoid competition. While plausible, an empirical test of these hypotheses is missing. Many insectivorous Eulipotyphla are more sociable than previously believed (Valomy et al., 2015b), and most canids as well as lions do live in groups (Dalerum, 2007). Comparative studies testing whether food distribution affects social organisation are needed. One empirical study was done by Dammhahn & Kappeler (2009) where they looked at two solitary foraging mouse lemur species that differed in the spatial ranging and sleeping associations of females. *Microcebus berthae* relied on small,

dispersed food patches causing within-group scramble competition that could explain low associations between females. In contrast, *M. murinus* made use of patchily distributed large food resources allowing females to overlap their home ranges and to form sleeping groups.

Territoriality

Territoriality is a way to monopolise access to resources, which can include food, mates, and shelter. The quantity of food available per square meter is one determinant of territory size, and higher food abundance leads to smaller territories (Schoepf et al., 2015; Schradin et al., 2010b). As more individuals need more resources, groups need to defend larger territories and also might have to travel larger distances. One benefit of solitary living is thus that smaller territories need to be defended and that smaller distances need to be travelled to obtain sufficient food, which would reduce energetic costs and could reduce predation risk when this means the individual can spend more time under cover. A solitary carnivore, the leopard, was found to display relaxed territoriality under high population density, which reduced the risk of injury due to territorial disputes (Roex et al., 2022).

Reproduction

It is sometimes argued that one benefit of group-living is easy access to mates. However, whether finding a mate is difficult rather depends on population density than on sociality. A solitary female that has several male neighbours might easily find a mate without being constrained in choosing which male to mate with compared to a female living in a group with a clear male hierarchy.

One major benefit of solitary living is the absence of direct reproductive competition within groups that can lead to reproductive suppression (behavioural or physiological) in both sexes, and infanticide by females (Clutton-Brock et al., 2006; Faulkes & Bennett, 2001; Schradin et al., 2012). Female infanticide also occurs in solitary species, but it is much more common in group-living species (Lukas & Huchard, 2019). While dominant individuals might benefit from living in groups where they can monopolise reproduction while they and their offspring benefit from helpers, for subordinate individuals of lower competitive ability, solitary living can be beneficial. But even large competitive females might prefer solitary breeding over communal breeding to avoid infanticide, if they are the first females to give birth during the season (Hill et al., 2015; Schradin et al., 2010a).

Infant care

Allo-parental care where offspring especially of dominant individuals benefit is common in group-living species (Kokko et al., 2002; Riedman, 1982). Allo-parental care can lead to indirect fitness benefits if directed towards close kin (Riedman, 1982; Schradin et al., 2018c). Allo-parental care evolves when constraints such as habitat saturation and reproductive suppression by dominants make independent reproduction costly (Emlen, 1982a, 1982b). Solitary living is a tactic to avoid such constraints (Schradin et al., 2010a). Solitary breeding females benefit from being able to invest all care into their own offspring, while solitary males can invest their time into further mate searching for direct reproductive success instead of investing into allo-parental care.

Thermoregulation

Thermoregulation via huddling can lead to significant energy savings, especially in small mammals (Canals et al., 1997; Schradin & Ancel, 2019). For solitary mammals, many of which are small, missing thermoregulatory benefits of huddling represents a major cost of solitary living. In some species, this can be compensated by spending the inactive period in well insulated burrows (Kinlaw, 1999), while other species use sun-basking to reduce thermoregulatory costs (Geiser et al., 2002; Zduniak et al., 2019). So far it is unknown whether solitary species invest more in these mechanisms than group-living species that can use the same mechanisms to save energy.

Benefits of solitary living: Simplicity of life?

Solitary living could be summarised as a strategy to avoid costs of group living: avoiding social transmission of disease and parasites, less conspicuous to predators, no competition for resources within the smaller territory, no social stress and dominance hierarchy causing reproductive suppression, and all parental investment goes into own offspring. However, it also induces the costs of missing all the benefits of group-living, and no study so far tried to measure the net benefit of solitary living. Figure 1 summarises a conceptual framework for such studies.

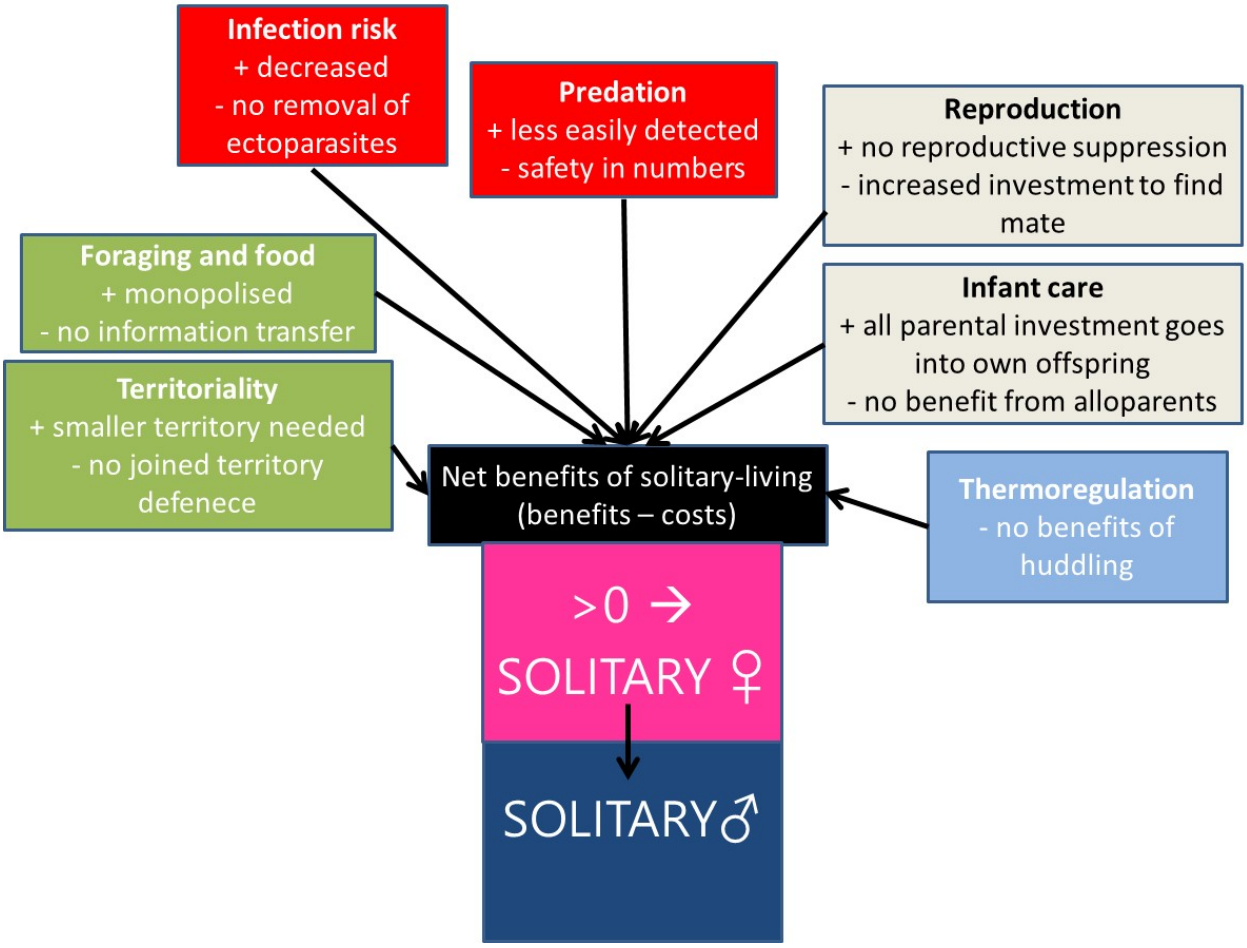

Figure 1. Socio-Ecological Model of Solitary Living. Seven main factors that influence the social organisation of a species. + indicates which condition favours solitary living, - which favours group-living. Combined, these factors influence whether solitary or group-living leads to a net benefit for females. In mammals, social organisation is mainly determined by how females distribute themselves, while males follow female social organisation, being solitary living when females are solitary, joining groups when females form groups.

Conclusions

Solitary living in mammals cannot be regarded as a primitive default stage that needs no scientific explanation. Instead, we must study why some species are solitary living in the same way as we investigated for decades why some species are group-living. For this, costs and benefits and associated ecological drivers of solitary living must be studied. Without a comprehensive understanding of solitary living our understanding of mammalian social evolution is significantly constrained and conservation efforts for solitary mammals will often be inadequate (Olivier et al., 2022c). We hope our review will motivate research groups worldwide to study the socio-ecology and broad diversity of solitary living mammals.

Acknowledgements

The comments of two anonymous referees significantly improved this manuscript. This study was supported by a joint Wits-CNRS PhD program.

Author contributions

Both authors contributed to reviewing the literature and writing the manuscript.

References

Agnani, P., Kauffmann, C., Hayes, L. D., & Schradin, C. (2018). Intra-specific variation in social organization of Strepsirrhines. *American Journal of Primatology, 80*(5), e22758. doi: doi:10.1002/ajp.22758

Andersen, G. E., McGregor, H. W., Johnson, C. N., & Jones, M. E. (2020). Activity and social interactions in a wide-ranging specialist scavenger, the Tasmanian devil (*Sarcophilus harrisii*), revealed by animal-borne video collars. *PLoS ONE, 15*(3), e0230216. doi: 10.1371/journal.pone.0230216

Anderson, T. A. (1998). *Hidden Splenour: A Guide to the Natural History of the Kalahari and Surrounds*: Wildlife and Environment Society of SA.

Andersson, M., & Wiklund, C. G. (1978). Clumping versus spacing out: Experiments on nest predation in fieldfares (*Turdus pilaris*). *Animal Behaviour, 26*, 1207-1212. doi: https://doi.org/10.1016/0003-3472(78)90110-0

Blumstein, D. T., Patton, M. L., & Saltzman, W. (2006). Faecal glucocorticoid metabolites and alarm calling in free-living yellow-bellied marmots. [Article]. *Biology Letters, 2*(1), 29-32. doi: 10.1098/rsbl.2005.0405

Bordes, F., Blumstein, D. T., & Morand, S. (2007). Rodent sociality and parasite diversity. *Biology Letters, 3*(6), 692-694. doi: doi:10.1098/rsbl.2007.0393

Brink, A. S. (1956). Speculations on some advanced mammalian characteristics in the higher mammal-like reptiles. *Palaeontologia Africana, 4*, 77-96.

Brown, C. R., & Brown, M. B. (1986). Ectoparasitism as a cost of coloniality in cliff swallows (*Hirundo pyrrhonot*a). *Ecology, 67*(5), 1206-1218. doi: https://doi.org/10.2307/1938676

Burda, H., Honeyscutt, R. L., Begall, S., Locker-Grütjen, O., & Scharff, A. (2000). Are naked and common mole-rats eusocial and if so, why? *Behavioral Ecology and Sociobiolgy, 47*, 293-303.

Canals, M., Rosenmann, M., & Bozinovic, F. (1997). Geometrical aspects of the energetic effectiveness of huddling in small mammals. *Acta Theriologica, 42*, 321-328.

Clutton-Brock, T. (2021). Social evolution in mammals. *Science, 373*(6561), eabc9699.

Clutton-Brock, T. H. (1989). Mammalian mating systems. *Proceedings of the Royal Society B: Biological Sciences, 236*, 339-372.

Clutton-Brock, T. H., Hodge, S. J., Spong, G., Russell, A. F., Jordan, N. R., Bennett, N. C., Sharpe, L. L., & Manser, M. B. (2006). Intrasexual competition and sexual selection in cooperative mammals. *Nature, 444*, 1065-1068.

Cresswell, W. (1994). Flocking is an effective anti-predation strategy in redshanks, Tringa totanus. *Animal Behaviour, 47*(2), 433-442. doi: https://doi.org/10.1006/anbe.1994.1057

Dalerum, F. (2007). Phylogenetic reconstruction of carnivore social organizations. *Journal of Zoology, 273*(1), 90-97. doi: 10.1111/j.1469-7998.2007.00303.x

Dammhahn, M., & Kappeler, P. M. (2009). Females go where the food is: does the socio-ecological model explain variation in social organisation of solitary foragers? *Behavioral Ecology and Sociobiology, 63*, 939–952. doi: 10.1007/s00265-009-0737-2

Eibl-Eibesfeldt, I. (1953). Zur Ethologie des Hamsters (*Cricetus cricetus* L.). *Zeitschrift für Tierpsychologie, 10*(2), 204-254. doi: https://doi.org/10.1111/j.1439-0310.1953.tb00392.x

Elbroch, L. M., Levy, M., Lubell, M., Quigley, H., & Caragiulo, A. (2017b). Adaptive social strategies in a solitary carnivore. *Science Advances, 3*(10), 10.1126/sciadv.1701218. doi: 10.1126/sciadv.1701218

Emlen, S. T. (1982a). The evolution of helping. I. An ecological constraints model. *Am Nat, 119*(1), 29-39.

Emlen, S. T. (1982b). The evolution of helping. II. The role of behavioral conflict. *Am Nat, 119*(1), 40-53.

Faulkes, C. G., & Bennett, N. C. (2001). Family values: group dynamics and social control of reproduction in African mole-rats. *Trends in Ecology and Evolution, 16*(4), 184-190.

Fernandez-Duque, E., Huck, M., Van Belle, S., & Di Fiore, A. (2020). The evolution of pair-living, sexual monogamy, and cooperative infant care: Insights from research on wild owl monkeys, titis, sakis, and tamarins. *American Journal of Physical Anthropology, 171*(S70), 118-173. doi: https://doi.org/10.1002/ajpa.24017

Gebo, D. L. (2004). A shrew-sized origin for primates. In S. Stinson (Ed.), *Yearbook of Physical Anthropology, Vol 47* (Vol. 47, pp. 40-62). New York: Wiley-Liss, Inc.

Geiser, F., Goodship, N., & Pavey, C. R. (2002). Was basking important in the evolution of mammalian endothemy? *Naturwissenschaften, 89*, 412-414.

Griesser, M., & Suzuki, T. N. (2016). Occasional cooperative breeding in birds and the robustness of comparative analyses concerning the evolution of cooperative breeding. *Zoological Letters, 2*, 7. doi: 10.1186/s40851-016-0041-8

Groenewald, G. H., Welman, J., & Maceachern, J. (2001). Vertebrate burrow complexes from the early Triassic Cynognathus zone (Driekoppen formation, Beaufort group) of the Karoo basin, South Africa. *PALAIOS, 16*, 148-160. doi: 10.2307/3515526

Hart, B., & Hart, L. (2019). Sickness behavior in animals: implications for health and wellness *Encyclopedia of animal behavior* (pp. 171-175): Elsevier Ltd.

Hill, D. L., Pillay, N., & Schradin, C. (2015). Alternative reproductive tactics in female striped mice: heavier females are more likely to breed solitarily than communally. *Journal of Animal Ecology, 84*(6), 1497-1508. doi: 10.1111/1365-2656.12431

Huck, M., Di Fiore, A., & Fernandez-Duque, E. (2020). Of apples and oranges? The evolution of "monogamy" in non-human primates. [Review]. *Frontiers in Ecology and Evolution, 7*(472). doi: 10.3389/fevo.2019.00472

Jackson, T. P. (1999). The social organisation and breeding system of Brants´ whistling rat (*Parotomys brantsii*). *Journal of Zoology (London), 247*, 323-331.

Jaeggi, A. V., Miles, M. I., Festa-Bianchet, M., Schradin, C., & Hayes, L. D. (2020). Variable social organization is ubiquitous in Artiodactyla and probably evolved from pair-living ancestors. *Proceedings of the Royal Society B: Biological Sciences, 287*(1926), 20200035. doi: doi:10.1098/rspb.2020.0035

Jasinoski, S. C., & Abdala, F. (2017). Aggregations and parental care in the Early Triassic basal cynodonts *Galesaurus planiceps* and *Thrinaxodon liorhinus*. *PeerJ, 5*, e2875. doi: 10.7717/peerj.2875

Kappeler, P. M. (2019). A framework for studying social complexity. [journal article]. *Behavioral Ecology and Sociobiology, 73*(1), 13. doi: 10.1007/s00265-018-2601-8

Kappeler, P. M., & Schaik, C. P. v. (2002). Evolution of primate social systems. *International Journal of Primatology, 23*, 707-740.

Kerth, G. (2022). Long-term field studies in bat research: importance for basic and applied research questions in animal behavior. *Behavioral Ecology and Sociobiology, 76*(6), 75. doi: 10.1007/s00265-022-03180-y

Kinlaw, A. (1999). A review of burrowing by semi-fossorial vertebrates in arid environments. *Journal of Arid Environments, 41*(2), 127-145. doi: https://doi.org/10.1006/jare.1998.0476

Kleiman, D. G., & Malcolm, J. R. (1981). The evolution of male parental investment in mammals. In D. J. Gubernick & P. H. Klopfer (Eds.), *Parental Care in Mammals* (pp. 347-387). New York: Plenum Press.

Kokko, H., Johnstone, R. A., & Wright, J. (2002). The evolution of parental and alloparental effort in cooperatively breeding groups: when should helpers pay to stay? *Behavioral Ecology, 13*, 291-300.

Krause, J., & Ruxton, G. D. (2002). *Living in Groups*. Oxford: Oxford University Press.

Kvarnemo, C. (2018). Why do some animals mate with one partner rather than many? A review of causes and consequences of monogamy. *Biological Reviews, 93*(4), 1795-1812. doi: doi:10.1111/brv.12421

Ladeveze, S., de Muizon, C., Beck, R. M. D., Germain, D., & Cespedes-Paz, R. (2011). Earliest evidence of mammalian social behaviour in the basal Tertiary of Bolivia. [10.1038/nature09987]. *Nature, 474*(7349), 83-86. doi: http://www.nature.com/nature/journal/v474/n7349/abs/10.1038-nature09987-unlocked.html#supplementary-information

Lawson Handley, L. J., & Perrin, N. (2007b). Advances in our understanding of mammalian sex-biased dispersal. *Molecular Ecology, 16*, 1559 - 1578.

LeRoux, A., Jackson, T. P., & Cherry, M. I. (2002). Differences in alarm vocalizations of sympatric populations of the whistling rat, *Parotomys brantsii* and *P. littledalei* (Rodentia: Muridae). *Journal of Zoology (London), 257*, 187-194.

Lopez, J., Wey, T., & Blumstein, D. (2013). Patterns of parasite prevalence and individual infection in yellow-bellied marmots. *Journal of Zoology, 291*. doi: 10.1111/jzo.12076

Lukas, D., & Clutton-Brock, T. (2017). Climate and the distribution of cooperative breeding in mammals. *Royal Society Open Science, 4*(1), doi:10.1098/rsos.160897. doi: 10.1098/rsos.160897

Lukas, D., & Clutton-Brock, T. H. (2013b). The evolution of social monogamy in mammals. *Science, 341*(6145), 526-530. doi: 10.1126/science.1238677

Lukas, D., & Huchard, E. (2019). The evolution of infanticide by females in mammals. *Philosophical Transactions of the Royal Society B: Biological Sciences, 374*(1780), 20180075. doi: doi:10.1098/rstb.2018.0075

Lutermann, H., Bennett, N., Speakman, J., & Scantlebury, D. (2013). Energetic benefits of sociality offset the costs of parasitism in a cooperative mammal. *PloS one, 8*, e57969. doi: 10.1371/journal.pone.0057969

Makuya, L., Olivier, C.-A., & Schradin, C. (2022). Field studies need to report essential information on social organisation – independent of the study focus. *Ethology, 128*(3), 268-274. doi: https://doi.org/10.1111/eth.13249

Makuya, L., Pillay, N., Rimbach, R., & Schradin, C. (2023). Suppressing the physiological stress response is not stress. *Trends in Ecology and Evolution*(3), 224-227.

Müller, A. E., & Thalmann, U. (2000). Origin and evolution of primate social organisation: a reconstruction. *Biological Reviews, 77*, 405-435.

Nowak, R. M., & Wilson, E. (1999). *Walker`s Mammals of the World* (6th ed.). Baltimore: John Hopkins University Press.

Olivier, C.-A. (2023). *Evolution de l'organisation sociale et sa variabilité chez les mammifères (Evolution of social organisation and its variation in Mammals). PhD Thesis.* PhD PhD Thesis, University of Strasbourg, Strasbourg, France.

Olivier, C.-A., Jaeggi, A. V., Hayes, L. D., & Schradin, C. (2022a). Revisiting the components of Macroscelidea social systems: Evidence for variable social organization, including pair-living, but not for a monogamous mating system. *Ethology, 128*(5), 383-394. doi: https://doi.org/10.1111/eth.13271

Olivier, C.-A., Martin, J. S., Pilisi, C., Agnani, P., Kauffmann, C., Hayes, L., Jaeggi, A. V., & Schradin, C. (2024). Primate social organization evolved from a flexible pair-living ancestor. *Proceedings of the National Academy of Sciences, 121*(1), e2215401120.

Olivier, C.-A., Schradin, C., & Makuya, L. (2022c). Global change and conservation of solitary mammals. [Opinion]. *Frontiers in Ecology and Evolution, 10*. doi: 10.3389/fevo.2022.906446

Patterson, J. E. H., & Ruckstuhl, K. E. (2013). Parasite infection and host group size: a meta-analytical review. *Parasitology, 140*(7), 803-813. doi: 10.1017/S0031182012002259

Qiu, J., Olivier, C.-A., Jaeggi, A. V., & Schradin, C. (2022). The evolution of marsupial social organization. *Proceedings of the Royal Society B: Biological Sciences, 289*(1985), 20221589. doi: doi:10.1098/rspb.2022.1589

Riedman, M. L. (1982). The evolution of alloparental care and adoption in mamals and birds. *The Quarterly Review of Biology, 57*(4), 405-435.

Roex, N. l., Mann, G. K. H., Hunter, L. T. B., & Balme, G. A. (2022). Relaxed territoriality amid female trickery in a solitary carnivore. *Animal Behaviour, 194*, 225 - 231. doi: https://doi.org/10.1016/j.anbehav.2022.09.022

Schoepf, I., Schmohl, G., König, B., Pillay, N., & Schradin, C. (2015). Manipulation of population density and food availability affects home range sizes of African striped mouse females. *Animal Behaviour, 99*(0), 53-60. doi: http://dx.doi.org/10.1016/j.anbehav.2014.10.002

Schradin, C. (2017). Comparative studies need to rely both on sound natural history data and on excellent statistical analysis. *Royal Society Open Science 4*(9), 170346. doi: http://dx.doi.org/10.1098/rsos.170346

Schradin, C., & Ancel, A. (2019). Huddling. In J. Vonk & T. Shackelford (Eds.), *Encyclopedia of Animal Cognition and Behavior* (pp. 1-5). Cham: Springer International Publishing.

Schradin, C., Hayes, L. D., Pillay, N., & Bertelsmeier, C. (2018a). The evolution of intraspecific variation in social organization. *Ethology, 124*, 527-536. doi: doi:10.1111/eth.12752

Schradin, C., König, B., & Pillay, N. (2010a). Reproductive competition favours solitary living while ecological constraints impose group-living in African striped mice. *Journal of Animal Ecology, 79*, 515-521.

Schradin, C., Lindholm, A. K., Johannesen, J., Schoepf, I., Yuen, C.-H., König, B., & Pillay, N. (2012). Social flexibility and social evolution in mammals: a case study of the African striped mouse (*Rhabdomys pumilio*). *Molecular Ecology, 21*, 541-553. doi: 10.1111/j.1365-294X.2011.05256.x

Schradin, C., Makuya, L., Pillay, N., & Rimbach, R. (2023). Harshness is not stress. *Trends in Ecology & Evolution, 38*(3), 224-227. doi: https://doi.org/10.1016/j.tree.2022.12.005

Schradin, C., Schmohl, G., Rödel, H. G., Schoepf, I., Treffler, S. M., Brenner, J., Bleeker, M., Schubert, M., König, B., & Pillay, N. (2010b). Female home range size is regulated by resource distribution and intraspecific competition: a long-term field study. *Animal Behaviour, 79*(1), 195-203.

Schradin, C., Vuarin, P., & Rimbach, R. (2018c). The neoteny-helper hypothesis: When to expect and when not to expect endocrine mechanisms to regulate allo-parental care? *Physiology and Behavior, 193*, 127-134. doi: https://doi.org/10.1016/j.physbeh.2017.12.008

Siracusa, E., Boutin, S. A., Dantzer, B., Lane, J. E., Coltman, D. W., & McAdam, A. G. (2021). Familiar neighbors, but not relatives, enhance fitness in a territorial mammal. *Current Biology, 31*, 438-455. doi: 10.1016/j.cub.2020.10.072

Tamura, N. (1989). Snake-directed mobbing by the Formosan squirrel *Callosciurus erythraeus thaiwanensis*. *Behavioral Ecology and Sociobiology, 24*(3), 175-180. doi: 10.1007/BF00292100

Tamura, N., Hayashi, F., & Miyashita, K. (1989). Spacing and kinship in the Formosan squirrel living in different habitats. *Oecologia, 79*(3), 344-352. doi: 10.1007/BF00384313

Taylor, W. A., & Skinner, J. D. (2003). Activity patterns, home ranges and burrow use of aardvarks (*Orycteropus afer*) in the Karoo. *Journal of Zoology (London), 261*, 291-297.

Thierry, B., Iwaniuk, A. N., & Pellis, S. M. (2000). The influence of phylogeny on the social behaviour of macaques (Primates: Cercopithecidae, genus *Macaca*). *Ethology, 106*, 713-728.

Valomy, M., Hayes, L. D., & Schradin, C. (2015b). Social organization in Eulipotyphla: evidence for a social shrew. *Biology Letters, 11*(11), doi:10.1098/rsbl.2015.0825.

Viblanc, V. A., Saraux, C., Murie, J. O., & Dobson, F. S. (2016a). Kin effects on energy allocation in group-living ground squirrels. *Journal of Animal Ecology, 85*, 1361-1369. doi: 10.1111/1365-2656.12541

Walmsley, S. F., Boutin, S., Dantzer, B., Lane, J. E., Coltman, D. W., & McAdam, A. G. (2023). Benefits of living closer to kin vary by genealogical relationship in a territorial mammal. *Proceedings of the Royal Society B: Biological Sciences, 290*, 20221569. doi: https://doi.org/10.1098/rspb.2022.1569

Weaver, L. N., Varricchio, D. J., Sargis, E. J., Chen, M., Freimuth, W. J., & Wilson Mantilla, G. P. (2020). Early mammalian social behaviour revealed by multituberculates from a dinosaur nesting site. *Nature Ecology and Evolution*. doi: 10.1038/s41559-020-01325-8

Zduniak, M., Pillay, N., & Schradin, C. (2019). Basking African striped mice choose warmer locations to heat up: evidence from a field study. *Journal of Zoology, 309*(2), 133-139. doi: 10.1111/jzo.12687

CHAPTER 3. Measuring range sizes in a 100 g rodent: mini-GPS are more reliable than transmitters, but the location error reduces reliability.

Makuya, L[1]., & Schradin, C[1,2]. (2023). Measuring range sizes in a 100-g rodent: mini-GPS are more reliable than transmitters, but the location error reduces reliability. *Mammalian Biology*, *103*(5), 455-465. doi: https://doi.org/10.1007/s42991-023-00365-4.

Authors' affiliations:

[1] School of Animal, Plant & Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS 2050, Johannesburg, South Africa

[2] IPHC, UNISTRA, CNRS, 23 rue du Loess, 67200 Strasbourg, France

Abstract

Home ranges of free-living mammals have typically been studied via radio-tracking to understand how individuals use their environment. Recently, GPS collars have become popular in large mammals. However, GPS collars are rarely used in small mammals, as they are too heavy, especially when needing coating to protect against gnawing. Here we test the efficiency of mini-GPS collars to measure range estimates compared to the use of radio-collars in a small rodent of 100 g body mass. We equipped 20 bush Karoo rats with mini-GPS loggers and thereafter with radio-transmitters to determine ranges. We validated the accuracy of the mini-GPS loggers by comparing them with the fixes from a handheld GPS and found both to be similar. We estimated range sizes using both traditional methods of Kernel and minimum convex polygon estimates as well as modern methods from movement ecology taking the location error of the mini-GPS into account. Using modern methods led to smaller range estimates, but results were in so far consistent that daily ranges for bush Karoo rats determined using mini-GPS were much larger than home range estimates from radio tracking. Using radio-tracking enabled us to establish the central shelter, while the mini-GPS revealed areas where rats had been observed foraging. We found a distinct location error and therefore suggest using modern approaches from movement ecology which can take this error into account. In sum, mini-GPS revealed more accurate estimates of the ranges than radio-tracking in a small rodent of 100 g body mass.

Keywords: data logger; home range; rodent; small mammal; territory

Introduction

Studying the home range sizes and daily ranges of animals is important to understand how they use their environment and the constituent resources (Börger et al., 2008; Burt, 1943; Potts & Lewis, 2014). Animals occupy areas that allow them to acquire sufficient resources for reproduction and survival, minimizing time and energy for territory defence (Maynard Smith, 1974; Powell, 2000). While the home range has been defined as "the area traversed by an animal during its normal activities such as food gathering, mating, and caring for young" (Burt, 1943), the home range sizes in turn vary by resource availability and distribution (Schoepf et al., 2015; Schradin et al., 2010b). For example, the female striped mice (*Rhabdomys pumilio*) displayed seasonal variation in home range size, with the home range size being larger in the wet season than in the dry season (Edelman & Koprowski, 2006; Schradin & Pillay, 2006). Lastly, significant sex differences exist during the breeding season, with males having larger home ranges than females (Eccard et al., 2004; Inoue, 1991). While ascertaining range size is important, this requires high investment.

Some techniques of assessing animal ranges date back to tracking spoors, direct observations, spooling animals, capture mark-recapture, and radio-tracking. For example, the home ranges of Kalahari lions (*Panthera leo*) and leopards (*Panthera pardus*) were studied via spoor tracking over several months (du P Bothma & Le Riche, 1984). These studies also involved direct focal animal observation. Spooling, where a thread is attached to the animal to detect its movement, has been used in studies of cryptic small mammals (Hawkins & Macdonald, 1992). Earlier studies of animal ranges also included capture mark-recapture trapping, whereby an animal was trapped, marked, released and trapped again (reviewed by Mohr (1947)). Improved technology has allowed animals to be equipped with radio-collars and then to be found using radio-tracking (Gardner & Serena, 1995; Loy et al., 1994; Ophir et al., 2008; Schneider & Kappeler, 2016) or even via automated radio-tracking (DeGregorio et al., 2018; Ward et al., 2013). Techniques to assess animal ranges differ in accuracy, with radio-tracking being more accurate than spoor tracking, spooling, and capture mark-recapture, yet all these methods require high-time investment (Kays et al., 2015).

The more recently developed GPS collars offer an accurate low-time investment technology and have been used to collect data for the range use of large mammals such as African ungulates (Owen-Smith & Goodall, 2014) and European brown bears (*Ursus arctos*) (De Angelis et al., 2021). Examples of medium sized mammals that have been equipped with GPS collars include the ~5 kg red fox (*Vulpes vulpes*, Walton & Mattisson (2021)), different mongoose species of ~4 kg (Herpestidae, Streicher et al. (2020)), the ~1 kg West European hedgehog (*Erinaceus europaeus*, Gazzard et al. (2022)), the 1.3 kg marsupial woylie (*Bettongia penicillate*; Bateman et al. (2017)), the 400 g pygmy rabbit (*Brachylagus idahoensis*, McMahon

et al. (2017)) and the 165 g golden-mantled ground squirrels (*Callospermophilus laterali*; Hefty & Stewart (2019)). However, the majority of mammal species are even smaller than that: more than 4000 out of all 6399 extant mammal species are small with a body mass below 100g, including members of orders Rodentia, Chiroptera, and Eulipotyphla (Burgin et al., 2018). Until now, GPS collars were typically too heavy to attach on small mammals. This is because most small mammals are rodents (2552 species; (Burgin et al., 2018)) that are known to gnaw at any devices. Thus, any collar (radio- or GPS) needs extensive protective coating that typically weighs more than the device itself. GPS collars have been used on bats that are less than 100 g, but these collars do not need heavy coating compared to when they would be used on rodents of a similar weight (Hefty & Stewart, 2019; Vleut et al., 2019). For example, the sizes of the coated collars used on the mongoose species and the red fox were 52 g and 210 g respectively (Streicher et al., 2020; Walton & Mattisson, 2021), both being too large to be used on smaller mammals.

So far, it is not well known whether the accuracy of GPS collar data is comparable, better or worse than that of radio-collars. This might depend on many factors, including range size and life history. GPS collars might generate more accurate data than radio-tracking in small mammals that are central place foragers as they interfere less with an animals' behaviour. Central place foragers have a defined shelter, from which they forage within close proximity and easily return to the shelter in case of any disturbance by a predator (Orians & Pearson, 1979). Examples include the colonial Brants' whistling rat (*Parotomys brantsii*, Jackson (2001)), Columbian ground squirrels (*Urocitellus columbianus*, Andrusiak & Harestad (1989)), golden-mantled ground squirrels (*Callospermophilus laterali*; Hefty & Stewart (2019)) and many species of marmots (*Marmota* spp., Lehrer et al. (2012)). Radio-tracking such small mammals often involves the 'homing in' technique to obtain an accurate measure of the home range, i.e. the researcher approaches the animal from different directions until its exact position is located (Kenward, 1987). However, this technique may disturb the natural ranging behaviour of the studied animals. In central place foragers this might lead to most fixes indicating the nest location and not the animal's entire home range. In contrast, GPS collars do not disturb the animal and collect data while the animal is moving in its environment. Another method that offers disturbance free tracking would be automated tracking of animals carrying radio-collars (automated very high frequency VHF tracking) that has been used on small mammal and also has similar inter-fix intervals as GPS (Eccard et al., 2022; Hoffmann et al., 2019; Kowalski et al., 2019; Schirmer et al., 2020; Schlägel et al., 2019). However, automated tracking is expensive in time and money. It needs base stations that are to be maintained which is why it is best done at permanent research sites that have a technician. As it requires more than one person to be in the field it is not suitable for a single PhD student or researcher collecting data at a remote field site.

GPS collars have the problem of the location error, which means the tendency for positions to drift when the unit is stationary, e.g., due to the Kalman filter (Frair et al., 2010; Gunner et al., 2022). The location error is the distance between the GPS-generated location and the true location. It occurs as every GPS has some inaccuracy associated with it. The location error is further affected by environmental characteristics (e.g., terrain, vegetation) satellite acquisition (e.g., number of satellites detected, satellite geometry) and animal behaviour (including the position of the GPS on the animal). The location error becomes more important when the measured range size becomes smaller. Thus, when using GPS collars on small mammals, it is important to determine the location error to validate the method and if possible, to correct for it. No study has compared the range sizes obtained via mini-GPS loggers with those obtained from radio-tracking in small mammals, especially not while taking the location error into account.

Here, we present data on one small central place forager, the bush Karoo rat (*Otomys unisulcatus*), and compare the calculated range sizes obtained from mini-GPS loggers with those obtained from radio-tracking. We also compared between traditional methods of range estimation (Kernel contours and minimum convex polygons) and modern approaches from movement ecology (namely the autocorrelated kernel density estimation (AKDE), Fleming et al. (2015)) which can take the location error into account. Bush Karoo rats live in the Succulent Karoo biodiversity hotspot in South Africa, characterized by very dry and hot summers. The bush Karoo rat was selected as the study animal as it is diurnal and occupies an open habitat and can be directly observed in the field (Schradin, 2005). It builds stick lodges within large shrubs, and these lodges create a favourable micro-climate to buffer against temperature extremes (Brown & Willan, 1991b; du Plessis & Kerley, 1991; du Plessis et al., 1992). It forages in proximity of its lodge(s), into which it withdraws if disturbed. Their territories can contain up to four lodges connected by runways and additional smaller shrubs providing cover. We expected bush Karoo rats to withdraw into their lodge when homing in during radio-tracking. Thus, we predicted mini-GPS data to reveal larger ranges than radio-tracking, which would represent a more accurate estimation of their range use.

Methods

Study area and period

The study was conducted at a 4-hectare field site adjacent to the Succulent Karoo Research Station located in the Goegap Nature Reserve, approximately 15 km from Springbok in the Northern Cape, South Africa. Data collection took place during the dry, non-breeding summer season from February until April 2022. The field site is in an open and flat environment, reducing risks of GPS inaccuracy.

Trapping, marking and observation of rats

Animal ethics clearance was provided by the University of the Witwatersrand (AESC 2018/03/15B). We placed a combination of foldable Sherman traps and locally produced metal traps of the Sherman style arranged around the entrances of occupied lodges (as indicated by the presence of faeces and plant material) and along active runways. Four traps were added per lodge and each lodge was trapped twice a month, each time for 3 consecutive days. Trapped rats were weighed, sexed and permanently marked with metal ear tags applied on both ears (National Band and Tag Co., Newport, KY, U.S.A.). We conducted focal animal observations before, during and after the attachment of collars to see if there was any indication that the collars influenced the behaviour of the rats.

Mini-GPS dataloggers

Altogether, 20 bush Karoo rats (14 females and 6 males) were fitted with Gipsy mini-GPS datalogger collars (Technosmart, Rome, Italy), which weighed 4.5 g, representing less than 5 % of body weight (Fig. 1) (Murray & Fuller, 2000). Rats weighed 96.6 $\pm$ 19.3 g (range: 67.5-131.2 g). Data collection started with a delay of 2 days after collar attachment to allow the bush Karoo rats to get accustomed to them (verified by focal animal observations at the lodge the day before the mini-GPS collected data). The Gipsys were programmed to collect GPS fixes (longitude, latitude, altitude, date, and time) on day 3 starting at 6 AM every 5 minutes, battery life did not allow us to collect data for more than one day. We chose this frequency as in our main running study (to be finished in 2023) we want to compare in how far neighbouring rats share their daily ranges (being in the same area at the same time), necessitating a higher frequency than for example one fix every 2 hours. One fix every 5minutes was the frequency allowing us to obtain the highest number of fixes possible within 24 hours given the battery size. On day 4, we started to re-trap animals to remove the Gipsys. Data were collected from 8 rats using the model Gipsy5 and for 12 rats using the model Gipsy6; the two logger models differed slightly in their assessment of fix accuracy (see below).



Figure 1. A Bush Karoo rat carrying a mini-GPS datalogger. This female is marked with blond hair dye on the head and back, for individual recognition during behavioural observations

Radio collars (radio transmitters)

Following removal of the mini-GPS dataloggers, the rats were fitted with PD-2C radio transmitters (Holohil, Carp, Ontario, Canada), which weighed 3.3 g including the collar and were less than 5 % of their body weight. GPS collars are less reliable than radio-collars, which is why we always first checked whether the Gipsys had successfully collected data (which was the case in 20 of 24 rats), before attaching radio-collars. This allowed us to reduce the number of individuals used in the study. Individuals were equipped with radio transmitters for a mean duration of 16.6 days (range 8–27 days). Radio-tracking was performed by using an AOR 8000 wide-range receiver (Tokyo, Japan) and a Telonics RA-14K antenna (Mesa, Arizona) and we followed the same procedure as in multiple studies for striped mice on the same field site (Schradin & Pillay, 2006; Schradin et al., 2010b). Both species are diurnal but with an activity peak during mornings and afternoons (Schradin, 2006). Striped mice seek shelter in different shrubs during the hot periods of the day and rarely in their main nesting site (Schradin, 2006). The same might be true for bush Karoo rats, which often have several lodges and shade providing shrubs in their territory, as indicated by our observations and trapping data (Schradin, 2006).

Individuals were radio-tracked for 7 days. Fixes were taken 6 times a day between 9 AM to 6 PM. To ensure independence of data points (Kenward, 1987), intervals between fixes were approximately 1h30 mins apart, giving rats enough time to traverse through their entire home range, except during the hottest part of the day (from 12 PM to 2 PM), when rats are inactive. Fixes were obtained by using the 'homing in' method: an individual was approached until we either saw it or identified the bush in which it was located. The location of the bush Karoo rat was then recorded with a handheld GPS (eTrex Venture, GARMIN International, Olathe, Kansas). In 4.2% of cases, the position of individuals was confirmed by visual sightings.

The coldest mean temperatures (recorded directly at the research station) during days we radio-tracked bush Karoo rats differed significantly from the temperatures on which they carried mini-GPS (14.8 ± 1.8 °C vs 17.3 ± 2.6°C, paired $t_{19} = 4.6$, $p = 0.0002$), but the hottest mean temperatures during days we radio-tracked did not differ significantly from the temperatures on which they carried mini-GPS (31.2 ± 2.3 °C vs 32.4 ± 1.4°C, paired $t_{19}=1.67$, $p = 0.113$). However, the rats are not active during the coldest times of the day and we would therefore not have obtained fixes from them with the mini-GPSs.

Validating mini-GPS accuracy

The accuracy of fixes with the handheld eTrex Venture GPS is specified to be ± 5 m by the manufacturer (GARMIN International). Accuracy of Gipsy5 dataloggers has been reported to be approximately 5 m (information provided by Technosmart) to 10 m (Fleming et al., 2021). Accuracy of the Gipsy6 mini-GPS logger was said to be approximately ± 5 m (information provided by Technosmart). As the accuracy of fixes can influence range estimates, we compared the two GPS instruments. For this, we took an 11 km hike through the Goegap Nature Reserve, with four mini-GPS loggers mounted onto a hat worn by one of the hikers while we used a handheld eTrex Venture GPS to collect the track data. We then compared fixes from the mini-GPS loggers with the track data and determined the distance of the logger fixes from the track. We did this in QGIS (QGIS, 2009) using the distance to nearest hub (points) toolkit. The source points were the Gipsy points, and the destination points were the track points from the handheld GPS. We excluded fixes with an accuracy so low that we would have removed them from data analysis (see below).

For a measure of accuracy, Gipsy5 units record the HDOP (Horizontal Dilution of Precision), a value which describes the geometric strength of satellite configuration on GPS accuracy; lower values indicate higher accuracy) (Technosmart, Rome, Italy). Fixes with an HDOP > 5 were excluded.

Gipsy6 units record horizontal accuracy estimates for fixes, and provide accuracy data in five categories: > 100 m, < 100 m, < 50 m, < 15 m, and < 10 m. It is important to note that these categories are rough estimates as in contrast to a large handheld GPS, the mini-GPS cannot directly determine their accuracy (information provided by Technosmart). In other words, a horizontal accuracy estimate of < 50 m means the fix is probably more accurate than one of the category < 100 m, but it does not mean the fix is 50 m away from the real position. To be able to rank the horizontal accuracy categories for how useful they are for our study species, we visualised the fixes of 15 individuals (of which 12 were also radio-tracked, 3 were used for other studies) in QGIS. We then related the five Gipsy categories to our own three accuracy categories: accuracy 1 - data are within the expected area, all fixes were at places where the individuals had been observed by us during field work; accuracy 2 - fixes with this accuracy were mainly in the area where the individual had been observed, with some being outside and some being more than 15 m away from the remaining fixes, indicating they could be possible outliers; and accuracy 3 - many fixes were more than 15 m or even more than 50 m away from the area where the individual had been observed, indicating data inaccuracy.

We determined the location error for the Gipsy6 units by collecting calibration data using stationary tests. Four units were placed near lodges where a bush Karoo rat carried a Gipsy and allowed to collect fixed location data for 24 hours. The calibration data were then used to estimate the location error through the *ctmm* (Calabrese et al., 2016) workflow in R and RStudio (Posit team, 2022; R Core Team, 2022a) and accounting for it before continuing with calculating the range sizes (Fleming et al., 2021). We also visually compared the data from the stationary Gipsys with the Gipsys carried by bush Karoo rats inhabiting the lodges.

Data analysis

For Gipsy5, fixes with an HDOP > 5 were excluded. The remaining fixes were visualised in QGIS and isolated outliers far (> 15 m) from the main lodge of the rat and from other fixes were excluded. For Gipsy6, fixes with a horizontal accuracy better than 50 m (i.e. < 50) were included; see results section for validation of this procedure.

We used several methods to calculate home range sizes. First, using the package *adehabitatHR* (Calenge, 2015) in R (R Core Team, 2022a) we calculated the traditional measures used in radio-tracking studies: For comparison with other species, home range size was estimated as minimum convex polygons (MCP) which calculate the home range size by drawing a convex polygon around the individuals' point data (Mohr, 1947; Powell, 2000). In contrast, Kernel density estimators calculate the probability of finding an individual in a

particular location by using un-parametric statistics (Worton, 1989). To account for the influence of outliers that may be missed during the cleaning process, we estimated 95 % MCP and Kernel range sizes.

Second, we used the autocorrelated Kernel density estimation (AKDE) as implemented in the R packages *ctmm* (Calabrese et al., 2016) and *ctmmweb* (Dong et al., 2018) used in movement ecology for both GPS and radio-tracking data. This approach allowed us to account for location error in the calculations of range sizes within the *ctmm* workflow through two stages (Fleming et al., 2021). First, the calculations from the autocorrelation and bandwidth are error-informed, which mitigates various biases in autocorrelation and bandwidth estimates that would otherwise occur had location error been ignored. Second, location estimates are fed through a Kalman smoother before kernel placement, which counteracts overdispersal (Silva et al., 2022). Range estimates using the *ctmm* workflow were only possible for the 12 Gipsy6 units and not the 8 Gipsy5 units, as location errors were determined after the main study (due to comments by colleagues), when no Gipsy5 unit was available anymore. We present data for all approaches to enable the reader to compare the traditional with the more modern ways of home range analyses.

We ran a linear mixed model using the *lme4* (Bates et al., 2015) package to analyse factors influencing the range size estimates. As the data were normally distributed, we used the gaussian family with the identity link function. The estimated range size was the dependent variable and as independent variable we included the equipment (Gipsy or radio-tracking), the used calculation model (MCP, Kernel, AKDE), and the sex of the individual. Due to the paired data, we included individual ID as random factor. We assessed model assumptions by inspecting Q–Q plots and by plotting model residuals against fitted values. We then performed post hoc analyses using the *emmeans* function from the *emmeans* package (Lenth, 2021) to determine pairwise differences in range size between the tracking method and estimation method. We adjusted *p* values using the Tukey method.

Results

Validating mini-GPS accuracy

Most of the fixes of the mini-GPS logger were on the track of the eTrex Venture handheld GPS (Fig. 2). Only at the start, when the Gipsy were switched on, and in a narrow valley, did some noticeable discrepancies occur (arrows in Fig. 2). For Gipsy5 units, fixes were on average 9 ± 12.3 m (excluding about 5 outliers) away from the track, and for Gipsy6 4.4 ± 3.7 m (excluding 3 outliers). As fixes of the handheld GPS and the Gipsys were not taken at the same time, the real accuracy was higher.

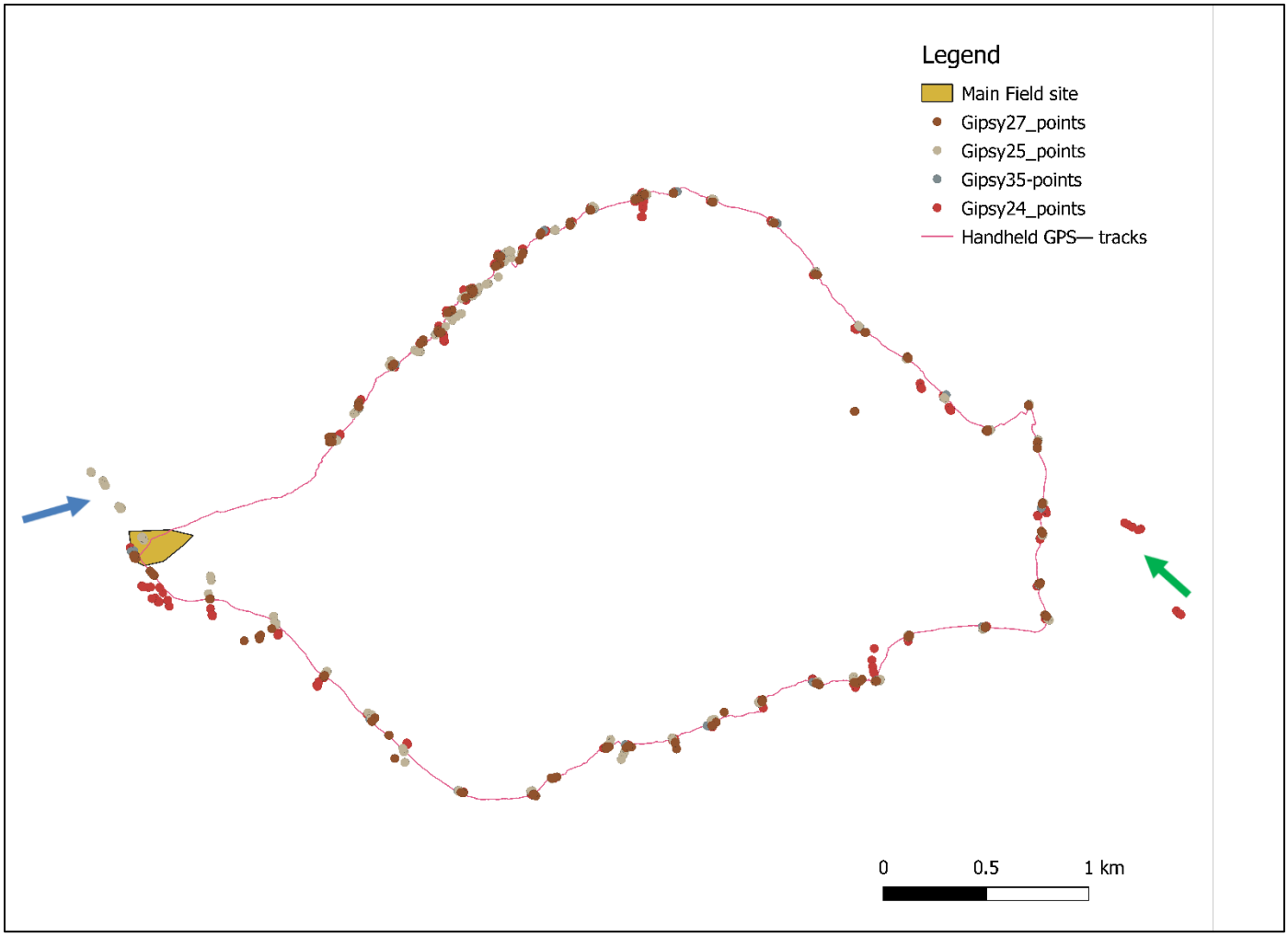

Figure 2. Track of a handheld extrex Venture GPS used during radio-tracking along a 11km hike. In addition, fixes taken every 5 minutes of 3 Gipsy 5 units (Gipsy units 24, 25 and 27) and one Gipsy 6 unit (Gipsy 35) are shown. At the start of the hike (left, blue arrow), fixes from Gipsy 25 were off the track, and during the middle of the hike (right, green arrow) in a narrow valley, fixes of Gipsy 24 were off the track. Otherwise, most Gipsy fixes were on the handheld GPS track

Gipsy6: Location error and accuracy depending on horizontal accuracy

The calculated location error for the four Gipsy6 units was 1.98 ± 0.16 m. Fixes with a horizontal accuracy of > 50 m were often unreliable outliers far away from areas where the animal had been observed during focal animal observations (Table 1). Therefore, we only included fixes with an accuracy better than 50 m for all further analysis.

Table 1 Categorising fixes of 12 bush Karoo rats (F: females, M: males) carrying Gipsy 6 units. Fixes were visualised depending on their horizontal accuracy category from > 100 m to < 10 m. When all fixes were close to each other and in the expected area where the individual had been observed, the accuracy was categorised as "Good". An accuracy where fixes with some potential outliers (fixes more than 15 m away from expected range) occurred that could be removed by using 95% contours was categorised as "Few outliers". Accuracy where many

fixes were far away from the expected area (more than 50 m) was categorised as "Many outliers". "NA": no fixes were recorded in this accuracy category. The Gipsy unit of M111 had a broken antenna and was not used anymore.

Individual	**> 100 m**	**< 100 m**	**< 50 m**	**< 15 m**	**< 10 m**
F98	Many outliers	Few outliers	Good	Good	NA
F105	Many outliers	Few outliers	Good	Good	NA
F126	Many outliers	Few outliers	Few outliers	Good	Good
F204	Many outliers	Few outliers	Good	Good	NA
F266	Many outliers	Few outliers	Good	Good	NA
F270	NA	Many outliers	Few outliers	Good	Good
F288	Many outliers	Few outliers	Few outliers	Good	Good
F356	NA	Many outliers	Good	Few outliers	Good
M111	Many outliers	Many outliers	NA	NA	NA
M129	Many outliers	Few outliers	Good	NA	NA
M299	Many outliers	Few outliers	Good	Good	NA
M303	NA	Good	Few outliers	Good	Good

Comparison of home ranges

The range estimates were significantly influenced by the tracking technique ($p < 0.001$), the calculation method used ($p = 0.04$), but not by sex ($p = 0.09$; Table 2). Home range estimates using radio-tracking were significantly smaller than those from mini-GPS units for all 3 measures (Fig. 3). During radio-tracking, bush Karoo rats were generally found in their main lodge (Fig. 4). Pairwise comparisons of range size between tracking method and estimation method showed no significant differences between the estimation methods (AKDE vs Kernel vs MCP; all $p > 0.09$; Fig. 3) but significant differences between the tracking methods (radio tracking vs gipsy, all $p < 0.001$).

Table 2. Results of the linear mixed models to test how range estimates were influenced by whether estimates were done using 3 different methods: minimum convex polygons (MCP), Kernel density (Kernel), or autocorrelated Kernel density estimation; and whether data were from radio-tracking or mini-GPS. Factors in bold type indicate significant predictors.

Predictors	*Estimates*	95% CI	*p*
(Intercept)	438.74	439.77 – 76.22	**<0.001**
Method (Kernel)	-150.54	149.93 – 73.93	**0.04**
Method (MCP)	-14.31	-14.92 – 73.93	0.84
Tracking (Radio-tracking)	-468.56	-468.97 – 58.23	**<0.001**
Sex (Male)	159.36	159.02 – 93.9	0.09
Random effects σ2/ICC	14727/0.153		
Marginal R^2/conditional R^2	0.407/0.498		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R^2 considers only the variance of the fixed effects, while the conditional R^2 takes both the fixed and random effects into account.

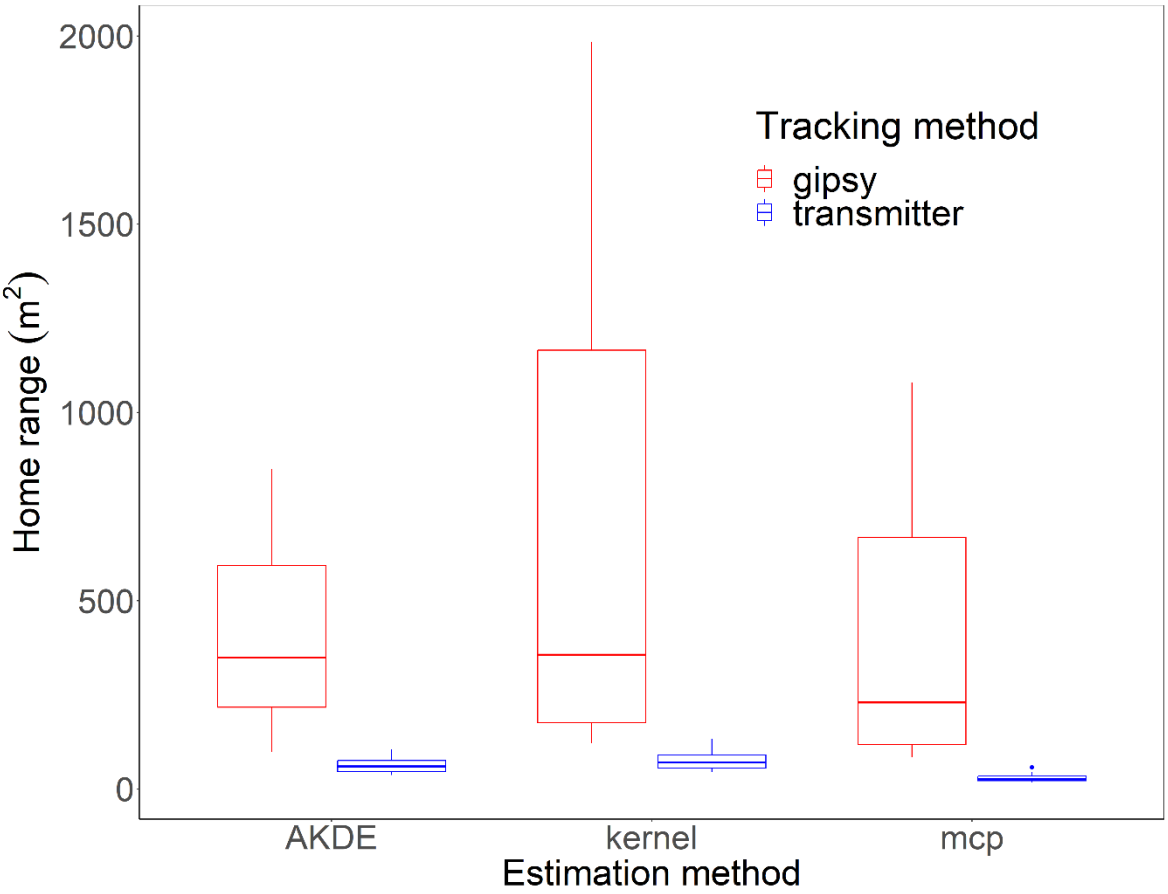

Figure 3 Estimated range sizes generated from mini-GPS (red bars) or radio-tracking (blue bars), calculated via different methods: autocorrelated kernel density estimation (AKDE); minimum convex polygons (MCP) 95%; Kernel density estimates (Kernel) 95%.

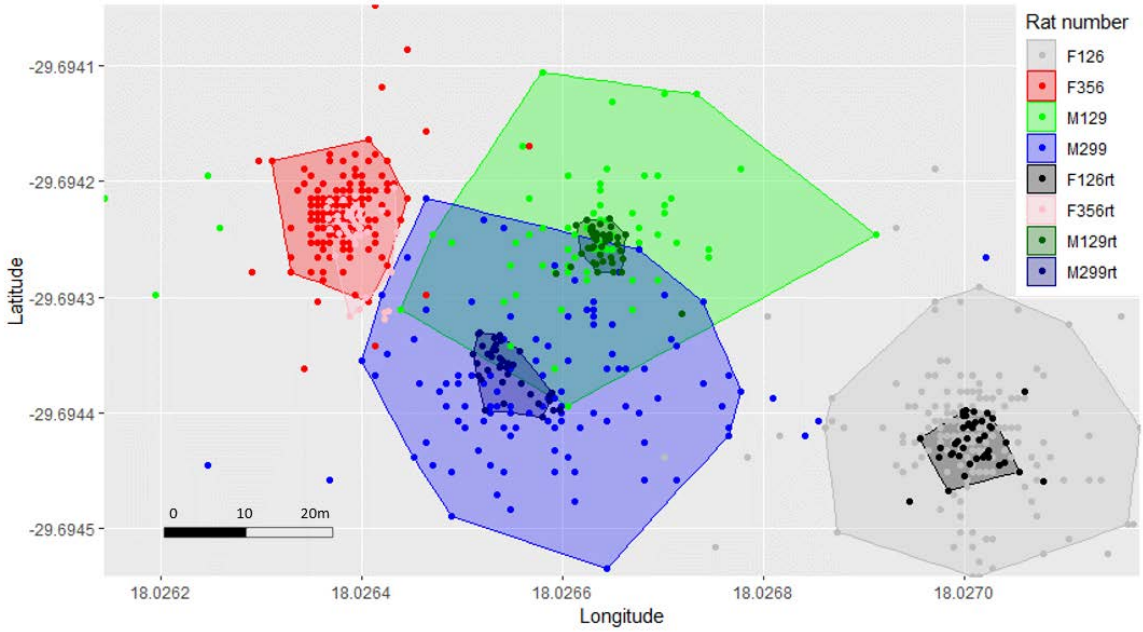

Figure 4. Daily ranges of four individuals (two males and two females) calculated using the MCP method for data obtained from mini-GPS (the large polygons) vs home range estimates from radio-tracking (the small polygons within). The coloured dots represent the different collection methods for the individuals and those outside the polygons represent the outliers excluded by the 95% method.

Discussion

Here we showed that mini-GPS dataloggers can provide reliable data on the range use of small rodents with a body mass of 100 g, even though the location error puts some limitations on the method. It is necessary to have reliable data of animal`s ranges if we want to understand their spatial structure (Powell, 2000). For decades, radio-tracking has been the most common technique to obtain such data, but this method is time consuming (Harris et al., 1990). Recently, in large mammals, radio-collars have been replaced by GPS dataloggers, but not in small mammals (Fujioka et al., 2020; Kays et al., 2015). We validated the use of min-GPS dataloggers in a small mammal and found range estimates to be larger and more realistic than those obtained from radio-tracking.

The main aim of our ongoing studies is to determine daily ranges using mini-GPS with a high frequency of fixes, and to do this simultaneously in neighbouring bush Karoo rats to determine whether they co-occur in space and time (studies to be completed in 2023/2024). Here we wanted to validate whether the ranges we obtain with mini-GPS are reliable or too small. For this we chose radio-tracking for comparison because it was used previously successfully on the same field site for another small rodent with the same activity rhythm (Schradin & Pillay, 2005). However, radio-tracking by homing in cannot be done every 5 minutes without permanent disturbance of the studied animals, we chose to determine home ranges instead. One shortcoming of our comparison is that the intervals at which fixes were taken differed between both methods. However, for both techniques we used the interval of fixes most appropriate for it, comparing two methods as actually used by field researchers. In a study focusing on home ranges (instead of daily ranges), it might be better to program the mini-GPS to collect fixes every 2 hours over several days (depending on battery size). If this method would yield smaller ranges than the daily ranges we obtained, this would indicate that our method is more valid. If it would yield larger ranges – which is to be expected – then this would strengthen our argument that mini-GPS provide larger and accurate estimates of (home) range sizes than does radio-tracking.

As predicted, the home range estimates obtained from radio-tracking were small. This was a result of the fixes obtained being concentrated at the individual's lodges, into which the rats withdrew when approached by the person radio-tracking. We rarely saw the rats during radio tracking (4 % of fixes), when they had withdrawn into their lodge, and which was in contrast to 40 % of sightings of striped mice on the same field site in a previous study (Schradin & Pillay, 2005). Rats had several lodges available in their home range but typically were always radio-tracked in the same lodge, which contrasts with the sympatric diurnal striped mouse, that during the day often rests away from its nesting site. In contrast to radio-tracking data,

the daily range estimates from the GPS dataloggers included the foraging grounds of rats which we knew from observations during our field work.

The range estimates using mini-GPS dataloggers were 5 to 10 times larger than those estimated from radio-tracking data. The range data obtained from bush Karoo rats via radio-tracking using the homing-in technique mainly provided us with the information where they are nesting, but not where they are foraging. Theoretically, it is possible to get more accurate radio-tracking data by not approaching the rats, but by using trigonometry from a larger distance of 30 - 50 m (the range of radio-transmitter signals), but this method has a larger error and with our rats having smaller ranges, this would lead to high inaccuracies. Automated radio-tracking with base stations (Eccard et al., 2022; Hoffmann et al., 2019; Kowalski et al., 2019; Schirmer et al., 2020; Schlägel et al., 2019) could be a good alternative, but it is work-intensive to establish such a system and to maintain it technically. For projects that need permanent data on range use and that have a larger research team working on it simultaneously, such automated radio-tracking might be the best solution.

Differences in range size estimates between radio-tracking and mini-GPS loggers were not due to differences in size of the devices nor the duration of data collection. Radio-transmitters were much smaller than mini-GPS loggers, and data were collected over seven days, and not only for one day as with the mini-GPS loggers. If rats would be more active carrying smaller devices and observed in more areas when monitored for a longer period, then this would suggest finding larger range sizes with radio-transmitters than with mini-GPS, but we found the opposite.

Every new method employed should be validated for accuracy. We found that the fixes obtained from the mini-GPS dataloggers matched those from the handheld GPS used for radio-tracking during a hike. Thus, we concluded that the larger ranges obtained from the mini-GPS dataloggers were not due to a lower accuracy of its fixes, as they were as accurate as the fixes from radio-tracking. We cleaned the mini-GPS data by excluding points with high inaccuracy. We excluded fixes with low estimated horizontal accuracy. While we included fixes which had a reported horizontal accuracy of < 50 m, our visualisation indicated that fixes where within a diameter of 10 - 20 m from each other (Fig. 4). By using the 95 % MCP, we were able to exclude obvious outliers and computed range estimates that included the areas in which we had observed the rats foraging. In sum, mini-GPS data had to be cleaned and by using 95 % MCP excluding obvious outliers, we obtained useable range data.

Another aspect that must be accounted for in studies using GPS dataloggers is the location error (Fleming et al., 2021; Thomson et al., 2017). Every GPS has some inaccuracy associated with it and the location error is the distance between the GPS-generated location and the true location (Frair et al., 2010; McMahon et

al., 2017 and references within). For animals with small ranges, the location error could be larger than the daily range, making it impossible to estimate it correctly. We accounted for the location error by calculating our range estimates using the *ctmm* package that not only takes calibrated data into account but also accounts for possible autocorrelation that may be associated with GPS data collection (Calabrese et al., 2016; Fleming et al., 2021; Fleming et al., 2015). When the location error is not accounted for, this may lead to improper conclusions on the range estimates of the animal under study (Fleming et al., 2021; McMahon et al., 2017). Traditional methods such as MCP and Kernel density do not account for biases in their calculations that have become inherent with the advances in technology, such as small sample sizes and autocorrelation. Furthermore, there has been no formal way to account for location error with these estimates before the introduction of certain mathematical models (such as *ctmm*). In sum, it was important to account for location error before estimating ranges using mini-GPS data (Fig. 5) to obtain reliable range data and doing so lead to more accurate estimates than radio-tracking (Fig. 3).

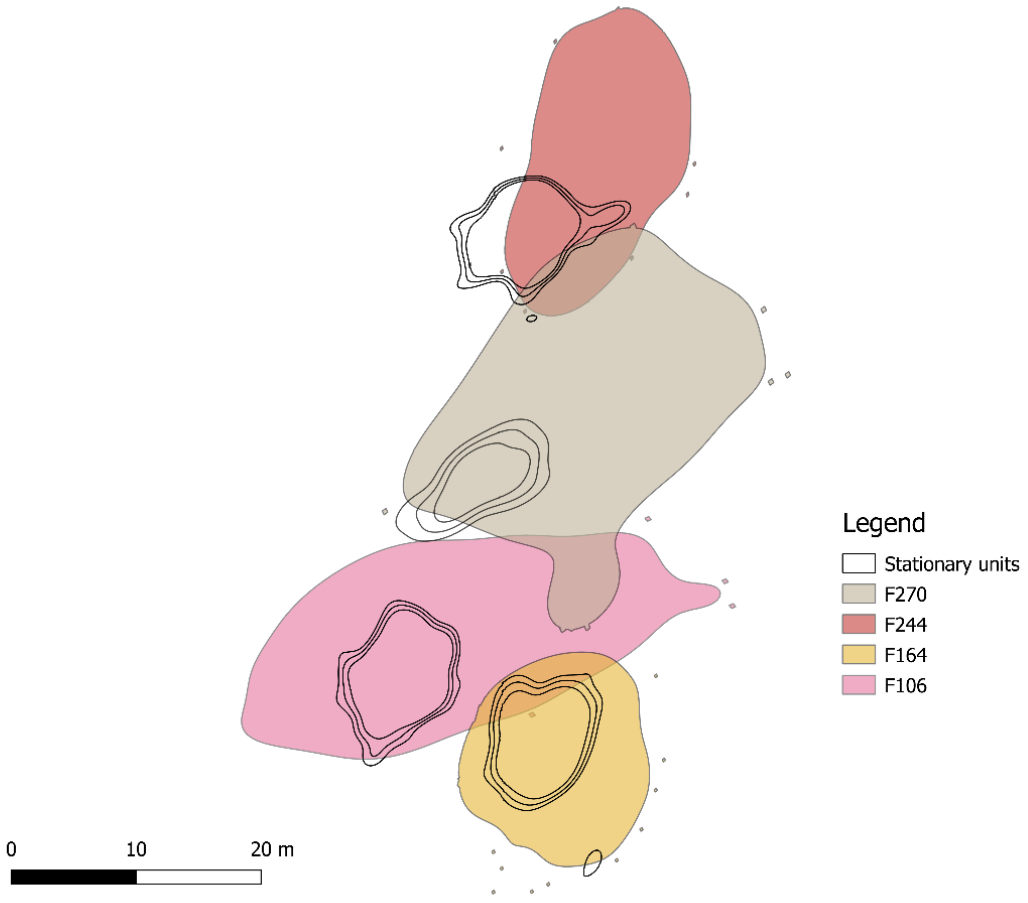

Figure 5. Daily ranges of four females calculated using the *ctmm* package method as autocorrelated Kernel density estimates correcting for the location error vs the stationary test units placed at the nests (the small lines within). This

diagram shows that while location errors are relatively large for these small daily ranges, our method allowed to calculate daily ranges that are clearly larger than the location error

Using radio-tracking to obtain range estimates needs a high time investment in collecting data over multiple days. Estimating ranges from mini-GPS datalogger data requires time for logger programming, value transformation, and data cleaning (about one hour per unit). For radio-tracking, we spent approximately 42 hours in the field to obtain data for three individuals (6x 1 hour every day, for 7 days) or 7 hours per individual, and one hour to download and organise the data. Mini-GPS are approximately twice the price of radio-collars (approx. 400 vs. 200 Euros per unit), but while radio-collars have to be sent in to replace batteries, the batteries of mini-GPS can be charged on location. Using mini-GPS dataloggers is a much lower time investment than radio-tracking but needs significant investment for validation.

Conclusions

Mini-GPS dataloggers have been successfully used in large mammals and in bats (Conenna et al., 2019; Corlatti et al., 2012; Vleut et al., 2019), but not in other small mammals below 100 g body mass. In contrast to bats, dataloggers used on rodents and Eulipotyphla need strong coating to protect them from gnawing. When using GPS dataloggers it is important to account for location error to obtain reliable range estimates, especially for small mammals with small ranges (McMahon et al., 2017). Here we showed that for a central place foraging rodent of less than 100 g, using mini-GPS dataloggers produce better range use data than radio-collars. Mini-GPS have been used previously in another central place forager, the golden-mantled ground squirrels (*Callospermophilus lateralis*) (Hefty & Stewart, 2019), but this species is nearly double as heavy as bush Karoo rats, and the data were not compared to radio-tracking. Future studies should test whether mini-GPS or radio-tracking would lead to more accurate range estimates in small mammals having foraging strategies different from central place foraging. Considering the progress in producing ever smaller GPS and the lower workload of using mini-GPS than radio-tracking, we encourage other researchers to test and validate whether this technology is also useful for small mammals with large home ranges and different foraging strategies.

Acknowledgements

We are thankful for comments by N. Pillay, M. Enstipp and C. Catoni that significantly improved the manuscript. C.H. Fleming gave important advice and help with analysing the data while taking the location error into account. This study was made possible by the administrative and technical support of the

Succulent Karoo Research Station (registered South African NPO 122-134). We are grateful for the support of Goegap Nature Reserve and its staff as well as the Northern Cape Department of Environment & Nature Conservation. This study was supported by the CNRS and the University of the Witwatersrand.

References

Andrusiak, L. A., & Harestad, A. S. (1989). Feeding behaviour and distance from burrows of Columbian ground squirrels. *Canadian journal of zoology, 67*(2), 381-384.

Bateman, A., Chambers, B., Pacioni, C., Rafferty, C., Jones, K., & Bencini, R. (2017). Application of underpasses to expand nature reserves: responses of a critically endangered marsupial, the woylie, *Bettongia penicillata*. [journal article]. *Hystrix, the Italian Journal of Mammalogy, 28*(2), 194-201. doi: 10.4404/hystrix-28.2-12255

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. . *Journal of Statistical Software, 67*(1), 1-48. doi: 10.18637/jss.v067.i01.

Börger, L., Dalziel, B. D., & Fryxell, J. M. (2008). Are there general mechanisms of animal home range behavior? A review and prospects for future research. *Ecol Let, 11*, 1-14.

Brown, E., & Willan, K. (1991b). Microhabitat selection and use by the bush Karoo rat *Otomys unisulcatus* in the Eastern Cape Province. *South African Journal of Wildlife Research, 21*, 69-75.

Burgin, C. J., Colella, J. P., Kahn, P. L., & Upham, N. S. (2018). How many species of mammals are there? *Journal of Mammalogy, 99*(1), 1-14. doi: 10.1093/jmammal/gyx147

Burt, W. H. (1943). Territoriality and home range concepts as applied to mammals. *J Mammal, 24*, 346-352.

Calabrese, J. M., Fleming, C. H., & Gurarie, E. (2016). *ctmm*: An r package for analyzing animal relocation data as a continuous-time stochastic process. *Methods in Ecology and Evolution, 7*(9), 1124-1132. doi: https://doi.org/10.1111/2041-210x.12559

Calenge, C. (2015). *adehabitatHR*: Home range estimation. *R package version 0.4.14*. doi: http://cran.r-project.org/web/packages/adehabitatHR/index.html

Conenna, I., López-Baucells, A., Rocha, R., Ripperger, S., & Cabeza, M. (2019). Movement seasonality in a desert-dwelling bat revealed by miniature GPS loggers. *Movement ecology, 7*(1), 1-10.

Corlatti, L., Béthaz, S., von Hardenberg, A., Bassano, B., Palme, R., & Lovari, S. (2012). Hormones, parasites and male mating tactics in Alpine chamois: identifying the mechanisms of life history trade-offs. *Animal Behaviour, 84*(4), 1061-1070.

De Angelis, D., Huber, D., Reljic, S., Ciucci, P., & Kusak, J. (2021). Factors affecting the home range of Dinaric-Pindos brown bears. *Journal of Mammalogy, 102*. doi: 10.1093/jmammal/gyab018

DeGregorio, B. A., Ravesi, M., Sperry, J. H., Tetzlaff, S. J., Josimovich, J., Matthews, M., & Kingsbury, B. A. (2018). Daily and seasonal activity patterns of the Massasauga (Sistrurus catenatus): an automated radio-telemetry study. *Herpetological Conservation and Biology, 13*(1), 10-16.

Dong, X., Fleming, C., Noonan, M., & Calabrese, J. (2018). Ctmmweb: A Shiny Web App for the Ctmm Movement Analysis Package.: https://github.com/ctmm-initiative/ctmmweb.

du P Bothma, J., & Le Riche, E. N. (1984). Aspects of the ecology and the behaviour of the leopard Panthera pardus in the Kalahari Desert. *Koedoe, 27*(2), 259-279.

du Plessis, A., & Kerley, G. I. H. (1991). Refuge strategies and habitat segregation in two sympatric rodents *Otomys unisulcatus* and *Parotomys brantsii*. *Journal of Zoology, London, 224*, 1-10.

du Plessis, A., Kerley, G. I. H., & Winter, P. E. D. (1992). Refuge microclimates of rodents: a surface nesting *Otomys unisulcatus* and a burrowing *Parotomys brantsii*. *Acta Theriologica, 37*, 351-358.

Eccard, J. A., Herde, A., Schuster, A. C., Liesenjohann, T., Knopp, T., Heckel, G., & Dammhahn, M. (2022). Fitness, risk taking, and spatial behavior covary with boldness in experimental vole populations. *Ecology and evolution, 12*(2), e8521.

Eccard, J. A., Meyer, J., & Sundell, J. (2004). Space use, circadian activity pattern, and matting system of the nocturnal tree rat Thallomys nigricauda. *Journal of Mammalogy, 85*(3), 440-445.

Edelman, A. J., & Koprowski, J. L. (2006). Seasonal changes in home ranges of Abert's squirrels: impact of mating season. *Can. J. Zool., 84*, 404-411.

Fleming, C. H., Dresher-Lehman, J., Noonan, M. J., Akre, T., LaPoint, S. C., Meyburg, B.-U., van Noordwijk, M. A., Pfeiffer, T., Roulin, A., Séchaud, R., Shephard, J. M., Stabach, J. A., Stein, K., Tonra, C. M., Vogel, E. R., & Calabrese, J. M. (2021). A comprehensive framework for handling location error in animal tracking data. *bioRxiv*. doi: https://doi.org/10.1101/2020.06.12.130195

Fleming, C. H., Fagan, W. F., Mueller, T., Olson, K. A., Leimgruber, P., & Calabrese, J. M. (2015). Rigorous home range estimation with movement data: A new autocorrelated kernel density estimator. *Ecology, 95*(5), 1182-1188. doi: https://doi.org/10.1890/14-2010.1

Frair, J. L., Fieberg, J., Hebblewhite, M., Cagnacci, F., DeCesare, N. J., & Pedrotti, L. (2010). Resolving issues of imprecise and habitat-biased locations in ecological analyses using GPS telemetry data. *Philosophical Transactions of the Royal Society B: Biological Sciences, 365*(1550), 2187-2200.

Fujioka, E., Yoshimura, K., Ujino, T., Yoda, K., Fukui, D., & Hiryu, S. (2020). Selection of foraging and commuting habitats of the greater horseshoe bat, revealed by high-resolution GPS-tracking.

Gardner, J., & Serena, M. (1995). Spatial-organization and movement patterns of adult male platypus, *Ornithorhynchus anatinus* (Monotremata, Ornithorhynchidae). *Australian Journal of Zoology, 43*(1), 91-103. doi: https://doi.org/10.1071/ZO9950091

Gazzard, A., Yarnell, R. W., & Baker, P. J. (2022). Fine-scale habitat selection of a small mammalian urban adapter: the West European hedgehog (Erinaceus europaeus). *Mammalian Biology, 102*(2), 387-403. doi: 10.1007/s42991-022-00251-5

Gunner, R. M., Wilson, R. P., Holton, M. D., Hopkins, P., Bell, S. H., Marks, N. J., Bennett, N. C., Ferreira, S., Govender, D., & Viljoen, P. (2022). Decision rules for determining terrestrial movement and the consequences for filtering high-resolution global positioning system tracks: a case study using the African lion (Panthera leo). *Journal of the Royal Society Interface, 19*(186), 20210692.

Harris, S., Cresswell, W., Forde, P., Trewhella, W., Woollard, T., & Wray, S. (1990). Home-range analysis using radio-tracking data–a review of problems and techniques particularly as applied to the study of mammals. *Mammal review, 20*(2-3), 97-123.

Hawkins, C. E., & Macdonald, D. W. (1992). A spool-and-line method for investigating the movements of badgers, Meies meles. *Mammalia, 56*(2), 322-326. doi: doi:10.1515/mamm-1992-0226

Hefty, K. L., & Stewart, K. M. (2019). Flexible resource use strategies of a central-place forager experiencing dynamic risk and opportunity. *Movement Ecology, 7*(1), 23. doi: 10.1186/s40462-019-0168-2

Hoffmann, J., Schirmer, A., & Eccard, J. A. (2019). Light pollution affects space use and interaction of two small mammal species irrespective of personality. *BMC ecology, 19*(1), 1-11.

Inoue, T. (1991). Sex differences in spatial distribution of the big-clawed shrew *Sorex unguiculatus.* [Article]. *Acta Theriologica, 36*(3-4), 229-237.

Jackson, T. (2001). Factors influencing food collection behaviour in Brants`whistling rat (*Parotomys brantsii*): a central place forager. *J Zool Lond, 255*, 15-23.

Kays, R., Crofoot, M. C., Jetz, W., & Wikelski, M. (2015). Terrestrial animal tracking as an eye on life and planet. *Science, 348*(6240), aaa2478.

Kenward, R. (1987). *Wildlife Radio Tagging*. London: Academic Press.

Kowalski, G. J., Grimm, V., Herde, A., Guenther, A., & Eccard, J. A. (2019). Does animal personality affect movement in habitat corridors? Experiments with common voles (Microtus arvalis) using different corridor widths. *Animals, 9*(6), 291.

Lehrer, E., Schooley, R. L., & Whittington, J. K. (2012). Survival and antipredator behavior of woodchucks (Marmota monax) along an urban–agricultural gradient. *Canadian Journal of Zoology, 90*(1), 12-21.

Lenth, R. V. (2021). emmeans: estimated marginal means, aka leastsquares means (Version 1.6.0).

Loy, A., Dupre, E., & Capanna, E. (1994). Territorial behavior in *Talpa romana*, a fosserial insectivore from south-central Italy. [Article]. *Journal of Mammalogy, 75*(2), 529-535. doi: 10.2307/1382577

Maynard Smith, J. (1974). The theory of games and the evolution of animal conflicts. *Journal of Theoretical Biology, 47*(1), 209-221. doi: 10.1016/0022-5193(74)90110-6

McMahon, L., Rachlow, J., Shipley, L., Forbey, J., Johnson, T., & Olsoy, P. (2017). Evaluation of micro-GPS receivers for tracking small-bodied mammals. *PLoS ONE, 12*(3). doi: https://doi.org/10.1371/journal.pone.0173185

Mohr, C. O. (1947). Table of Equivalent Populations of North American Small Mammals. *American Midland Naturalist, 37*, 223.

Murray, D. L., & Fuller, M. R. (2000). *Research techniques in animal ecology: Controversies and Consequences*. New York: Columbia University Press.

Ophir, A. G., Wolff, J. O., & Phelps, S. M. (2008). Variation in neural V1aR predicts sexual fidelity and space use among male prairie voles in semi-natural settings. [Article]. *Proceedings of the National Academy of Sciences of the United States of America, 105*(4), 1249-1254.

Orians, G. H., & Pearson, N. E. (1979). On the theory of central place foraging. . In D. J. Horn & R. D. Mitchell (Eds.), *Analysis of ecological systems* (pp. 155-177). Columbus: Ohio State University Press.

Owen-Smith, N., & Goodall, V. (2014). Coping with savanna seasonality: comparative daily activity patterns of African ungulates as revealed by GPS telemetry. *Journal of Zoology, 293*(3), 181-191. doi: 10.1111/jzo.12132

Posit team. (2022). RStudio: Integrated Development Environment for R. PBC, Boston, MA: Posit Software. Retrieved from http://www.posit.co/

Potts, J. R., & Lewis, M. A. (2014). How do animal territories form and change? Lessons from 20 years of mechanistic modelling. *Proceedings of the Royal Society B: Biological Sciences, 281*(1784). doi: 10.1098/rspb.2014.0231

Powell, R. A. (2000). Animal home ranges and territories and home range estimators. In L. Boitani & T. K. Fuller (Eds.), *Research techniques in animal ecology: controversies and consequences* (Vol. 442, pp. 65-110). United States of America: Columbia University Press.

QGIS, D. T. (2009). QGIS Geographic Information System.

R Core Team. (2022a). R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.

Schirmer, A., Hoffmann, J., Eccard, J. A., & Dammhahn, M. (2020). My niche: individual spatial niche specialization affects within-and between-species interactions. *Proceedings of the Royal Society B, 287*(1918), 20192211.

Schlägel, U. E., Signer, J., Herde, A., Eden, S., Jeltsch, F., Eccard, J. A., & Dammhahn, M. (2019). Estimating interactions between individuals from concurrent animal movements. *Methods in Ecology and Evolution*, *10*(8), 1234-1245.

Schneider, T. C., & Kappeler, P. M. (2016). Gregarious sexual segregation: the unusual social organization of the Malagasy narrow-striped mongoose (Mungotictis decemlineata). [journal article]. *Behavioral Ecology and Sociobiology*, *70*(6), 913-926. doi: 10.1007/s00265-016-2113-3

Schoepf, I., Schmohl, G., König, B., Pillay, N., & Schradin, C. (2015). Manipulation of population density and food availability affects home range sizes of African striped mouse females. *Animal Behaviour*, *99*(0), 53-60. doi: http://dx.doi.org/10.1016/j.anbehav.2014.10.002

Schradin, C. (2005). Nest-Site Competition in Two Diurnal Rodents from the Succulent Karoo of South Africa. *Journal of Mammalogy*, *86*(4), 757-762. doi: 10.1644/1545-1542(2005)086[0757:Ncitdr]2.0.Co;2

Schradin, C. (2006). Whole day follows of the striped mouse. *J Ethol*, *24*, 37-43.

Schradin, C., & Pillay, N. (2005). Intraspecific variation in the spatial and social organization of the African striped mouse. *Journal of Mammalogy*, *86*, 99-107.

Schradin, C., & Pillay, N. (2006). Female striped mice (*Rhabdomys pumilio*) change their home ranges in response to seasonal variation in food availability. *Behav Ecol*, *17*, 452-458.

Schradin, C., Schmohl, G., Rödel, H. G., Schoepf, I., Treffler, S. M., Brenner, J., Bleeker, M., Schubert, M., König, B., & Pillay, N. (2010b). Female home range size is regulated by resource distribution and intraspecific competition: a long-term field study. *Animal Behaviour*, *79*(1), 195-203.

Silva, I., Fleming, C. H., Noonan, M. J., Alston, J., Folta, C., Fagan, W. F., & Calabrese, J. M. (2022). Autocorrelation-informed home range estimation: A review and practical guide. . *Methods in Ecology and Evolution*, *13*, 534-544. doi: https://doi.org/10.1111/2041-210X.13786

Streicher, J. P., Ramesh, T., & Downs, C. T. (2020). Home range and core area utilisation of three co-existing mongoose species: large grey, water and white-tailed in the fragmented landscape of the KwaZulu-Natal Midlands, South Africa. *Mammalian Biology*, *100*(3), 273-283. doi: 10.1007/s42991-020-00028-8

Thomson, J., Borger, L., Christianen, M., Esteban, N., Laloe, J. O., & Hays, G. (2017). Implications of location accuracy and data volume for home range estimation and fine-scale movement analysis: comparing argos and fastloc-gps tracking data. . *Marine Biology*, *16*(204).

Vleut, I., Carter, G. G., & Medellín, R. A. (2019). Movement ecology of the carnivorous woolly false vampire bat (Chrotopterus auritus) in southern Mexico. *Plos one*, *14*(7), e0220504.

Walton, Z., & Mattisson, J. (2021). Down a hole: missing GPS positions reveal birth dates of an underground denning species, the red fox. *Mammalian Biology, 101*(3), 357-362. doi: 10.1007/s42991-020-00089-9

Ward, M. P., Sperry, J. H., & Weatherhead, P. J. (2013). Evaluation of automated radio telemetry for quantifying movements and home ranges of snakes. *Journal of Herpetology, 47*(2), 337-345.

Worton, B. J. (1989). Kernel Methods for Estimating the Utilization Distribution in Home-Range Studies. *70*, 164-168. doi: 10.2307/1938423

CHAPTER 4. Kin based spatial structure in a solitary small mammal as indicated by GPS dataloggers.

Makuya, L[1]., Pillay, N[1]., & Schradin, C[1,2]. (2024). Kin based spatial structure in a solitary small mammal as indicated by GPS dataloggers. *Animal Behaviour*, *215*, 45-54. doi: https://doi.org/10.1016/j.anbehav.2024.06.022

Authors, affiliations:

[1] School of Animal, Plant & Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS 2050, Johannesburg, South Africa

[2] IPHC, UNISTRA, CNRS, 23 rue du Loess, 67200 Strasbourg, France

Abstract

Kin selection is important for understanding the evolution of social behaviour in group-living species. Yet, the role of kinship in solitary species has received little attention. We studied how kinship influences intra-specific variation in social organisation and spatial structure in a predominantly solitary species, the bush Karoo rat (*Otomys unisulcatus*) from the Succulent Karoo semi-desert of South Africa. We predicted that if social groups occur, they would consist of close kin. We further predicted that the spatial structure is not random, but that close kin live closer to each other. Over five years we performed trapping and focal animal observations and fitted mini-GPS dataloggers simultaneously on neighbouring female bush Karoo rats (N = 125) to investigate how their spatial structure was influenced by kinship. Female bush Karoo rats were mainly solitary living, although small social groups also occurred, all consisting of close kin, typically females, such as a mother and her adult daughter or sisters. Although females did have more non-kin than kin neighbours, kin lived closer to each other than non-kin. Daily ranges were larger in the breeding than in the non-breeding season and overlapped more between kin than non-kin females. We conclude that kinship should be considered when studying solitary living species as it might influence variation in social organisation and spatial structure.

Keywords: intra-specific variation in social organisation; kinship; kin selection; social organisation; social structure; social system; solitary; spatial structure

Introduction

Kin selection (Hamilton, 1964a) has been used to explain why species live in groups (Lacey & Wieczorek, 2004) but has rarely been considered of importance for solitary species. Kin groups often form when individuals delay dispersal and remain philopatric (Williams & Rabenold, 2005). But dispersal can also influence spatial kin structure in solitary species (Cutrera et al., 2005).

A species' spatial structure forms the basis on which their social system (i.e., social organization, social structure, care and mating systems) develops and evolves (He et al., 2019; Kappeler, 2019; Webber et al., 2023; Webber & Vander Wal, 2018). The spatial structure of a species is influenced by different factors such as food availability, the presence of conspecifics, geographic barriers and dispersal (Schlichting et al., 2022). Of these factors, dispersal is one of the most important ones (Cooper & Randall, 2007; Cutrera et al., 2005; He et al., 2019; Webber et al., 2023). In mammals, females are often philopatric while males disperse (Li & Kokko, 2019; Williams & Rabenold, 2005). If females disperse, then it is often for shorter distances from their natal territory, whereas males disperse over larger distances (Lawson Handley & Perrin, 2007a). In the solitary giant kangaroo rat (*Dipodomys ingens*), the spatial structure is characterized by female kin living closely together, especially at high population densities (Meshriy et al., 2011). While philopatry is considered a promotor of group living, dispersal in its broad sense (i.e. remaining in the natal area) can also be important to understand the social systems of solitary species (Cooper & Randall, 2007).

In mammals, solitary species are characterised by males and females only meeting for courtship and mating, and offspring leaving their mother in the period between weaning and the end of puberty (Makuya & Schradin, 2024a). Examples of solitary species are some shrews (Valomy et al., 2015a), and large predators, such as black bears (*Ursus americanus*) (Kovach & Powell, 2003) and leopards (*Panthera pardus*) (Rafiq et al., 2020). Solitary living is often assumed to be an adaptation to sparsely distributed food that cannot be shared (Dalerum, 2007; Makuya & Schradin, 2024a; Nowak & Wilson, 1999). While individuals of solitary species do not share the same home range and sleeping sites, they do interact with conspecifics (Makuya & Schradin, 2024a). For example, the solitary puma (*Puma concolor*) has complex social networks with individualized relationships (Elbroch & Wittmer, 2012). In the solitary giant kangaroo rat (*Dipodomys ingens*), the spatial structure is characterized by female kin living closely together, especially at high population densities (Meshriy et al., 2011). Many solitary species do not have a random distribution but display a kin-based spatial structure, though this has been rarely studied.

The spatial structure of solitary living species can change by season (Johansson et al., 2018). In particular, males of solitary species increase their home ranges more than females during the breeding season to

overlap with many females to increase their reproductive success (Dillon & Kelly, 2008). Females might change their home ranges seasonally as a response to changes in resource availability (Potts & Lewis, 2014). For example, female striped mice show variation in their home range sizes depending on the seasonal availability of food (Schradin et al., 2010b). The impact of season and sex on home range size is well understood in group-living species (Schradin et al., 2010b; Stamps, 1994) but not much is known about solitary species. Studying variation in home range size of solitary species will provide an indication of intraspecific variation in social organization (IVSO) in solitary species, particularly among the females.

Some species do not have a fixed but varying social organization (Schradin, 2013). Factors that influence such intraspecific variation in social organization (IVSO) include population density and reproductive competition (Schradin et al., 2018a). The African striped mouse (*Rhabdomys pumilio*) shows IVSO, being able to switch from solitary living at low population density to group living when both population density increases and reproductive competition decreases (Schoepf & Schradin, 2012a; Schradin et al., 2010a). Generally, population density and competition are the main drivers of IVSO at the species level (Schradin, 2013). IVSO may be adaptive in harsh environments (Makuya et al., 2023) and also occurs in species that are typically regarded as solitary living (Valomy et al., 2015a). To date we don't know whether population density and reproductive competition might lead to IVSO in predominantly solitary species, leading to some of them forming groups when population density is very high.

Solitary living mammals are often either small, nocturnal and live in dense habitats (shrews, rodents), or large and nocturnal, occupying large areas (large predators), making them challenging to study (Kovach & Powell, 2003; Waser & Jones, 1983). The solitary bush Karoo rat (*Otomys unisulcatus*) from the Succulent Karoo semi-desert is a good model to study solitary living since it is a diurnal small mammal that occupies an open habitat characterized by low growing annuals and dispersed shrubs, has small home ranges and is easily habituated to the presence of observers (Schradin, 2005; Vermeulen & Nel, 1988; Wolhuter et al., 2022).

We studied how kinship influences the social organization, especially deviation from its primarily solitary living, and spatial distribution of the bush Karoo rat. We focussed on females, which is the more philopatric sex. We ascertained the social organization of the bush Karoo rat from trapping data and behavioural observations and made five predictions. 1) Bush Karoo rats are mainly solitary living, but when groups occur, these will be mainly female kin groups, occurring after the breeding season and when population density is high. 2) Females have more female kin than non-kin neighbours. 3) The geographic distance between female kin neighbours is shorter than between non-kin neighbours. We then used mini-GPS data-loggers to provide a better assessment of the spatial structure and ascertain the daily range sizes and overlap of daily ranges, and how these are influenced by kinship and season. We predicted 4) that the daily ranges

of females vary seasonally, being larger in the food restricted dry season, and 5) the daily ranges of kin neighbours overlap more than that of non-kin.

Methods

Study species

The 100 g bush Karoo rat (*Otomys unisulcatus*) is endemic to semi-arid and arid regions of South Africa, particularly the Karoo and the Succulent Karoo, one of the world's most important biodiversity hotpots (Cowling et al., 1999; Myers et al., 2000). The bush Karoo rat builds stick-lodges inside shrubs, which provide a favourable micro-climate with high humidity and mild temperatures for protection against the external environment (Brown & Willan, 1991b; du Plessis & Kerley, 1991; du Plessis et al., 1992). It is a central-place forager, foraging within a short distance (less than 25m) from its lodge and then returning to the lodge (Schradin, 2005).

Study site and study period

We conducted our study on a 3.5-hectare site next to the Succulent Karoo Research Station in the Goegap Nature Reserve, Northern Cape, South Africa. The Succulent Karoo is characterized by cold winters (mean temperatures of 16.8°C in June 2019 at our field site with a minimum of -2,6°C on one night) and hot summers (mean temperature in January 2019 was 27.3°C, maximum was 46.7°C). The study site occurs in a winter rainfall area with average rainfall of 160 mm per year at our field site (Schradin, 2005). In our study area, the breeding season of bush Karoo rats is from June until November (austral winter and spring), and the non-breeding season is from December to May (austral summer and autumn). We used trapping and observation data collected from bush Karoo rats from September 2017 to March 2020 (closure of field site due to the COVID-19 pandemic) and January 2021 to July 2023.

Sampling regime

Trapping and marking of bush Karoo rats

Trapping was conducted at lodges that showed signs of being occupied (fresh faeces, active runways, rats observed). The field site was divided into four to six areas, with one to two areas trapped at the same time by two teams (Figure 1). All lodges within one area were trapped for three consecutive days before moving

on to the next area. Traps were set in the morning before sunrise and checked after 45 and 90 minutes and then un-set during the hottest times of the day. In the afternoon, traps were set 45 minutes before sundown, checked after sundown, and then un-set during the night. We used a combination of foldable Sherman traps (https://shermantraps.com/) and locally produced heavy metal traps of the Sherman style. Traps were baited with a combination of bran flakes, salt and sunflower oil and rebaited each morning and afternoon. Traps were arranged around the entrances of the lodges and along runways. No injuries due to trapping were recorded. The details of trapped rats were recorded, including their weight to the nearest 0.1-gram, reproductive status and lodge number. Bush Karoo rats were removed from the traps in a plastic bag and weighed with the bag, before being taken out for inspection and then released at their lodge. Then the weight of the plastic bag was subtracted from the total weight. We marked individuals with metal band ear tags with a unique reference number for later identification (National Band and Tag Co., Newport, KY, U.S.A.) (Schoepf & Schradin, 2012a). To aid in visual identification during observations, individuals were marked with non-toxic hair dye (Inecto Rapido, Pinetown, South Africa), in combinations (females: head and chest/sides/back; males: hindquarters and chest/sides/back). Age was estimated from body mass at first capture, using a species-specific growth curve validated by our field data (Pillay, 2001). The bush Karoo rats were classified according to their age, with pups being up to two weeks old and weighing less than 30 g (weaning is at 14 days; (Pillay, 2001)), juveniles being 2-6 weeks old and weighing between 30 and 70 g, and adults being older than six weeks and weighing more than 70 g, when both sexes can start reproduction (Pillay, 2001).

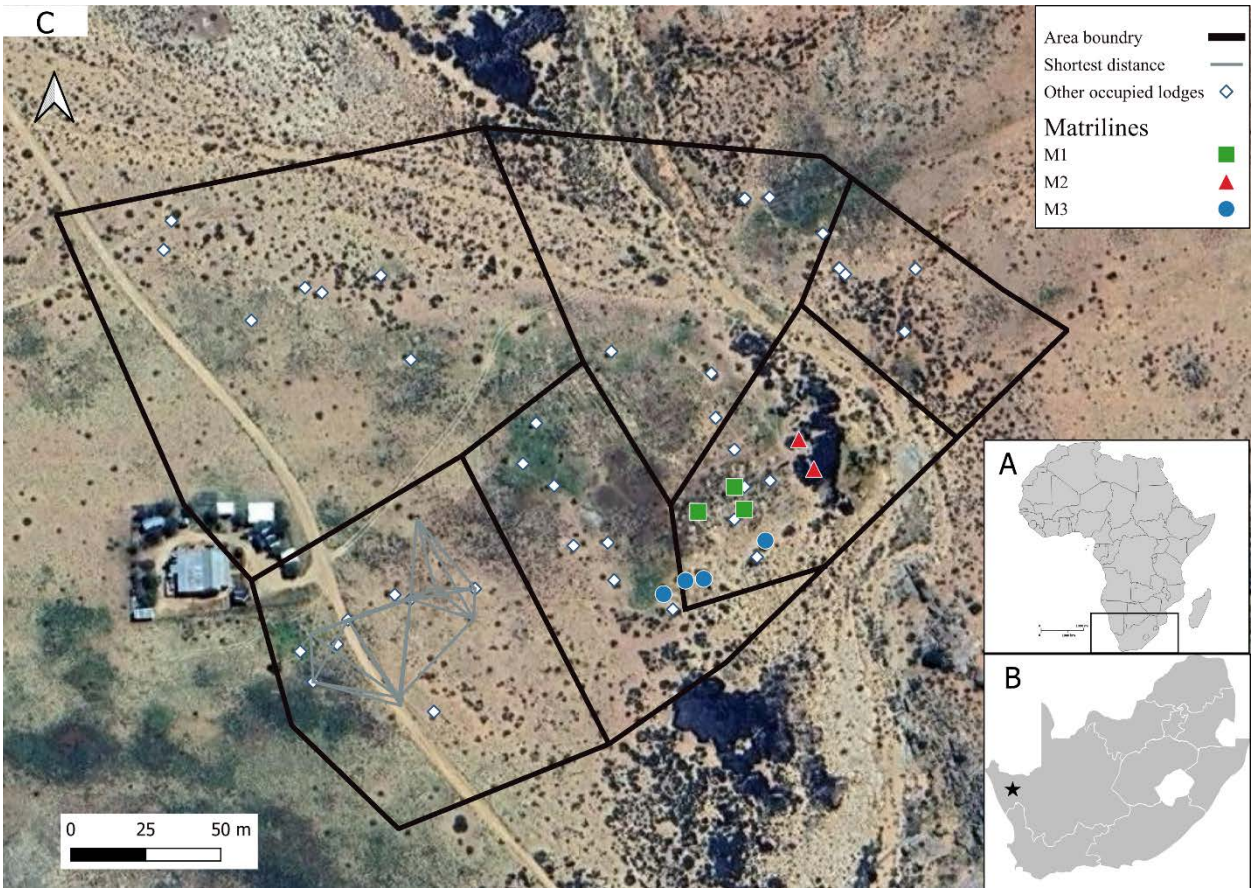

Figure 1. Satellite image of the field site (star in insert map). The black polygons are the boundaries of the six trapping areas within the field-site that we demarcated for trapping purposes. In one trapping area we show an example of matrilines in part of the study site for August 2022. Symbols and colours represent lodges that were occupied by bush Karoo rats of three different matrilines (white: rats from other matrilines). In another trapping area the grey lines represent an example of the shortest distance between the occupied lodges calculated in qGIS.

Determination of kinship

Relatedness between individuals was assessed from trapping records and observations. Pups and juveniles that were trapped at a lodge with an adult female were considered as the offspring of that female. Pups and juveniles were always trapped at only one lodge, such that our combination of trapping and observations allowed us to reliably estimate female kinship for them, although we have no information on paternal kinship. Following dispersal, juveniles either lived alone or with a sibling. This was confirmed through focal animal observations which we conducted at the lodges where pups and juveniles were located. Females from one matriline were regarded to be closely related kin: mother and her offspring, maternal siblings, females which had the same grandmother as well as maternal cousins, i.e., females whose mothers were sisters. All other individuals were considered non-kin.

Determination of reproduction

The reproductive condition of the females was determined from trapping data when pups / juveniles were trapped at the adult female's lodge. The month of birth of the pups / juveniles (estimated from their body mass at first trapping) was used as the reproductive month for the adult female. The gestation period of the bush Karoo rat is 39 days (Pillay, 2001). It was possible to tell when females were pregnant from their weight and when they gave birth due to a sudden drop in weight. We used this information to confirm the calculated pup/juvenile date of birth and therefore the breeding period of the females.

Behavioural observations in the field

Focal observations were conducted five times a week at lodges with evidence of being occupied by bush Karoo rats to determine occupancy of lodges (which individuals were present) and to identify when pups emerged, and juveniles / young adults dispersed. The morning observations started five minutes before the sun started shining on the lodges and lasted for 30 minutes; afternoon observations started 25 minutes before the sun stopped shining on the lodges. Lodges were usually observed on two days a month. The lodges that contained juveniles or a pregnant female were monitored more than twice in a month. The behaviours recorded during the observations included activities (e.g., foraging and feeding), lodge building and social behaviours including the following groups of behaviour: (i) amicable behaviours (i.e., grooming, body contact); (ii) social investigation (i.e., sniffing); and (iii) aggression (i.e., chasing, fighting). Behavioural data will be presented in another publication.

Determination of social organization

We used a combination of trapping and behavioural observations to assess the social organization (i.e., group size and composition) of each adult bush Karoo rat for every month they were observed in the study site. We identified four forms of social organizations (Table 1). Because lodges occupied by bush Karoo rats showed clear signs of use such as collected plant material, active runways and fresh faeces, we were certain that we had trapped at all occupied lodges on the field site (bush Karo rats on our field site always live in a lodge). The open habitat (no trees, low growing shrubs) ensured we did not miss a lodge and we continuously surveyed the field site for occupied lodges. If we did not trap bush Karoo rat at a lodge with an indication of occupancy within the normal trapping regime of three days, we continued trapping and also observed the lodge, to ensure that trap shy individuals were trapped and marked. Our behavioural

observations confirmed that bush Karoo rats trapped at a lodge also inhabited that lodge, although some bush Karoo rats inhabited several neighbouring lodges. Apart from roaming males, we very rarely observed any bush Karoo rats at a lodge that it did not inhabit.

Table 1. Observed forms of social organization in bush Karoo rats.

Social organization	**Definition**
Solitary living	Only one adult bush Karoo rat was trapped and observed at a lodge. We differentiated between solitary males and solitary females. Mother families, i.e., one mother with one or more offspring that were not adult were also considered to be solitary living.
Sibling pairs	Adult sister and adult brother.
Female groups	Two or more adult females were trapped and observed at one lodge together.
Mother and adult son	Adult female with her adult son

Pups: less than 2 weeks old; juveniles: 2-6 weeks; adults: > 6 weeks old.

We used a linear mixed effects model (LME) run in the *lme4 R* package using the *lmer* function (Bates et al., 2015) to analyse factors that influence the percentage of solitary living adult females. We used data collected from September 2017 to July 2023 (54 months), with season and population density as fixed effects and the year as a random variable. The population density was calculated every month ($N = 54$) from the total number of adult rats divided by the study site size. The model used was as follows:

percentage of solitary individuals = season + population density + (1|year).

Monthly determination of neighbours and distance between neighbours

We ascertained the number of adult kin and adult non-kin neighbours for each adult female every month. A neighbour was defined as any adult individual that occupied a lodge not more than 25 metres away from the focal individual with no other individual occupying a lodge in between. This distance was selected since we never observed a rat foraging more than 25 metres away from its focal lodge, which is confirmed by data from GPS dataloggers (Makuya & Schradin, 2023). For each female, we recorded the distance from its lodge to the lodges of its nearest kin and its closest non-kin neighbour in QGIS using the "shortest distance feature" tool (QGIS, 2009) (Figure 1). We ran a generalized linear mixed effects model (GLMM) with a Poisson distribution to analyse factors influencing the number of neighbours, with the focal rat's ID number as a random effect and the relationship between neighbours (kin VS non-kin) and season as fixed effects. We had a sample size of 172 focal rats over 21 months (from January 2021 to July 2023) in the following model:

Number of neighbours = relationship (kin VS non-kin) + season (breeding VS non-breeding) + (1|Focal ID) + (1|year).

We ran a linear mixed effects model to test for a difference in the distance between the closest kin versus non-kin neighbour. For every female, we took the mean distance to the kin neighbours and the mean distance to the non-kin neighbours such that each focal rat had a maximum of two neighbour values per month for the analyses. This was done to control for pseudo-replication. We included relationship (kin VS non-kin) and season as fixed effects and the ID of the focal rat and the year as random effects:

Distance = relationship (kin VS non-kin) + season (breeding VS non-breeding) + (1|Focal ID) + (1|Month)

Mini GPS dataloggers

Three to five neighbouring adult females were fitted with mini-GPS dataloggers (Gipsy 6 model from Technosmart, Italy https://www.technosmart.eu/gipsy-remote-copy/) on the same day, from August 2021 to April 2023. The mini-GPS weighed 4.5 g including the protective coating, and the rats weighed 96.6 + 19.3 g (range: 67.5-131.2 g) Therefore, the mini-GPS represented less than 7 % and in most cases less than 5 % of their body weight (Murray & Fuller, 2000). GPS dataloggers were fitted like a rucksack with a neck and a belly belt consisting of a wire running through a small rubber tube to avoid any skin irritation, fastened with a fishing crimp. The procedure lasted 1-3 minutes and involved holding the rat in one hand and fitting the unit. We fitted 227 Gipsys on a total of 125 females. Of these, useful data were obtained from 156 Gipsys; the remaining units (N = 73) either failed (did not switch on), were lost or switched off too early. Gipsy collars collected the location (longitude, latitude and altitude) of an individual and the date, time, and daily ranges used, including the occupied lodges and foraging grounds (Makuya & Schradin, 2023). The Gipsys were programmed following a successfully validated protocol (Makuya & Schradin, 2023). Data collection was delayed by 2 days after fitting the collar to allow the rats to become accustomed to the Gipsys. On the third day, the Gipsys collected GPS fixes every 5 minutes; the battery life did not allow us to collect data for more than one day. On day 4, we started to re-trap rats to remove the Gipsys. Because range estimates can be skewed due to outliers and locations with low accuracy, we cleaned the data from the mini-GPS loggers by first removing fixes with a horizontal accuracy of > 50 metres and then visualising fixes obtained from the Gipsys in QGIS to exclude obvious outliers, as validated in Makuya & Schradin (2023). To analyse the daily range sizes and overlap, we used the autocorrelated Kernel density estimation (AKDE) implemented in the R packages *ctmm* (Calabrese et al., 2016) and *ctmmweb* (Dong et al., 2018). The overlap was calculated for every pair on the daily range data from the same month. The AKDE approach allowed us to account for location error in the calculations of range sizes within the *ctmm*

workflow through two stages (Fleming et al., 2021). The calculations from the autocorrelation and bandwidth were error-informed, which mitigates various biases in autocorrelation and bandwidth estimates that would otherwise occur had location error been ignored. Then, location estimates were fed through a Kalman smoother before kernel placement, which counteracts overdispersal (Silva et al., 2022). To determine the overlap between a pair of home range estimates, we used the Bhatttacharyya coefficient within the *ctmmweb* R package, following Winner et al. (2018). We used a linear mixed effects model to determine what factors influenced the daily range sizes (DRS), in which season and relationship (kin versus non-kin) were used as fixed predictors and the ID of the rat was a random effect. This was followed by a linear mixed effects model to assess the influence of the season and relationship on the overall overlap between ranges. The models we used were:

$DRS = season + (1|ID)$

$Overlap = season + relationship + (1|ID)$

Statistical analysis

All statistical analyses were done in R (version 4.3.1; R Core Team, 2022b). Model assumptions were assessed by inspecting Q–Q plots and by plotting model residuals against fitted values for every model used. Before running each model, we checked for multi-collinearity in models that contained more than one fixed factor by calculating variance inflation factors (vifs) (Zuur et al., 2010) for the predictor variables using the *vif* function in the *car* package (Fox & Weisberg, 2011). Models that contained vifs above two were removed to prevent multi-collinearity. The study focussed on social organization, which by definition is the adult composition of social units, with pups and juveniles not considered (Kappeler, 2019). First, we describe social organization considering both sexes. Since males disperse early and then float (have no permanent lodge) and are only present for a few days at the field site before disappearing for weeks, the social units on the field site were characterised by females and all further statistical tests therefore involved females only.

Ethical note

We adhered to the ASAB/ABS Guidelines for the Use of Animals in Research (Buchanan et al., 2012). Bush Karoo rats (N = 507 rats trapped 10711 times over the study period) were captured and handled using protocols approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (AESC clearance numbers 2003/34/3 and 2018-03-15B). The field site was selected such that even under

low population density conditions, at least 8 solitary females were present, and regular trapping was necessary to ensure we kept track of all individuals on the field site. To reduce the stress caused to the animals after fitting them with mini-GPS dataloggers, we placed the rats in cages with food in a field laboratory and monitored them for one hour before releasing them into the field. Following their release, we also conducted focal animal observations on the rats that carried loggers and found that they foraged like normal, and the loggers did not seem to negatively affect them. We adhered to the relevant South African laws concerning the use of animals for experimental purposes, including obtaining Section 20 permit.

Results

Social organization

Most adult female and adult male bush Karoo rats were solitary living (mean ± SD % solitary living: females: 96.6 ± 10.26 %, males: 98.9 ± 2.97 %; Fig. 2). Adult groups made up a small percentage of the population and consisted of adult female kin (mother and adult daughter or sisters, 6.6 ± 9.5 % of all social units), mother and adult son (0.32 ± 0.93 %) and sibling pairs (brother and sister; 0.13 ± 0.66 %; Fig. 2). In total, we observed 97 groups in 23 of the 51 months of study, with a group size of two or three individuals (2.2 ± 0.397). Of these, 50 groups consisted of a mother and one (N =31 groups) or two daughters (N = 19 groups) and 31 of two (N = 28 groups) or three sisters (N = 3 groups). We observed 12 groups consisting of a mother and her adult son and four groups of a sister and brother. For the groups consisting of two or three females, none of the females bred in 65 groups (end of the breeding season), one bred in seven groups and both females bred in five groups. There were no significant effects of season (LME; t_{46} = 0.4, P = 0.69; Table A1) or population density on the percentage of solitary living females (LME; t_{48} = -1.6, P = 0.116; Table A1).

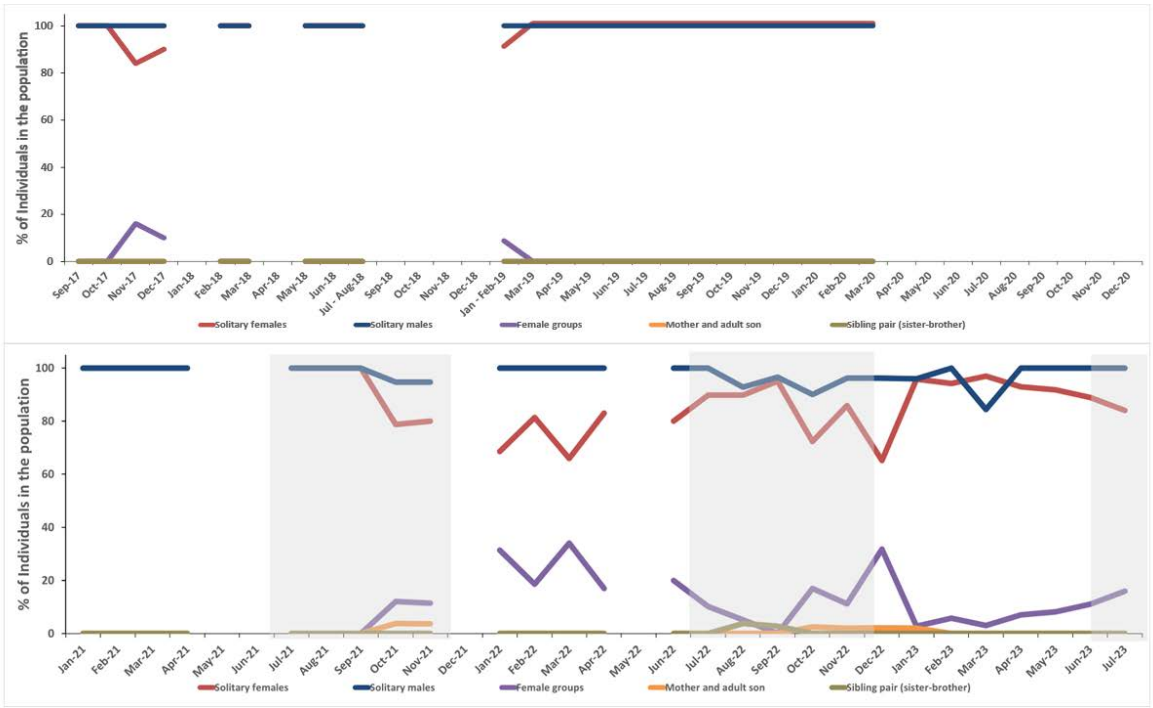

Figure 2. Distribution of social units in bush Karoo rats in seven years. The top panel is from September 2017 – December 2020, and the bottom panel is from January 2021 – July 2023. The breaks in the lines are from periods when no data was collected, for example due to the Corona lockdown (March to December 2020). The definitions for the different social units are given in Table 1. The grey boxes indicate the wet, breeding season in winter / spring. The number next to the respective month refers to the year (i.e. Sept-17 = September 2017).

Spatial structure

The mean values of kin and non-kin neighbours were very similar (Fig. 3(a), but female bush Karoo rats had more non-kin than kin neighbours (GLMM; $t = 2.53$, $P = 0.01$); Table A2). However, kin neighbours lived closer to each other than non-kin neighbours (LME; $t_{966} = 21.36$, $P < 0.001$) (Fig. 2 and 3(b); Table A3). Season did not influence the number of neighbours (GLMM; $t = 1.01$, $P = 0.29$), or the distance to the nearest neighbour (LME; $t_{11} = -0.882$, $P = 0.40$) (Table A2 and A3).

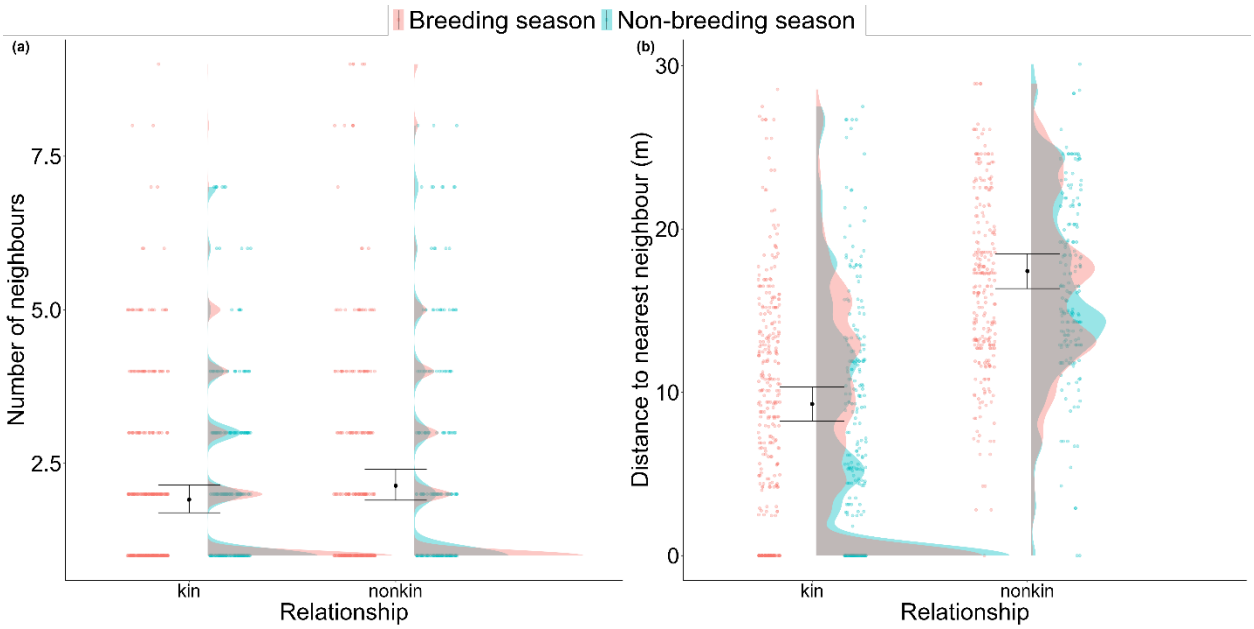

Figure 3. The number of female neighbours by kinship (a left; P < 0.01) and the mean distance to the nearest neighbour depending on kinship (b right; *P* < 0.001) in bush Karoo rats, for both seasons. points represent individual values (kin: *N* = 556, non-kin: *N* = 444). The black point is the model estimate and the error bars are the confidence intervals of the model. The half-violin plot shows the raw data distribution.

Daily range size and overlap

The daily range sizes were influenced by season (LME; t_{62} = 3.04, *P* < 0.001; Fig. 4(a)), with range sizes being significantly larger in the breeding season. Overlap of ranges was not influenced by season (LME; t_{104} = -0.99, *P* =0.32; Fig. 4(b)). However, it was influenced by kinship (LME; t_{97} = -4.57, *P* < 0.001; Table A5), with females overlapping significantly more with kin than with non-kin (Fig. 4(b); Table A5).

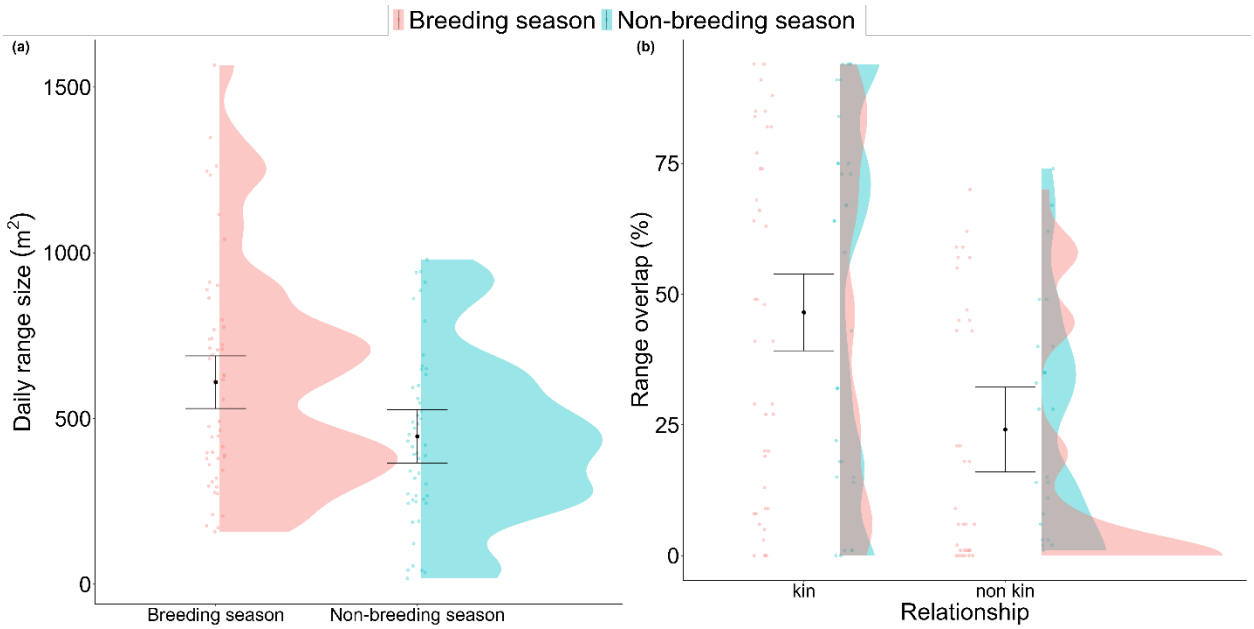

Figure 4. Seasonal comparison of daily range size (a left; non-breeding: N = 59, breeding: N = 81) and overlap (right; non-kin: *N* = 54, kin: *N* = 72) in bush Karoo rats. The black point is the model estimate and the error bars are the confidence intervals of the model. The half-violin plot shows the raw data distribution.

Discussion

We showed that kinship is important in the social system of a solitary species, suggesting the potential for kin selection. Specifically, we found that the spatial distribution of the solitary bush Karoo rat is not random but follows kin-based matrilines. Females live closer with and overlap more with female kin than with non-kin. We also found variation in social organization, with few small groups, which always consisted of close kin and most often closely related females. Our study adds to more recent studies indicating that solitary living is not primitive and simple and that solitary species might have more complex social structures than recognised (Baker et al., 2021; Elbroch et al., 2017a; Makuya & Schradin, 2024a, 2024b; Meshriy et al., 2011; Roex et al., 2022).

Bush Karoo rats are mainly solitary living but do show some deviation, leading to intraspecific variation in social organization. Ours is the first peer-reviewed study describing the social organization of bush Karoo rats. Unpublished data indicate that in other populations, group-living in the bush Karoo rat might be more common than in our study population (Do Linh San et al., 2016), indicating the possibility for variation in social organization within and between populations.

At our field site, groups rarely formed. These were always kin groups, as predicted, most often consisting of a mother and her adult daughter(s), and sometimes sisters. Males were more often solitary than females,

and few of them lived together with their mother or sister. In contrast to our prediction, groups were not more often found during the non-breeding season than the breeding season, even though the data indicate that they might be more common at the end of the breeding season (Fig. 2), which warrants future studies. In striped mice studied on the same field site, one main factor leading to solitary living is reproductive competition, through female infanticide of another female's pups (Schradin et al., 2010a). In the few bush Karoo rat groups, we observed either no reproduction taking place or only one female reproduced, such that reproduction competition played no role. Group formation in bush Karoo rats could best be explained by the delayed dispersal of females reaching adulthood during the end of the breeding season, which should be studied in more detail in the future.

Population density was not significantly related to solitary living as we had predicted. In our current study, sufficient free territories were available for most female bush Karoo rats to disperse and start solitary living. The small territories of these central place foragers, as seen from the daily range data, might be one explanation for the lack of association between population density and solitary living. In contrast, the sympatric striped mice have larger home ranges and are more likely to form groups when population density is high (Schoepf & Schradin, 2012a; Schradin et al., 2010a; Schradin et al., 2019). The striped mice study generated more data collected over several more years than we collected in our current study (8 years vs 5 years). Future analyses on a larger data set for bush Karoo rats over more years considering population density and additional co-factors such as predation pressure, food availability, and availability of lodges might help us to understand when groups form. It is possible that during years with very high population density, we might find more and even larger groups.

Non-random spatial structures of amicably interacting individuals have been described for crocodiles (Baker et al., 2021; Baker et al., 2023) and sharks (Anderson et al., 2021; Mourier et al., 2012). In solitary felids and rodents this is known to depend on kinship (Meshriy et al., 2011; Roex et al., 2022). In our study, female bush Karoo rats had significantly more non-kin than kin neighbours, though the mean values were very similar (Fig. 3a). This might simply be due to the fact that overall, a female had many more non-kin than kin in the population. However, female bush Karoo rats lived closer to kin than to non-kin neighbours. This spatial kinship structure is similar to that of the giant kangaroo rat (*Dipomys ingens*) which formed female kin groups with increasing population density (Meshriy et al., 2011) and woodchucks (*Marmota monax*), which lived closer to kin and shared home ranges (Maher, 2009). The spatial kinship pattern in the bush Karoo rat seems to be related to females dispersing from their natal lodge to unoccupied lodges in close proximity. There are fitness benefits of living near kin and familiar neighbours (Siracusa et al., 2021). An individual might be able to increase its inclusive fitness by associating and collaborating with closely related conspecifics, even in solitary species, such as alerting each other to predators (warning calls) or by

social tolerance allowing them to share food sources (Dobson et al., 2012; Griffiths & Armstrong, 2001; Viblanc et al., 2016b; Viblanc et al., 2022; Walmsley et al., 2023). This can have significant fitness benefits as shown in Columbian ground squirrels (*Urocitellus columbianus*), which have an overall greater number of weaned offspring when they have female kin neighbours (Viblanc et al., 2016b). Solitary animals may lose some of the direct benefits of group living (Makuya & Schradin, 2024a), and forming a kinship spatial structure could be a strategy to reduce such costs (Makuya & Schradin, 2024b).

When individuals derive benefits from close kin, they increase their inclusive fitness, which often occurs in pair- and group-living species (Ferriere & Michod, 2011; Gardner & West, 2014; Hamilton, 1964a; Hamilton, 1964b). That the ranges of close female kin of solitary species overlap more than those of non-kin might be a result of them living closer to each other, which indicates higher tolerance between close kin. Such situations might then enable some collaboration, as described for other solitary mammals (Elbroch & Wittmer, 2012; Lührs & Kappeler, 2013; Meshriy et al., 2011). Future studies should test whether closely related bush Karoo rats share important foraging grounds and whether they might collaborate in territorial defence of areas with resources. While individuals overlap in space, they may not always overlap in time, which can be confirmed studying the fine scale interactions of individuals (Schlägel et al., 2019). Kin selection might provide a basis for understanding the social system of solitary species with a kin-based spatial structure, although this has been largely ignored in the literature.

GPS data-loggers have been used in several large mammal studies to assess their spatial structure, such as in cougars (*Puma concolor*) and fossas (*Cryptoprocta ferox*) where this technique revealed previously unknown relationships, often influenced by kinship (Elbroch et al., 2017a; Lührs & Kappeler, 2013). By using min-GPS for the first time in a small rodent to determine spatial structure, we found that the ranges of bush Karoo rats overlapped more with their kin than with their non-kin neighbours. Large carnivores which display territorial defence had greater overlap in spatial ranges when food availability was low (Nams et al., 2023). In our study, season did not significantly influence range overlap, but we cannot exclude this possibility (see Fig. 4b). However, we found that bush Karoo rats had larger ranges in the wet, breeding season than in the dry, non-breeding season. This is surprising, since more food is available in the wet season, and one might expect bush Karoo rats to instead extend their home ranges in the food-deprived dry season. In the wet breeding season, females might have to forage over larger areas to obtain both energy and protein rich food necessary for breeding. The shrubs into which lodges are built offer some food (leaves) even in the dry season. Smaller ranges in the dry season might help save energy, as occurs in the synoptic striped mouse which also has smaller home ranges in the dry season (Schradin et al., 2010b). Our GPS data confirmed that the spatial structure in the solitary bush Karoo rat is not random but influenced by kinship.

While solitary living has long been regarded as an ancestral and primitive trait, recent comparative studies indicate that solitary living might be a derived state in mammals, and an adaptation to specific environments (Makuya & Schradin, 2024a). However, studying solitary living is important to understand mammalian social evolution (Clutton-Brock, 2021; Makuya & Schradin, 2024a). Multiple studies using modern network analysis indicate that even solitary living mammals (social organization) might have complex social interactions with their neighbours (social structure) (Elbroch & Wittmer, 2012; Lührs & Kappeler, 2013; Meshriy et al., 2011). Our study adds to uncovering the complexity of solitary mammals. It provides insight into the importance of kinship in a solitary species and that intra-specific variation in social organization can also occur a primarily solitary species. Our observations of female kin structure and the rare occurrence of female kin groups indicate the possibility of evolution leading to social groups.

Conclusion

The evolution of sociality can only be explained when also conducting studies on solitary species (Clutton-Brock, 2021; Makuya & Schradin, 2024b). Understanding solitary species will help us to better conserve them (Olivier et al., 2022c). Several years of field data showed that bush Karoo rats are mainly solitary living, yet their spatial organization is influenced by kinship. Solitary species may miss out on the benefits gained by group living species (Makuya & Schradin, 2024a), but the costs of solitary living can be reduced by a kin-based spatial structure. The groups we observed consisted of close kin, and these groups likely formed as a result of delayed dispersal. This is a potential starting point for the evolution of group-living. In conclusion, our study supports the notion that kinship and kin selection might be important in solitary species.

Acknowledgements

We are grateful for the help in data collection provided by several volunteers, research assistants, and students, especially Jennifer Thomson (2018), Lindy Wolhuter (2019) and Inbar Schnitzer (2019/2020). The comments of three referees significantly improved the manuscript. This study was made possible by the administrative and technical support of the Succulent Karoo Research Station (registered South African NPO 122-134). We are grateful for the support of Goegap Nature Reserve and its staff as well as the Northern Cape Department of Environment & Nature Conservation. This study was supported by the CNRS and the University of the Witwatersrand via a CNRS-Wits joint PhD program. This study is part of the long-term Studies in Ecology and Evolution (SEE-Life) program of the CNRS.

Appendix

Table A 1 Results of the generalized linear mixed effects models to identify whether population density and the season have an impact on the percentage of solitary living females in the bush Karoo rat.

Predictors	Estimates	95% CI	*P*
(Intercept)	99.4	90.68 – 108.1	**< 0.001**
Population density	-0.43	-0.96 – 0.09	0.116
Season (non-breeding)	0.895	-5.36 – 3.43	0.691
Random effects σ2/ICC	37.21/0.386		
Marginal R2/conditional R2	0.41/0.039		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.
Factors in bold type indicate significant predictors.
%solitary living = season + population density + (1|year)

Table A 2 Results of the linear mixed effects models to identify which relationship category predicts the number of neighbours a rat has.

Predictors	Estimates	95% CI	P
(Intercept)	0.61	0.45 – 0.74	< 0.001
Relationship (non-kin)	0.11	0.02 – 0.199	< 0.01
Season (non-breeding)	0.1	-0.098 – 0.293	0.35
Random effects σ2/ICC	0.40/0.198		
Marginal R2/conditional R2	0.011/0.207		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.
Factors in bold type indicate significant predictors.
Number of neighbours = relationship (kin VS non-kin) + (1|Focal ID) + (1|Month)

Table A 3 Results of the linear mixed effects models to identify which relationship category predicts the distance to the nearest neighbour.

Predictors	Estimates	95% CI	*P*
(Intercept)	8.89	7.98 – 9.79	**< 0.001**
Relationship (non-kin)	10.35	9.68 – 11.02	**< 0.001**
Season (non-breeding)	-0.44	-1.17 – 0.28	0.23
Random effects σ2/ICC	28.79/0.509		
Marginal R2/conditional R2	0.321/0.667		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.
Factors in bold type indicate significant predictors.

Distance = relationship (kin VS non-kin) + season + (1|Focal ID: Month) + (1|Neighbour ID: Month)

Table A 4 Results of the linear mixed effects models to identify whether season influences daily range size.

Predictors	Estimates	95% CI	*P*
(Intercept)	445.94	365.17 – 526.71	< **0.001**
Season (Breeding)	163.75	57.01 – 273.09	< **0.01**
Random effects σ2/ICC	18,276/0.213		
Marginal R2/conditional R2	0.073/0.271		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.
Factors in bold type indicate significant predictors.
HRS = season + relationship + (1|ID)

Table A 5 Results of the linear mixed effects models to identify whether season influences daily range size overlap.

Predictors	Estimates	95% CI	*P*
(Intercept)	49.34	40.26 – 58.42	< **0.001**
Relationship (non-kin)	-22.41	-31.98 – -12.8	< **0.001**
Season (non-breeding)	-4.96	-14.69 – 4.79	0.32
Random effects σ2/ICC	195.0/0.223		
Marginal R2/conditional R2	0.132/0.326		

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.
The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.
Factors in bold type indicate significant predictors.
Overlap = season + relationship + (1|ID)

References

Anderson, J. M., Burns, E. S., Meese, E. N., Farrugia, T. J., Stirling, B. S., White, C. F., Logan, R. K., O'Sullivan, J., Winkler, C., & Lowe, C. G. (2021). Interannual Nearshore Habitat Use of Young of the Year White Sharks Off Southern California. [Original Research]. *Frontiers in Marine Science, 8*. doi: 10.3389/fmars.2021.645142

Baker, C. J., Frère, C. H., Franklin, C. E., Campbell, H. A., Irwin, T. R., & Dwyer, R. G. (2021). Crocodile social environments dictated by male philopatry. *Behavioral Ecology, 33*(1), 156-166. doi: 10.1093/beheco/arab120

Baker, C. J., Frère, C. H., Franklin, C. E., Campbell, H. A., Irwin, T. R., & Dwyer, R. G. (2023). Long-term tracking reveals a dynamic crocodylian social system. *Animal Behaviour, 199*, 59-78. doi: https://doi.org/10.1016/j.anbehav.2023.02.015

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. . *Journal of Statistical Software, 67*(1), 1-48. doi: 10.18637/jss.v067.i01.

Brown, E., & Willan, K. (1991b). Microhabitat selection and use by the bush Karoo rat *Otomys unisulcatus* in the Eastern Cape Province. *South African Journal of Wildlife Research, 21*, 69-75.

Buchanan, K., Burt de Perera, T., Carere, C., Carter, T., Hailey, A., Hubrecht, R., Jennings, D., Metcalfe, N., Pitcher, T., & Peron, F. (2012). Guidelines for the treatment of animals in behavioural research and teaching.

Calabrese, J. M., Fleming, C. H., & Gurarie, E. (2016). *ctmm*: An r package for analyzing animal relocation data as a continuous-time stochastic process. *Methods in Ecology and Evolution, 7*(9), 1124-1132. doi: https://doi.org/10.1111/2041-210x.12559

Clutton-Brock, T. (2021). Social evolution in mammals. *Science, 373*(6561), eabc9699.

Cooper, L. D., & Randall, J. A. (2007). Seasonal changes in home ranges of the giant kangaroo rat (Dipodomys ingens): a study of flexible social structure. *Journal of Mammalogy, 88*(4), 1000-1008.

Cowling, R. M., Esler, J. J., & Rundel, P. W. (1999). Namaqualand, South Africa - an overview of a unique winter-rainfall desert ecosystem. *Plant Ecology, 142*, 3-21.

Cutrera, A. P., Lacey, E. A., & Busch, C. (2005). Genetic structure in a solitary rodent (*Ctenomys talarum*): implications for kinship and dispersal. *Molecular Ecology, 14*(8), 2511-2523. doi: https://doi.org/10.1111/j.1365-294X.2005.02551.x

Dalerum, F. (2007). Phylogenetic reconstruction of carnivore social organizations. *Journal of Zoology, 273*(1), 90-97. doi: 10.1111/j.1469-7998.2007.00303.x

Dillon, A., & Kelly, M. J. (2008). Ocelot home range, overlap and density: comparing radio telemetry with camera trapping. *Journal of Zoology, 275*(4), 391-398.

Do Linh San, E., Babu, N., Xalu, M., Le Gars, S., Perquin, J., & Baxter, R. (2016). A conservation assessment of Otomys unisulcatus. *The red list of mammals of South Africa, Swaziland and Lesotho*.

Dobson, F. S., Viblanc, V. A., Arnaud, C. M., & Murie, J. O. (2012). Kin selection in Columbian ground squirrels: direct and indirect fitness benefits. *Molecular ecology, 21*(3), 524-531.

Dong, X., Fleming, C., Noonan, M., & Calabrese, J. (2018). Ctmmweb: A Shiny Web App for the Ctmm Movement Analysis Package.: https://github.com/ctmm-initiative/ctmmweb.

du Plessis, A., & Kerley, G. I. H. (1991). Refuge strategies and habitat segregation in two sympatric rodents *Otomys unisulcatus* and *Parotomys brantsii*. *Journal of Zoology, London, 224*, 1-10.

du Plessis, A., Kerley, G. I. H., & Winter, P. E. D. (1992). Refuge microclimates of rodents: a surface nesting *Otomys unisulcatus* and a burrowing *Parotomys brantsii*. *Acta Theriologica, 37*, 351-358.

Elbroch, L. M., Levy, M., Lubell, M., Quigley, H., & Caragiulo, A. (2017a). Adaptive social strategies in a solitary carnivore. *Science Advances, 3*(10), e1701218.

Elbroch, L. M., & Wittmer, H. U. (2012). Puma spatial ecology in open habitats with aggregate prey. *Mammalian Biology, 77*(5), 377-384.

Ferriere, R., & Michod, R. E. (2011). Inclusive fitness in evolution. [10.1038/nature09834]. *Nature, 471*(7339), E6-E8.

Fleming, C. H., Dresher-Lehman, J., Noonan, M. J., Akre, T., LaPoint, S. C., Meyburg, B.-U., van Noordwijk, M. A., Pfeiffer, T., Roulin, A., Séchaud, R., Shephard, J. M., Stabach, J. A., Stein, K., Tonra, C. M., Vogel, E. R., & Calabrese, J. M. (2021). A comprehensive framework for handling location error in animal tracking data. *bioRxiv*. doi: https://doi.org/10.1101/2020.06.12.130195

Fox, J., & Weisberg, S. (2011). *An R companion to applied regression*: Sage publications.

Gardner, A., & West, S. A. (2014). Inclusive fitness: 50 years on. *Philosophical Transactions of the Royal Society B: Biological Sciences, 369*(1642). doi: 10.1098/rstb.2013.0356

Griffiths, S. W., & Armstrong, J. D. (2001). The benefits of genetic diversity outweigh those of kin association in a territorial animal. *Proceedings of the Royal Society of London B, 268*, 1293 - 1296.

Hamilton, W. D. (1964a). The genetical evolution of social behaviour. I. *J Theoret Biol, 7*, 1-16.

Hamilton, W. D. (1964b). The genetical evolution of social behaviour. II. *Journal of Theoretical Biology, 7*(1), 17-52. doi: https://doi.org/10.1016/0022-5193(64)90039-6

He, P., Maldonado-Chaparro, A. A., & Farine, D. R. (2019). The role of habitat configuration in shaping social structure: a gap in studies of animal social complexity. *Behavioral Ecology and Sociobiology, 73*(1), 9. doi: 10.1007/s00265-018-2602-7

Johansson, Ö., Koehler, G., Rauset, G. R., Samelius, G., Andrén, H., Mishra, C., Lhagvasuren, P., McCarthy, T., & Low, M. (2018). Sex-specific seasonal variation in puma and snow leopard home range utilization. *Ecosphere, 9*(8), e02371.

Kappeler, P. M. (2019). A framework for studying social complexity. [journal article]. *Behavioral Ecology and Sociobiology, 73*(1), 13. doi: 10.1007/s00265-018-2601-8

Kovach, A. I., & Powell, R. A. (2003). Effects of body size on male mating tactics and paternity in black bears, Ursus americanus. *Canadian Journal of Zoology, 81*(7), 1257-1268.

Lacey, E. A., & Wieczorek, J. R. (2004). Kinship in colonial tuco-tucos: evidence from group composition and population structure. *Behavioral Ecology, 15*(6), 988-996. doi: 10.1093/beheco/arh104

Lawson Handley, L., & Perrin, N. (2007a). Advances in our understanding of mammalian sex-biased dispersal. *Molecular ecology, 16*(8), 1559-1578.

Li, X. Y., & Kokko, H. (2019). Sex-biased dispersal: A review of the theory. *Biological Reviews, 94*(2), 721-736.

Lührs, M. L., & Kappeler, P. M. (2013). Simultaneous GPS tracking reveals male associations in a solitary carnivore. *Behavioral Ecology and Sociobiology, 67*, 1731-1743.

Maher, C. R. (2009). Genetic relatedness and space use in a behaviorally flexible species of marmot, the woodchuck (Marmota monax). *Behavioral Ecology and Sociobiology, 63*(6), 857-868. doi: 10.1007/s00265-009-0726-5

Makuya, L., Pillay, N., Rimbach, R., & Schradin, C. (2023). Suppressing the physiological stress response is not stress. *Trends in Ecology and Evolution*(3), 224-227.

Makuya, L., & Schradin, C. (2023). Measuring range sizes in a 100-g rodent: mini-GPS are more reliable than transmitters, but the location error reduces reliability. *Mammalian Biology, 103*(5), 455-465. doi: https://doi.org/10.1007/s42991-023-00365-4

Makuya, L., & Schradin, C. (2024a). Costs and benefits of solitary living in mammals. *Journal of Zoology, 323*, 9-18. doi: https://doi.org/10.1111/jzo.13145

Makuya, L., & Schradin, C. (2024b). The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences, 121*(13). doi: https://doi.org/10.1073/pnas.2402871121

Meshriy, M. G., Randall, J. A., & Parra, L. (2011). Kinship associations of a solitary rodent, Dipodomys ingens, at fluctuating population densities. *Animal Behaviour, 82*(4), 643-650.

Mourier, J., Vercelloni, J., & Planes, S. (2012). Evidence of social communities in a spatially structured network of a free-ranging shark species. *Animal Behaviour, 83*(2), 389-401. doi: https://doi.org/10.1016/j.anbehav.2011.11.008

Murray, D. L., & Fuller, M. R. (2000). *Research techniques in animal ecology: Controversies and Consequences*. New York: Columbia University Press.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., Fonseca, G. A. B. D., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature, 403*, 853-858.

Nams, V. O., Parker, D. M., Weise, F. J., Patterson, B. D., Buij, R., Radloff, F. G., Vanak, A. T., Tumenta, P. N., Hayward, M. W., & Swanepoel, L. H. (2023). Spatial patterns of large African cats: a large-scale study on density, home range size, and home range overlap of lions Panthera leo and leopards Panthera pardus. *Mammal Review, 53*(2), 49-64.

Nowak, R. M., & Wilson, E. (1999). *Walker`s Mammals of the World* (6th ed.). Baltimore: John Hopkins University Press.

Olivier, C.-A., Schradin, C., & Makuya, L. (2022c). Global change and conservation of solitary mammals. [Opinion]. *Frontiers in Ecology and Evolution, 10*. doi: 10.3389/fevo.2022.906446

Pillay, N. (2001). Reproduction and postnatal development in the bush Karoo rat Otomys unisulcatus (Muridae, Otomyinae). *Journal of Zoology, 254*(4), 515-520.

Potts, J. R., & Lewis, M. A. (2014). How do animal territories form and change? Lessons from 20 years of mechanistic modelling. *Proceedings of the Royal Society B: Biological Sciences, 281*(1784). doi: 10.1098/rspb.2014.0231

QGIS, D. T. (2009). QGIS Geographic Information System.

R Core Team. (2022b). R: A Language and Environment for Statistical Computing. Vienna, Austria. Retrieved from https://www.R-project.org/

Rafiq, K., Jordan, N. R., Meloro, C., Wilson, A. M., Hayward, M. W., Wich, S. A., & McNutt, J. W. (2020). Scent-marking strategies of a solitary carnivore: Boundary and road scent marking in the leopard. *Animal Behaviour, 161*, 115-126.

Roex, N. l., Mann, G. K. H., Hunter, L. T. B., & Balme, G. A. (2022). Relaxed territoriality amid female trickery in a solitary carnivore. *Animal Behaviour, 194*, 225 - 231. doi: https://doi.org/10.1016/j.anbehav.2022.09.022

Schlägel, U. E., Signer, J., Herde, A., Eden, S., Jeltsch, F., Eccard, J. A., & Dammhahn, M. (2019). Estimating interactions between individuals from concurrent animal movements. *Methods in Ecology and Evolution, 10*(8), 1234-1245.

Schlichting, P. E., Boughton, R. K., Anderson, W., Wight, B., VerCauteren, K. C., Miller, R. S., & Lewis, J. S. (2022). Seasonal variation in space use and territoriality in a large mammal (Sus scrofa). *Scientific Reports, 12*(1), 4023. doi: 10.1038/s41598-022-07297-y

Schoepf, I., & Schradin, C. (2012a). Better off alone! Reproductive competition and ecological constraints determine sociality in the African striped mouse (*Rhabdomys pumilio*). *Journal of Animal Ecology, 81*(3), 649-656. doi: 10.1111/j.1365-2656.2011.01939.x

Schradin, C. (2005). Nest-Site Competition in Two Diurnal Rodents from the Succulent Karoo of South Africa. *Journal of Mammalogy, 86*(4), 757-762. doi: 10.1644/1545-1542(2005)086[0757:Ncitdr]2.0.Co;2

Schradin, C. (2013). Intraspecific variation in social organization by genetic variation, developmental plasticity, social flexibility or entirely extrinsic factors. [Review]. *Philosophical Transactions of the Royal Society B-Biological Sciences, 368*(1618). doi: 10.1098/rstb.2012.0346

Schradin, C., Hayes, L. D., Pillay, N., & Bertelsmeier, C. (2018a). The evolution of intraspecific variation in social organization. *Ethology, 124*, 527-536. doi: doi:10.1111/eth.12752

Schradin, C., König, B., & Pillay, N. (2010a). Reproductive competition favours solitary living while ecological constraints impose group-living in African striped mice. *Journal of Animal Ecology, 79*, 515-521.

Schradin, C., Pillay, N., & Bertelsmeier, C. (2019). Social flexibility and environmental unpredictability in African striped mice. *Behavioral Ecology and Sociobiology, 73*(7), 94. doi: 10.1007/s00265-019-2707-7

Schradin, C., Schmohl, G., Rödel, H. G., Schoepf, I., Treffler, S. M., Brenner, J., Bleeker, M., Schubert, M., König, B., & Pillay, N. (2010b). Female home range size is regulated by resource distribution and intraspecific competition: a long-term field study. *Animal Behaviour, 79*(1), 195-203.

Silva, I., Fleming, C. H., Noonan, M. J., Alston, J., Folta, C., Fagan, W. F., & Calabrese, J. M. (2022). Autocorrelation-informed home range estimation: A review and practical guide. . *Methods in Ecology and Evolution, 13*, 534-544. doi: https://doi.org/10.1111/2041-210X.13786

Siracusa, E., Boutin, S. A., Dantzer, B., Lane, J. E., Coltman, D. W., & McAdam, A. G. (2021). Familiar neighbors, but not relatives, enhance fitness in a territorial mammal. *Current Biology, 31*, 438-455. doi: 10.1016/j.cub.2020.10.072

Stamps, J. (1994). Territorial behavior: testing the assumptions. *Adv Study Behav, 23*, 173-232.

Valomy, M., Hayes, L. D., & Schradin, C. (2015a). Social organization in Eulipotyphla: evidence for a social shrew. *Biology Letters, 11*(11). doi: 10.1098/rsbl.2015.0825

Vermeulen, H., & Nel, J. (1988). The bush Karoo rat *Otomys unisulcatus* on the Cape West coast. *African Zoology, 23*(2), 103-111.

Viblanc, V. A., Saraux, C., Murie, J. O., & Dobson, F. S. (2016b). Kin effects on energy allocation in group-living ground squirrels. *Journal of Animal Ecology*, n/a-n/a. doi: 10.1111/1365-2656.12541

Viblanc, V. A., Saraux, C., Tamian, A., Criscuolo, F., Coltman, D. W., Raveh, S., Murie, J. O., & Dobson, F. S. (2022). Measuring fitness and inferring natural selection from long-term field studies: different measures lead to nuanced conclusions. *Behavioral Ecology and Sociobiology, 76*(6), 79.

Walmsley, S. F., Boutin, S., Dantzer, B., Lane, J. E., Coltman, D. W., & McAdam, A. G. (2023). Benefits of living closer to kin vary by genealogical relationship in a territorial mammal. *Proceedings of the Royal Society B: Biological Sciences, 290*, 20221569. doi: https://doi.org/10.1098/rspb.2022.1569

Waser, P. M., & Jones, W. T. (1983). Natal philopatry among solitary mammals. *The quarterly review of biology, 58*(3), 355-390.

Webber, Q. M. R., Albery, G. F., Farine, D. R., Pinter-Wollman, N., Sharma, N., Spiegel, O., Vander Wal, E., & Manlove, K. (2023). Behavioural ecology at the spatial–social interface. *Biological Reviews, 98*(3), 868-886. doi: https://doi.org/10.1111/brv.12934

Webber, Q. M. R., & Vander Wal, E. (2018). An evolutionary framework outlining the integration of individual social and spatial ecology. *Journal of Animal Ecology, 87*(1), 113-127. doi: https://doi.org/10.1111/1365-2656.12773

Williams, D. A., & Rabenold, K. N. (2005). Male-biased dispersal, female philopatry, and routes to fitness in a social corvid. *Journal of Animal Ecology, 74*(1), 150-159.

Winner, K., Noonan, M. J., Fleming, C. H., Olson, K. A., Mueller, T., Sheldon, D., & Calabrese, J. M. (2018). Statistical inference for home range overlap. *Methods in Ecology and Evolution, 9*(7), 1679-1691. doi: https://doi.org/10.1111/2041-210X.13027

Wolhuter, L., Thomson, J., Schradin, C., & Pillay, N. (2022). Life history traits of free-living bush Karoo rats (Otomys unisulcatus) in the semi-arid Succulent Karoo. *Mammal Research, 67*(1), 73-81.

Zuur, A. F., Ieno, E. N., & Elphick, C. S. (2010). A protocol for data exploration to avoid common statistical problems. *Methods in Ecology and Evolution, 1*(1), 3-14. doi: 10.1111/j.2041-210X.2009.00001.x

CHAPTER 5. Tolerant mothers: aggression does not explain solitary living in the bush Karoo rat

Makuya, L[1]., Pillay, N[1]., Sangweni, S. P[1]., & Schradin, C[1,2]. (2024). Tolerant mothers: aggression does not explain solitary living in the bush Karoo rat. *Proc Roy Soc B* 291 (20241534) doi: https://doi.org/10.1098/rspb.2024.1534

Authors, affiliations:

[1] School of Animal, Plant & Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS 2050, Johannesburg, South Africa

[2] IPHC, UNISTRA, CNRS, 23 rue du Loess, 67200 Strasbourg, France

Abstract

Many mammal species are thought to adopt solitary living due to mothers becoming intolerant of adult offspring and social intolerance between adults. However, field studies on how solitary mammals interact are rare. Here we show that solitary living can occur without social intolerance. Over three years, we recorded interactions between free-living bush Karoo rats (*Otomys unisulcatus*) and conducted dyadic encounter experiments between kin and non-kin female neighbours, both in a neutral test arena and in field intruder experiments. Social interactions were rare (230/2062 observations), and aggressive in only 34% of the cases. In dyadic encounters, mothers interacted amicably with young offspring. Aggression between mothers and offspring was almost absent. This mother-offspring relationship remained amicable even after the offspring had dispersed. Aggression between neighbouring adult females was low in neutral arena tests, independent of kinship and season. However, in the field, females reacted more aggressively towards non-kin than kin intruders, especially during the breeding season. Tolerance between mothers and adult offspring indicates that aggression is not the mechanism leading to dispersal and solitary living. We found a solitary social system characterised by social tolerance, suggesting that dispersal and a lack of social attraction rather than aggression can lead to solitary living.

Keywords: Aggression; kin neighbours; natal dispersal; social organisation; social structure; social system; social tolerance; solitary living

Introduction

To understand the diversity of social systems, many studies have focused on pair and group-living species, assuming solitary living to be the ancestral stage that does not require any explanation for its occurrence (Lukas & Clutton-Brock, 2013a; Makuya & Schradin, 2024a, 2024b). Solitary, pair- and group-living refer to the social organisation, describing the composition of social units (Kappeler, 2019). Social organisation is one of four components of the social system which further includes the care system, mating system and social structure (Kappeler, 2019; Kappeler & Schaik, 2002). Of all the different forms of social organisation, solitary living is the most understudied (Makuya & Schradin, 2024a).

Previous studies regarded solitary living as the ancestral default form of social organisation in mammals (Lukas & Clutton-Brock, 2013a). However, recent comparative studies have shown that this is often not the case (Olivier et al., 2024; Schradin, 2017). These studies showed that pair-living was most likely the ancestral form of social organisation in artiodactyls (Jaeggi et al., 2020), primates (Olivier et al., 2022b), and possibly in marsupials (Qiu et al., 2022) and Eulipotyphla (Valomy et al., 2015a), indicating that solitary living is often a derived state (Makuya & Schradin, 2024a). However, we know little about the mechanisms leading to solitary living in mammals (Makuya & Schradin, 2024a). To understand the mechanisms of group-living, we also must understand the alternative, which is solitary living (Makuya & Schradin, 2024a).

It is generally assumed that solitary living in mammals is due to aggression. This opinion is based on work done on the European hamster (*Cricetus cricetus*) in the 1950s (Eibl-Eibesfeldt, 1953). In this species, mothers become intolerant of their offspring when they reach puberty and adults are highly intolerant of each other (Eibl-Eibesfeldt, 1953). The assumption that social intolerance is the main reason of solitary living in mammals was further based on standardized laboratory experiments on solitary rodents to examine the proximate mechanisms of aggression (Elidio et al., 2021; Fischer et al., 2007; Krause & Schüler, 2010). However, whether this is always the mechanism leading to solitary living under natural conditions is unknown. One of the main reasons for solitary living is high reproductive competition between females which often leads to female infanticide (Clutton-Brock et al., 2006; Hodge et al., 2011; Wolff, 2007). As an alternative to social intolerance and aggression, a lack of social attraction combined with a motivation to disperse when reaching sexual maturity could lead to a solitary lifestyle. In solitary mustelids, for example, individuals commonly meet in a non-aggressive context (Twining & al., 2024). Social interactions

in nature have been studied for a few solitary living species such as the puma (*Puma concolor*) (Roex et al., 2022) and the giant Kangaroo rat (*Dipomys ingens*) (Meshriy et al., 2011). While social interactions in these solitary species were rare, these studies highlight the lack of aggressive interactions when individuals met. More field studies are needed to understand the mechanisms that cause solitary living.

Solitary species can have complex social structures in which individuals interact in a non-random way (Makuya et al., 2024b). They often display kinship-determined spatial patterns where kin live close to each other and share part of their range and engage in non-aggressive interactions when they meet (Meshriy et al., 2011; Roex et al., 2022). The kinship patterns in these solitary species are driven by philopatry, a behaviour usually displayed by group living species, with individuals dispersing only short distances and forming kin clusters (Cooper & Randall, 2007). For example, the social structure of the giant kangaroo rat is formed by female kin neighbours, with shorter distances between neighbours leading to increased social interactions (Meshriy et al., 2011). The frequency of social interactions was positively related to population density, indicating an influence of season on the social and spatial structure (Johansson et al., 2018). Amicable interactions at territory boundaries are not a contradiction to the theoretical assumption that aggression is a main driver of solitary living, but could simply be due to the dear enemy phenomenon, where individuals are tolerant of known neighbours as long as they do not cross territory boundaries (Temeles, 1994). To test whether solitary living can arise without aggression, one would need to measure aggression also inside territories and at nesting sites. Field experiments are needed to test the level of aggression in solitary species, and whether close kin are more tolerant towards each other than non-kin.

Our aim was to test the general assumption that aggression and social intolerance are the mechanisms leading to solitary living in our study species, the bush Karoo rat (*Otomys unisulcatus*) from South Africa. In particular, if aggression is the main mechanism leading to natal dispersal and solitary living, we predicted (1) that the behaviour of mothers changes towards their offspring as the offspring become older, with mothers showing more aggression towards dispersed adult offspring than juvenile offspring still living with the mother. Since our study species has a kin based spatial structure with significant overlap of home ranges between close kin (Makuya et al., 2024b), we further predicted (2) that they would show higher levels of aggression towards non-kin neighbours than towards kin neighbours. To test these predictions, we conducted more than 2000 focal animal observations over a period of 3 years, conducting tests in a neutral presentation arena in a field laboratory, and conducted field experiments where we presented kin and non-kin neighbours at the nesting sites of resident females.

Materials and methods

Study site

The study was conducted in the Goegap Nature Reserve, in the Northern Cape, South Africa. The study site is located in the Succulent Karoo biome (Cowling et al., 1999). The climate is arid with temperatures falling below 0°C in winter and exceeding 40°C in summer (Schradin, 2005). Mean precipitation at the field site is 160mm per annum. Seasons are divided into the hot dry non-breeding season (December to May) and the cold wet breeding season (June to November). The bush Karoo rat reproduces in the wet season.

Study species

The 100g heavy bush Karoo rat offers a model to study solitary living because it is diurnal, occupies an open habitat, has small home ranges (0.06 ± 0.04ha in the dry, non-breeding season and 0.04 ± 0.03ha in the wet, breeding season) (Makuya et al., 2024b), and easily habituates to the presence of observers. It inhabits the semi-arid regions of South Africa including the Succulent Karoo (less than 200mm of rain per annum), one of the world's most important biodiversity hotspots (Myers et al., 2000). The bush Karoo rat builds stick-lodges inside shrubs, which offers a favorable microclimate with high humidity and mild temperatures as protection against the harsh outside environment that is characterized by unpredictable rain in cold winters, and long, hot summer dry seasons (Brown & Willan, 1991b; du Plessis & Kerley, 1991; du Plessis et al., 1992). The bush Karoo rat is a central place forager, foraging around its stick lodge and bringing food back to the lodge, where it can be easily observed, and where experiments can be conducted. In the Succulent Karoo, around 95% of the rats are solitary, although a few small groups of 2-3 closely related females occur (Makuya et al., 2024b). They have a kin based spatial structure, with females having more kin than non-kin neighbours, and home ranges of close kin overlapping more with each other than of non-kin (Makuya et al., 2024b). Young male bush Karoo rats behave similarly to females at the beginning of the dry season and stay in an area close to their natal lodge. However, they disperse in winter when food availability increases just before the breeding season starts. Adult males roam over very large areas and there are no resident males on the field site during the breeding season. Thus, there are no male neighbours in the breeding season.

Sampling regime

Marking and trapping

Trapping was conducted at lodges that showed signs of being occupied (fresh faeces, active runways, rats observed). The field site was divided into six areas, with 1-2 areas trapped simultaneously by research teams. All lodges within one area were trapped for three consecutive days before moving on to the next

area. Traps were set in the morning before sunrise and checked after 45 and 90 minutes and then un-set during the hottest times of the day. In the afternoon, traps were set 45 minutes before sundown, checked after sundown, and then un-set during the night. We used a combination of foldable Sherman traps (https://shermantraps.com/) and locally produced heavy metal traps of the Sherman style. Traps were baited with a combination of bran flakes, salt and sunflower oil and re-baited each morning and afternoon. Traps were arranged around the entrances of the lodges and along runways. We recorded the body weight of individuals to the nearest 0.1-gram, as well as their sex, reproductive status and lodge number. We marked individuals with single, metal-band ear tags with a unique reference number (National Band and Tag Co., Newport, KY, U.S.A.) (Schoepf & Schradin, 2012a). To aid in visual identification during observations, individuals were marked with non-toxic hair dye (Inecto Rapido, Pinetown, South Africa), in combinations (females: head and chest/sides/back; males: hindquarters and chest/sides/back). Age was estimated from body mass at first capture, using a species-specific growth curve (Pillay, 2001) validated for our field data. The bush Karoo rats were classified according to their age, with pups being up to two weeks old and weighing less than 30 g (weaning is at 14 days; (Pillay, 2001)), juveniles being 2-6 weeks old and weighing between 30 and 70 g, and adults being older than 6 weeks and weighing more than 70 g, when both sexes can start reproduction (Pillay, 2001).

Determining dispersal

The onset of dispersal was determined for the rats used during the dyadic encounter tests (explained below). We determined these occurrences from the time that the rat was trapped consistently over a period of more than 4 weeks at a lodge that was not its natal lodge, without the mother being trapped and observed at the same lodge. Using these data, we calculated the age at dispersal.

Focal animal observations in the field

Focal animal observations were conducted between March 2021 and October 2023 for a total of 2279 observations for 246 rats at lodges with identified bush Karoo rats to establish 1) which individuals occupy the lodges, and 2) record behaviours, including interactions with conspecifics. The observations were done for 30 minutes in the morning after sunrise and 30 minutes in the afternoon before sunset. Observations were done using focal animal sampling and one zero recording for 30min. We recorded all social behaviours in 1-minute intervals. The social behaviours including the following groups of behaviour: (i) amicable behaviours (e.g., grooming, body contact); (ii) social investigation (i.e., sniffing); and (iii) aggression (e.g., chasing, fighting).

Dyadic encounter tests

Dyadic encounter experiments were used to assess whether interactions with neighbours were amicable (predicted for close kin) or aggressive (predicted for non-kin). Bush Karoo rats were trapped and brought inside their trap to a laboratory at the research station, located 100 m away, and allowed to acclimatize for 10 minutes before the start of the experiment. The experiments were conducted in a neutral test arena that was constructed of wood chip panels (80 cm x 65 cm x 94 cm) and had a partition in the middle (Figure S 1, supplementary file). The testing arena was cleaned between encounters using diluted Dettol Antiseptic Liquid and then air dried. All tests were done between 10 am and 12 pm from August 2021 until March 2023, for a total of 143 tests on 52 focal rats. Each rat was tested an average of 2.80 ± 2.2 times.

Each bush Karoo rat was introduced into the arena and allowed to settle for 5 minutes with the partition down. We first tested mothers as the focal individual against their offspring which were between one month and 20 months old (N = 80 tests, 3 with male offspring and 77 with female offspring, on 31 focal mother rats). The offspring tested were either still living in their mothers' lodge i.e., not dispersed (N = 43) or already dispersed (N = 36). We attempted to test each mother with the same offspring at different ages, but because some rats were not re-trapped and disappeared, some mothers were tested with different offspring at different ages. Next, we tested adult female focal rats on two different days with an adult female kin neighbour and an adult female non-kin neighbour respectively (N = 63 tests on 41 rats). Half of the rats were first tested with a kin neighbour, the other half with a non-kin neighbour. We defined a neighbour as a rat that occupied a lodge not more than 25 metres away from the focal rat. A total of 22 of the focal rats tested with a neighbour were used in the tests with offspring. Because all stimulus animals were direct neighbours, our experimental design controlled for the dear enemy phenomenon, whereby the owner of a territory responds less aggressively to a familiar neighbour than towards a stranger (Temeles, 1994). Presentations lasted for 15 minutes each. The focal adult individual was heavier than the stimulus female (mean weight difference 25.8g ± 20.3 *SD*), because we wanted the focal individual, which we assigned as the owner of the territory, to initiate the encounters and body mass difference was expected to have a positive influence on the initiation of aggression (Schradin, 2004). At the end of each test, bush Karoo rats were returned to their lodge. Focal animal sampling was used to record the frequency of social behaviours as described for focal animal observation above (Martin & Bateson, 1993). The behaviours were recorded for 15 minutes using a webcam. The observer was present in the same room as the animals being tested but was separated from the animals by a black curtain and monitored the behaviours live on a computer. Tests would have been immediately terminated as soon as individuals started damaging fights (biting and/or standing upright and boxing for more than 2 seconds) to avoid any injury. However, this was never necessary in our study. Interactions videos were then scored using BORIS (Behavioural Observation Research Interactive Software, (Friard & Gamba, 2016)).

Field intruder presentation tests

Since we observed little aggression during the neutral arena tests, we conducted field intruder tests directly at the lodge of the rats. A similar experiment had been done on group living African ice rats (*Otomys sloggetti robertsi*) from the alpine regions of the southern African Drakensberg and Maluti mountains (Hinze et al., 2013). We expected higher levels of aggression here due to the focal rats defending their territory, which was not the case in the neutral arena in the laboratory. Again, we tested whether focal animals are more aggressive towards non-kin than kin neighbours. These tests were done 1-2 years after the dyadic encounter tests in the neutral test arena, and of the individuals tested in the field, only nine had participated as the focal (N = 9) / stimulus (N = 6) in the previous experiments. Stimulus individuals were trapped (as described above) and then transferred into a wire mesh cage (30 x 15 cm, 12 cm height) (Figure S2). This wire mesh cage allowed other bush Karoo rats to see and smell the stimulus animal in the trap, but not to reach and touch it. The trapped individual was then presented at a neighbouring rat`s lodge (i.e. the focal individual). The focal individual was not caged and its response to the trapped stimulus individual was recorded. The wire cage was positioned within an active runway 30 cm away from the lodge. Observations started when the focal animal was observed outside its lodge and lasted for 15 minutes thereafter. The maximum duration of the presentation was 45 min; thus, when the focal animal was not seen within the first 30 min, the experiment was terminated. During the 15 min of observations, we recorded aggressive behaviours of the focal animal towards the caged stimulus animal, including charging towards the cage and emitting chit sounds. We also recorded the latency until the investigation of the stimulus individual and the first aggression as well as the total time spent at the cage. Thereafter, the stimulus animal was returned to its lodge from where it was trapped. Each focal was tested, once with a kin neighbour and once with a non-kin neighbour on separate days. The tests were conducted in the early mornings between 6:00 and 9:00 from January – November 2023. We conducted a total of 54 tests for 26 focal rats with each rat being tested for an average of 2.03 ± 0.87 times. For both the dyadic and field encounter tests. a focal rat could be used as a stimulus in another experiment, and a stimulus rat could be used multiple times for different focal rats. In the dyadic tests, 15 rats were used as both focal and stimulus and 12 rats were used in the intruder tests.

Statistical analysis

All statistical analyses were done in R (version 4.3.1; R Core Team, 2022b). We used linear and generalized mixed models to analyse factors that influence the frequency of sniffing events, or the time spent in body contact, fighting, or grooming, and included the age of offspring, and season (breeding vs non-breeding) as factors for the dyadic encounter tests between mother and offspring (Table 1).

Table 1: Hypothesis and associated models tested in the dyadic and field intruder tests.

Hypothesis	Type experiment	Model	Model type
The behaviour of mothers changes towards their offspring as the offspring become older.	Dyadic encounter – mother : offspring	Time in body contact = season + age of offspring.	LME
		Time fighting = season + age of offspring.	LME
		Time grooming = season + age of offspring.	LME
		Frequency of sniffing = season + age of offspring.	GLMM - Poisson
Bush Karoo rats show higher levels of aggression towards non-kin than towards kin neighbours	Dyadic encounter - neighbours	Fight = season + relatedness + body size difference + (1|ID-focal).	LME
		Body contact = season + relatedness + body size difference + (1|ID-focal).	LME
		Groom = season + relatedness + body size difference + (1|ID-focal).	LME
		Sniff = season + relatedness + body size difference + (1|ID-focal).	GLMM - Poisson
	Field intruder test	Latency to first aggression = season * relatedness + body size difference + (1|ID-focal).	LME
		Latency to approach = season * relatedness + body size difference + (1|ID-focal).	LME
		Charging = season * relatedness + body size difference + (1|ID-focal).	GLMM - Poisson

	Trills (chit) = season * relatedness + body size difference + (1|ID-focal).	LME
	Time at cage = season * relatedness + body size difference + (1|ID-focal).	LME

We ran linear mixed models (LME) in *lme4* to investigate factors that influenced the behaviour exhibited by bush Karoo rats during dyadic encounter tests toward neighbours. For affiliative behaviours, we tested the time spent sitting in body contact, and time spent grooming. For aggressive behaviours we tested the duration of fights. Finally, for social investigative behaviours, we fit a generalized linear mixed model (GLMM) with a Poisson distribution, and we tested the number of sniffing events displayed. For all the model, the duration/frequency of the behaviours were fitted in linear models as predictors, and we included season, body size difference, and relatedness as fixed effects and the ID of the focal individual as random effects (Table 1).

We used both generalized linear mixed models (GLMM) and linear mixed models (LME) to analyse factors that influence the behaviour displayed by the focal rat towards a stimulus for the field intruder presentation tests. We tested the latency to approach the cage and to first aggression, and the number of charging events (fitted with a Poisson distribution) and the number of chit sounds, and total time spent at the cage. One model was fit for each behaviour. The behaviours were again fitted as predictors, and the interaction between season and relatedness included as fixed effects. To avoid singularity, we only fitted the ID of the focal rat as a random effect (Table 1).

Results

Mother- offspring interactions

During 87 of the 2062 field observations sessions, pups were present at the mother's nest. On 11 occasions, we observed amicable interactions with the mother (grooming, and body contact), and no interactions occurred on 76 occasions; we never observed aggression towards the offspring by the mother. Juveniles were present on 240 occasions during the field observations and were observed in amicable interactions with the mother on 20 occasions, and only once in an aggressive interaction.

The mean age of all offspring (N = 79) used as stimulus animals was 4.13 ± 3.72 *SD* months. The age of non-dispersed rats in the experiments was 3.81 ± 3.25 *SD* months (range 1 – 14, N = 43) compared to 5.21 ± 4.01 months (range: 1 – 20, N = 36) of dispersed rats. We calculated AIC values for models including the

age of offspring vs dispersed or undispersed. We found that the AIC values for models including the age of offspring as a predictor were higher than for the models that included whether the offspring had dispersed (Table S2, supplementary file). However, the summary tables for both these models were very similar (results and conclusions did not change) and we reported the models including the age of offspring in the electronic supplement. Dyadic encounters between mothers and their female offspring were characterised by sniffing and body contact with little grooming and nearly no aggression (Table S1, supplementary file). Social interactions were not influenced by season and did not significantly change after offspring dispersed (Figure S3; Tables S3-S6: supplementary file). Specifically, mothers were not more aggressive towards dispersed offspring than non-dispersed (Table S4, Figure S4), nor did they decrease the level of body contact (Table S6. Figure S3).

Focal animal observation

In 230 of the 2062 observations, conducted between July 2021 and October 2023, two or more adult rats were present at the focal rats' lodge. Only 97 chases occurred in 79 of the 230 observations. Chases occurred more often during the non-breeding season (t-test; t_{1728} = -2.99, p < 0.01). The chases occurred between a male and an unrelated female (41%) or an unidentified neighbour (41%), and between related females (18%).

Dyadic encounter tests (neutral test arena)

We tested whether adult female bush Karoo rats (focal individuals) showed less aggression towards kin neighbours than towards non-kin neighbours (adult females). The weight difference between the individuals was not a significant predictor in any of the models. Adult female bush Karoo rats showed very little aggression towards their neighbours, irrespective of the season (LME; estimate = 0.016, s.e. = 0.013, CI: -0.0098/0.042, t_{63} = 1.2, p = 0.22) and relatedness (LME; estimate = 0.009, s.e. = 0.015, CI: -0.02/0.04, t_{62} = 0.6, p = 0.55) (Figure S2 and Table S7: supplementary file). However, they spent more time in body contact with kin than with non-kin (LME; estimate = -2.36, s.e. = 1.316, CI: -5.04/0.31, t_{63} = -1.79, p = 0.08; Figure S5 A), but the season was not a significant predictor (LME; estimate: 0.57 + s.e. = 1.094, CI: -1.63/2.8,t_{61} = 0.52, p = 0.60) (Table S8: supplementary file). The time spent grooming neighbours was significantly affected by season (LME; estimate = 0.008, s.e. = 0.019, CI: -0.0295/0.045, t_{63} = 0.42, p = 0.673) or relatedness (LME; estimate = -0.02, s.e. = 0.022, CI: -0.0645/0.025, t_{58} = -0.93, p = 0.356) (Figure S5 B and Table S9: supplementary file). Bush Karoo rats spent a significant amount of time investigating neighbours through sniffing in the breeding season (GLMM; estimate = 0.73, s.e. = 0.27, CI: 0.22/1.29, t = 2.697, p < 0.01) but this was not affected by relatedness (estimate = 0.54, s.e. = 0.36, CI: -0.17/1.27, p = 0.14) (Figure S6 and Table S10: supplementary file).

Intruder field presentation tests

In both seasons, female bush Karoo rats were mildly aggressive towards non-kin neighbours but tolerated kin neighbours when presented at their lodge. There was no difference in the latency to approach the cage with a kin or non-kin focal neighbour (estimate = -0.98, s.e. = 1.61, CI: -4.27/2.29, p = 0.6; Figure S8 and Table S12), indicating resident bush Karoo rat females investigated the stimulus animal regardless of kinship. However, non-kin females were attacked much faster (LME; t_{38} = 2.54, p <0.05, Figure 2 and Table S11), their cages were charged more often (GLMM; estimate = 0.75, s.e. = 0.22, CI: 0.32/1.2, t = 3.4, p < 0.001; Figure 2 and Table S13), and they produced more trill sounds (Figure S8; although this was not significant because a large outlier: LME; estimate = 9.53, s.e. = 18.2, CI: -25.45/44.77, t = 1.56, df = 37.59, p = 0.61; Table S14). Also, focal individuals spent significantly more time at the cage with the non-kin than kin neighbours (LME; estimate = 4.63, s.e = 1.37, CI: 1.76/7.39, t_{39} = 4.06, p < 0.01, Figure. 4 and Table S15). These differences occurred in both the breeding and the non-breeding seasons. However, in the breeding season, female bush Karoo rats showed more aggression towards intruders, regardless of kinship (LME; t = -4.32, p < 0.001) by quickly attacking (LME; estimate = 4.287, s.e. = 1.82, CI: 0.61/7.95, t_{43} = 2.349, p = 0.02), and charging at cages (LME; estimate = -1.92, s.e. = 0.44, CI: -2.899/-1.10) (Figures. 2-3).

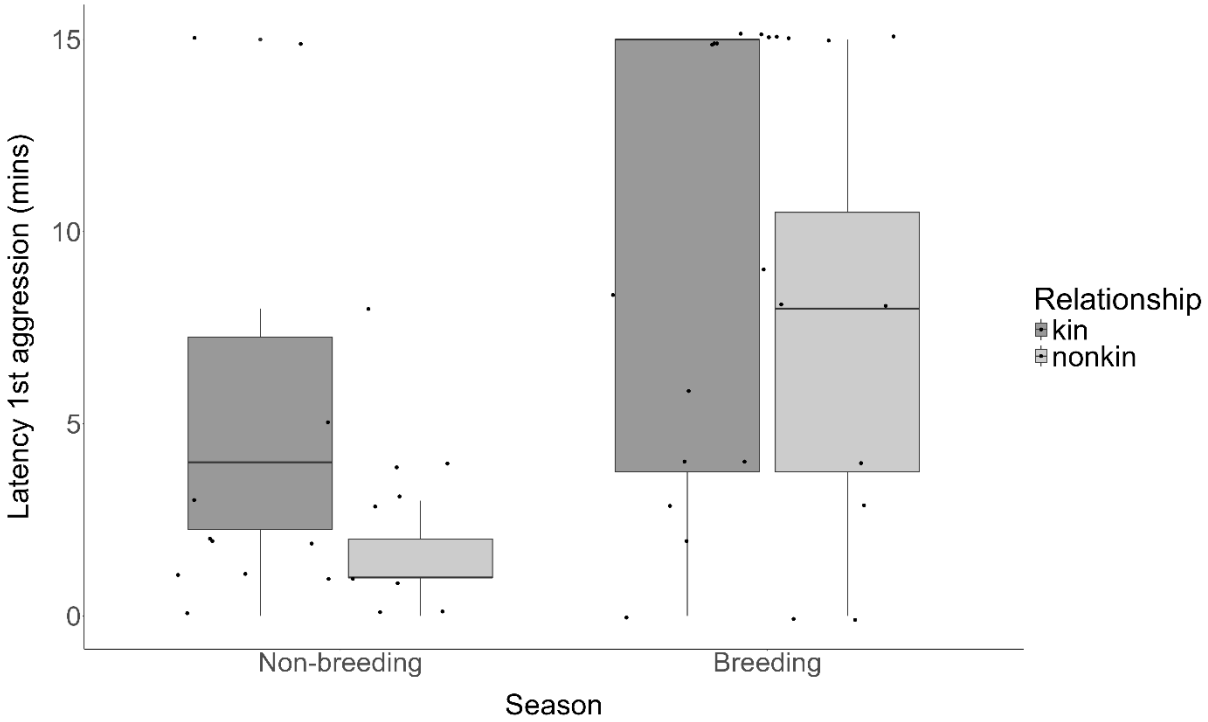

Figure 1. The latency to first aggression by focal females to neighbouring cage-housed female bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values.

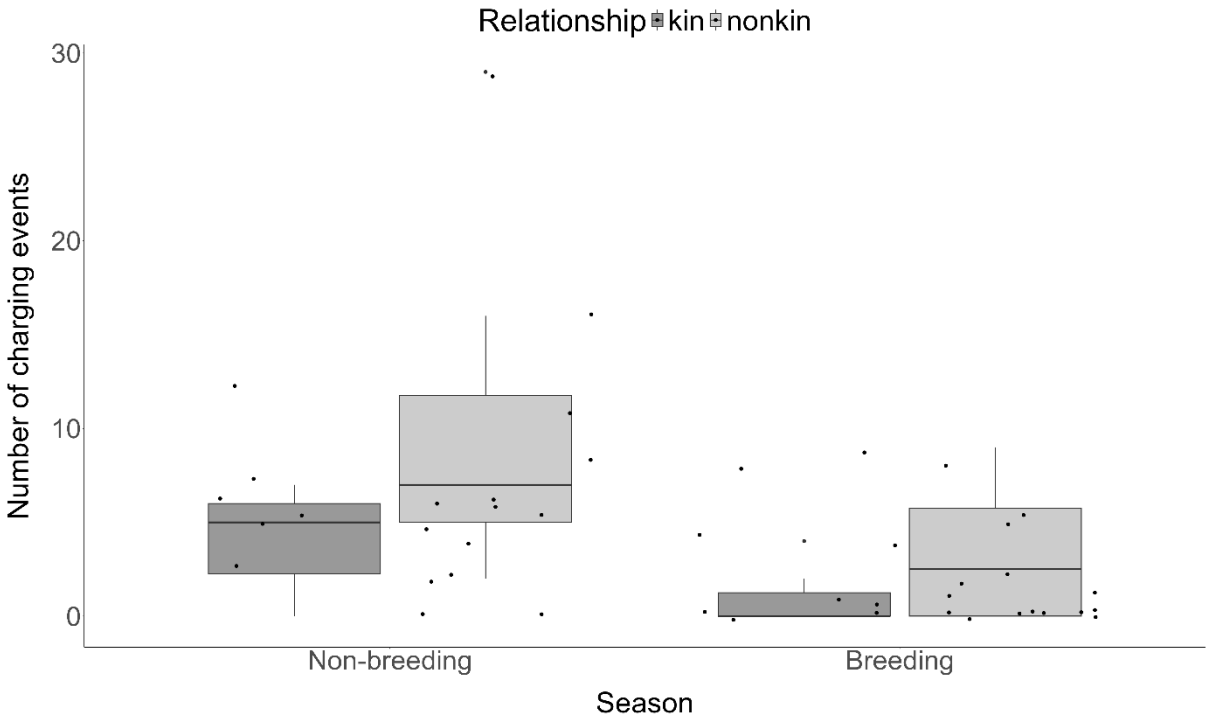

Figure 2. The number of charging events by focal females to neighbouring cage-housed bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values.

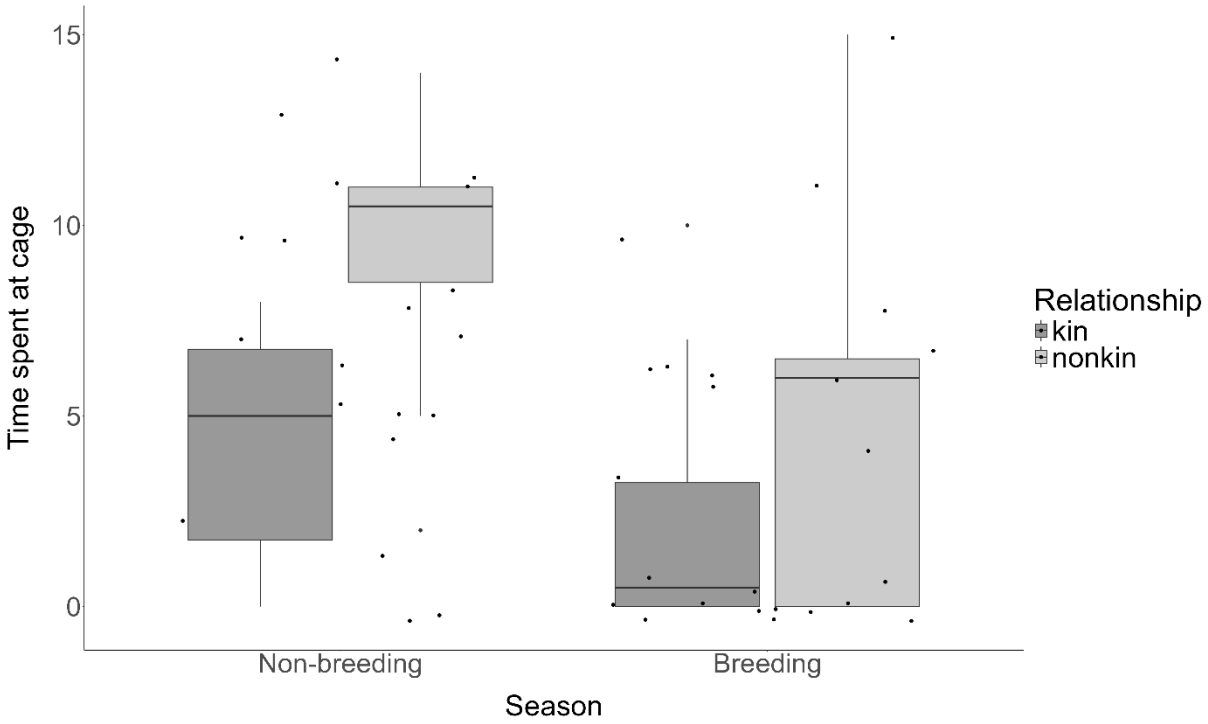

Figure 3. The time spent at the cage by focal females to neighbouring cage-housed bush Karoo rats during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data, and points represent individual values.

Discussion

It is typically assumed that individuals of solitary species are intolerant of each other (Clutton-Brock, 2021). Here we tested whether female bush Karoo rats live solitarily because of high intra-specific aggression. However, we observed no aggression between mothers and their juvenile or even adult offspring. The mother-offspring relationship did not change when the offspring became older and had dispersed from their mother. In dyadic encounter tests, mother and offspring were regularly in body contact which did not decline as offspring became older. During field observations, mothers interacted rarely with pups and juveniles, but when they did so, it was mainly amicable. Thus, both experimental and field observation data indicate that aggression rarely occurs between mothers and offspring, aggression does not increase as offspring age and have become solitary and is thus unlikely to be the mechanism leading to solitary living. This makes the alternative a likely explanation, i.e. motivation to disperse when reaching sexual maturity together with an absence of social attraction, such that dispersing individuals settle alone inside an unoccupied lodge.

It is typically assumed that aggression is the main mechanism leading to offspring dispersal and solitary living (Bekoff, 1977). The association between aggression and dispersal in which subordinate individuals (juveniles) are driven out by dominant individuals (adults) is well known (Bekoff, 1977; Christian, 1970).

However, there are equally many studies showing that aggression and dispersal are not always associated with one another (Bekoff, 1977). In our study, mothers did not react more aggressively towards their dispersed offspring than towards their offspring still living with them. Female bush Karoo rats reach sexual maturity at six weeks (Pillay, 2001) and are thus expected to leave their mother's lodge (i.e. disperse) earliest at this age. Accordingly, females dispersed at 2 months of age but with a large variation that needs investigation in the future. For example, food availability, population density, and start vs end of breeding season are factors expected to influence dispersal. Our study showed the importance of dispersal in becoming solitary while there was no indication that aggression by the mother drives natal dispersal.

Aggressive interactions are often considered as the main underlying reason for solitary living and the reduction of aggression as a first step towards the evolution of sociality (Kleiman & Eisenberg, 1973). Dyadic encounters between adult neighbouring bush Karoo rats in a neutral arena were characterised by few interactions, independent of kinship, with sniffing as the predominant form of social investigation. Nearly no aggression occurred, not even between unrelated females. However, close kin were more likely to spend time in body contact with each other than non-kin. Thus, while female bush Karoo rats differentiated between kin and non-kin neighbours, neither relationship was driven by aggression. But there was also no social attraction between adults. We did however observe a high number of chases of striped mice by the bush Karoo rats, mostly in the breeding season (figure in supplementary file), demonstrating that bush Karoo rats can be aggressive. Within the same field site, and in similar experiments to ours, group-living striped mice were highly aggressive towards non-kin and interacted amicably with kin (Schoepf & Schradin, 2012b). We had expected much more aggression in the solitary bush Karoo rat than in the sociable striped mice, and our ethical clearance protocol included that as soon as damaging fighting started, experiments would be terminated. We expected this to be the case regularly when non-kin met, but we never had to stop the encounter experiments. Therefore, absence of social attraction seems to be sufficient to lead to solitary living in bush Karoo rats, without the need for social intolerance and aggression.

Interactions between individuals are not random and instead reflect relatedness or familiarity (Siracusa et al., 2021). In field intruder tests, where we expected to find more aggression towards conspecifics, due to the defence of resources (Wolff & Peterson, 1998), aggression was much more common during the breeding than the non-breeding season, and females were much more aggressive towards non-kin than kin. This indicates that female bush Karoo rats can differentiate between kin and non-kin, even when their kin had been living away from them (solitarily) for several months. Kin recognition thus occurs post-dispersal and indicates that mothers remember adult, dispersed offspring. This tells us that remembering kin (the mechanism for kin recognition) is persistent.

Female territoriality functions to defend resources and offspring (Sinn et al., 2008; Wolff, 2007). This theoretical consideration can explain why we observed nearly no aggression during dyadic encounter tests in a neutral test arena because no resources could be defended but did observe some during intruder tests when individuals defended their lodge. Does this indicate defence of resources such as food and shelter, or defence of offspring? More aggression was observed during the breeding season, which is also when more lodges are available due to low population density at the start of the season, and when food is highly abundant. Thus, our data support the female hypothesis of Wolff & Peterson (1998) that territorial aggression is linked more to the defence of offspring (maternal aggression) than the defence of food resources. Reproductive competition can be high between female mammals (Clutton-Brock et al., 2006), often leading to female infanticide (Hodge et al., 2011), and is considered as one of the main reasons for solitary living (Schradin et al., 2010a). Several other mammal species show maternal aggression, with females being aggressive at their nests in the breeding season while showing amicable behaviour at communal foraging grounds (reviewed by Wolff & Peterson, 1998). For example, female Arctic squirrels (*Urocitellus parryii*) and grey squirrels (*Sciurus carolinensis*) defend their territories near their nests but show considerable overlap in foraging areas. This behaviour is not limited to mammals but has been shown also in female social lizards (e.g., White's skinks, *Egernia whitii*) where female aggression increased during pregnancy and after birth (Sinn et al., 2008). Aggression in female bush Karoo rats thus rather functions to protect their offspring than to establish a solitary social organisation.

Conclusion

While solitary species have been considered to be generally asocial and aggressive, we have evidence that the solitary bush Karoo rat shows low levels of aggression. Instead, its social system is characterised by social tolerance, which suggests that lack of social attraction after dispersal and not aggression leads to solitary living. Female bush Karoo rats were not aggressive to their offspring even after they had dispersed from the maternal lodge and lived solitarily for months. In the solitary bush Karoo rat maternal aggression is not the mechanism driving solitary living. Tolerance of conspecifics is a condition for sociality. Therefore, solitary species that can tolerate their conspecifics may provide a model to understand the evolution of sociality in mammals.

Ethical note

We adhered to the ASAB/ABS Guidelines for the Use of Animals in Research (Buchanan et al., 2012). Bush Karoo rats were captured and handled using protocols approved by the Animal Ethics Screening Committee of the University of the Witwatersrand (AESC clearance number: 2018-03-15B). We further received additional ethical clearance for both the dyadic encounter tests and the field intruder presentation tests (AESC clearance numbers 2021/05/02/B and 22-12-024B respectively).

References

Bekoff, M. (1977). Mammalian dispersal and the ontogeny of individual behavioral phenotypes. *The American Naturalist, 111*(980), 715-732.

Brown, E., & Willan, K. (1991b). Microhabitat selection and use by the bush Karoo rat *Otomys unisulcatus* in the Eastern Cape Province. *South African Journal of Wildlife Research, 21*, 69-75.

Buchanan, K., Burt de Perera, T., Carere, C., Carter, T., Hailey, A., Hubrecht, R., Jennings, D., Metcalfe, N., Pitcher, T., & Peron, F. (2012). Guidelines for the treatment of animals in behavioural research and teaching.

Christian, J. J. (1970). Social subordination, population density, and mammalian evolution. *Science, 168*(3927), 84-90.

Clutton-Brock, T. (2021). Social evolution in mammals. *Science, 373*(6561), eabc9699.

Clutton-Brock, T. H., Hodge, S. J., Spong, G., Russell, A. F., Jordan, N. R., Bennett, N. C., Sharpe, L. L., & Manser, M. B. (2006). Intrasexual competition and sexual selection in cooperative mammals. *Nature, 444*, 1065-1068.

Cooper, L. D., & Randall, J. A. (2007). Seasonal changes in home ranges of the giant kangaroo rat (Dipodomys ingens): a study of flexible social structure. *Journal of Mammalogy, 88*(4), 1000-1008.

Cowling, R. M., Esler, J. J., & Rundel, P. W. (1999). Namaqualand, South Africa - an overview of a unique winter-rainfall desert ecosystem. *Plant Ecology, 142*, 3-21.

du Plessis, A., & Kerley, G. I. H. (1991). Refuge strategies and habitat segregation in two sympatric rodents *Otomys unisulcatus* and *Parotomys brantsii*. *Journal of Zoology, London, 224*, 1-10.

du Plessis, A., Kerley, G. I. H., & Winter, P. E. D. (1992). Refuge microclimates of rodents: a surface nesting *Otomys unisulcatus* and a burrowing *Parotomys brantsii*. *Acta Theriologica, 37*, 351-358.

Eibl-Eibesfeldt, I. (1953). Zur Ethologie des Hamsters (*Cricetus cricetus* L.). *Zeitschrift für Tierpsychologie, 10*(2), 204-254. doi: https://doi.org/10.1111/j.1439-0310.1953.tb00392.x

Elidio, H. d. S. M., Coelho, J. W. R., da Silva, L. C. C. P., & Dos Santos, I. B. (2021). Housing density and aggression in syrian hamsters. *Journal of the American Association for Laboratory Animal Science, 60*(5), 506-509.

Fischer, K., Gebhardt-Henrich, S., & Steiger, A. (2007). Behaviour of golden hamsters (Mesocricetus auratus) kept in four different cage sizes. *Animal welfare, 16*(1), 85-93.

Friard, O., & Gamba, M. (2016). BORIS: a free, versatile open-source event-logging software for video/audio coding and live observations. *Methods in Ecology and Evolution, 7*, 1325-1330. doi: DOI:10.1111/2041-210X.12584

Hinze, A., Rymer, T., & Pillay, N. (2013). Spatial dichotomy of sociality in the African ice rat. *Journal of Zoology, 290*(3), 208-214. doi: 10.1111/jzo.12028

Hodge, S. J., Bell, M. B. V., & Cant, M. A. (2011). Reproductive competition and the evolution of extreme birth synchrony in a cooperative mammal. *Biology Letters, 7*(1), 54-56. doi: 10.1098/rsbl.2010.0555

Jaeggi, A. V., Miles, M. I., Festa-Bianchet, M., Schradin, C., & Hayes, L. D. (2020). Variable social organization is ubiquitous in Artiodactyla and probably evolved from pair-living ancestors. *Proceedings of the Royal Society B: Biological Sciences, 287*(1926), 20200035. doi: doi:10.1098/rspb.2020.0035

Johansson, Ö., Koehler, G., Rauset, G. R., Samelius, G., Andrén, H., Mishra, C., Lhagvasuren, P., McCarthy, T., & Low, M. (2018). Sex-specific seasonal variation in puma and snow leopard home range utilization. *Ecosphere, 9*(8), e02371.

Kappeler, P. M. (2019). A framework for studying social complexity. [journal article]. *Behavioral Ecology and Sociobiology, 73*(1), 13. doi: 10.1007/s00265-018-2601-8

Kappeler, P. M., & Schaik, C. P. v. (2002). Evolution of primate social systems. *International Journal of Primatology, 23*, 707-740.

Kleiman, D., & Eisenberg, J. (1973). Comparisons of canid and felid social systems from an evolutionary perspective. *Animal behaviour, 21*(4), 637-659.

Krause, S., & Schüler, L. (2010). Behavioural and endocrinological changes in Syrian hamsters (Mesocricetus auratus) under domestication. *Journal of Animal Breeding and Genetics, 127*(6), 452-461.

Lukas, D., & Clutton-Brock, T. H. (2013a). The evolution of social monogamy in mammals. *Science, 341*. doi: 10.1126/science.1238677

Makuya, L., Pillay, N., & Schradin, C. (2024b). Kin based spatial structure in a solitary small mammal as indicated by GPS dataloggers. *Animal Behaviour, 215*, 45-54. doi: https://doi.org/10.1016/j.anbehav.2024.06.022

Makuya, L., & Schradin, C. (2024a). Costs and benefits of solitary living in mammals. *Journal of Zoology, 323*, 9-18. doi: https://doi.org/10.1111/jzo.13145

Makuya, L., & Schradin, C. (2024b). The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences, 121*(13). doi: https://doi.org/10.1073/pnas.2402871121

Martin, P., & Bateson, P. P. G. (1993). Measuring behaviour: An introductory guide.

Meshriy, M. G., Randall, J. A., & Parra, L. (2011). Kinship associations of a solitary rodent, Dipodomys ingens, at fluctuating population densities. *Animal Behaviour, 82*(4), 643-650.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., Fonseca, G. A. B. D., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature, 403*, 853-858.

Olivier, C.-A., Martin, J. S., Pilisi, C., Agnani, P., Kauffmann, C., Hayes, L., Jaeggi, A. V., & Schradin, C. (2024). Primate social organization evolved from a flexible pair-living ancestor. *Proceedings of the National Academy of Sciences, 121*(1), e2215401120.

Olivier, C.-A., Martin, J. S., Pilisi, C., Agnani, P., Kaufmann, C., Hayes, L. D., Jaeggi, A., & Schradin, C. (2022b). Primate Social Organization Evolved from a Flexible Pair-Living Ancestor. *bioRxiv*, 2022.2008.2029.505776. doi: 10.1101/2022.08.29.505776

Pillay, N. (2001). Reproduction and postnatal development in the bush Karoo rat Otomys unisulcatus (Muridae, Otomyinae). *Journal of Zoology, 254*(4), 515-520.

Qiu, J., Olivier, C.-A., Jaeggi, A. V., & Schradin, C. (2022). The evolution of marsupial social organization. *Proceedings of the Royal Society B: Biological Sciences, 289*(1985), 20221589. doi: doi:10.1098/rspb.2022.1589

R Core Team. (2022b). R: A Language and Environment for Statistical Computing. Vienna, Austria. Retrieved from https://www.R-project.org/

Roex, N. l., Mann, G. K. H., Hunter, L. T. B., & Balme, G. A. (2022). Relaxed territoriality amid female trickery in a solitary carnivore. *Animal Behaviour, 194*, 225 - 231. doi: https://doi.org/10.1016/j.anbehav.2022.09.022

Schoepf, I., & Schradin, C. (2012a). Better off alone! Reproductive competition and ecological constraints determine sociality in the African striped mouse (*Rhabdomys pumilio*). *Journal of Animal Ecology, 81*(3), 649-656. doi: 10.1111/j.1365-2656.2011.01939.x

Schoepf, I., & Schradin, C. (2012b). Differences in social behaviour between group-living and solitary African striped mice, Rhabdomys pumilio. *Animal Behaviour, 84*(5), 1159-1167. doi: 10.1016/j.anbehav.2012.08.019

Schradin, C. (2004). Territorial defense in a group-living solitary forager: who, where, against whom? *Behavioral Ecology and Sociobiology, 55*(5), 439-446.

Schradin, C. (2005). Nest-Site Competition in Two Diurnal Rodents from the Succulent Karoo of South Africa. *Journal of Mammalogy, 86*(4), 757-762. doi: 10.1644/1545-1542(2005)086[0757:Ncitdr]2.0.Co;2

Schradin, C. (2017). Comparative studies need to rely both on sound natural history data and on excellent statistical analysis. *Royal Society Open Science 4*(9), 170346. doi: http://dx.doi.org/10.1098/rsos.170346

Schradin, C., König, B., & Pillay, N. (2010a). Reproductive competition favours solitary living while ecological constraints impose group-living in African striped mice. *Journal of Animal Ecology, 79*, 515-521.

Sinn, D. L., While, G. M., & Wapstra, E. (2008). Maternal care in a social lizard: links between female aggression and offspring fitness. *Animal Behaviour, 76*(4), 1249-1257. doi: https://doi.org/10.1016/j.anbehav.2008.06.009

Siracusa, E., Boutin, S. A., Dantzer, B., Lane, J. E., Coltman, D. W., & McAdam, A. G. (2021). Familiar neighbors, but not relatives, enhance fitness in a territorial mammal. *Current Biology, 31*, 438-455. doi: 10.1016/j.cub.2020.10.072

Temeles, E. J. (1994). The role of neighbours in territorial systems: when are they `dear enemies`? *Anim Behav, 47*, 339-350.

Twining, J. P., & al., e. (2024). Using global remote camera data of a "solitary" species complex to evaluate the drivers of group formation. *PNAS*.

Valomy, M., Hayes, L. D., & Schradin, C. (2015a). Social organization in Eulipotyphla: evidence for a social shrew. *Biology Letters, 11*(11). doi: 10.1098/rsbl.2015.0825

Wolff, J. O. (2007). Social biology of rodents. *Integrative Zoology, 2*(4), 193-204. doi: 10.1111/j.1749-4877.2007.00062.x

Wolff, J. O., & Peterson, J. A. (1998). An offspring-defense hypothesis for territoriality in female mammals. *Ethology Ecology and Evolution, 10*, 227- 239.

Supplementary tables

Table S1: Sample sizes for the number of sniffing events and the time spent in fights, grooming and body contact between mothers and offspring during dyadic encounter tests grouped by season and the age of the offspring.

Breeding season					
Age of offspring (in months)	*Number of individuals*	*Mean number of sniffing events (tests with events)*	*Mean duration of fights (min), (tests with aggression)*	*Mean duration of grooming (min),*	*Mean duration of body contact (min),*

				(tests with grooming)	*(tests with body contact)*
1	22	5.545 (N=20)	0.003 (N=2)	0.206 (N=5)	6.385 (N=18)
2	7	6.714 (N=3)	0.015 (N=1)	0.071 (N=2)	3.655 (N=6)
3	5	3.6 (N=3)	0 (N=0)	0.018 (N=1)	1.190 (N=4)
6	1	2 (N=1)	0 (N=0)	0 (N=0)	10.279 (N=1)
8	1	1 (N=1)	0 (N=0)	0 (N=0)	0 (N=0)
10	1	1 (N=1)	0 (N=0)	0.079 (N=1)	10.152 (N=1)
12	1	0 (N=0)	0 (N=0)	0 (N=0)	0 (N=0)
14	2	0 (N=0)	0 (N=0)	0 (N=0)	0 (N=0)
Means (Totals)	40	2.482 (N=29)	0.002 (N=3)	0.047 (N=9)	3.958 (N=30)
	STDEV	2.549	0.005	0.072	4.456
Non-breeding season					
Age of offspring (in months)	*Number of individuals*	*Mean number of sniffing events (tests with events)*	*Mean duration of fights (min), (tests with aggression)*	*Mean duration of grooming (min), (tests with grooming)*	*Mean duration of body contact (min), (tests with body contact)*
3	8	5.375 (N=5)	0.019 (N=1)	0.054 (N=2)	3.451 (N=8)
4	8	1.625 (N=2)	0.026 (N=2)	0.014 (N=1)	4.494 (N=5)
5	12	3.167 (N=6)	0.003 (N=1)	0.077 (N=4)	3.943 (N=8)
6	5	3.6 (N=5)	0 (N=0)	0.087 (N=1)	5.771 (N=5)
7	4	8.5 (N=1)	0 (N=0)	0 (N=0)	1.576 (N=3)
18	1	3 (N=1)	0 (N=0)	0.092 (N=1)	5.354 (N=1)
20	1	0 (N=0)	0 (N=0)	0 (N=0)	0.496 (N=1)
Means (Totals)	39	3.61 (N=20)	0.007 (N=4)	0.046 (N=9)	3.584 (N=31)
	STDEV	2.725	0.011	0.041	1.935

Model selection

Table S2: AIC-based model selection results examining reparameterization of age of offspring vs whether offspring had dispersed or not.

body contact	**K**	**-2logL**	**AIC**	**ΔAIC**	**ωAIC**
dispersed	6	-232.99	479.16	0	1
age	17	-226.42	496.88	17.73	0

fight	**K**	**-2logL**	**AIC**	**ΔAIC**	**ωAIC**
dispersed	6	167.41	-321.66	0	1
age	17	169.65	-295.28	26.38	0
sniff	**K**	**-2logL**	**AIC**	**ΔAIC**	**ωAIC**
dispersed	5	-236.93	484.68	0	1
age	16	-231.41	503.59	18.91	0
groom	**K**	**-2logL**	**AIC**	**ΔAIC**	**ωAIC**
dispersed	6	-25.2	63.57	0	1
age	17	-31.35	106.73	43.15	0

Table S3. Sniffing: Results of the ANOVA models to test whether dispersal and the season have an impact on the frequency of sniffing events in the bush Karoo rat.

Sniff = season + dispersed

Predictors	*df*	*Sum sqs*	F value	*p*
Season	1	18	0.723	0.398
Dispersed	1	49.2	1.978	0.164
Season : Dispersed	1	20.5	0.822	0.368

Table S4. Fighting: Results of the ANOVA model to test whether dispersal and the season have an impact on the time spent fighting in the bush Karoo rat.

Fighting = season + dispersed

Predictors	*df*	*Sum sqs*	F value	*p*
Season	1	0.00061	0.678	0.41
Dispersed	1	0.0015	1.656	0.2
Weight difference	1	0.00014	0.15	0.7

Table S5 Grooming: Results of the ANOVA models to test whether dispersal and the season have an impact on the time spent grooming in the bush Karoo rat. Factors in bold type indicate significant predictors.

Grooming = season + dispersed

Predictors	*df*	*Sum sqs*	F value	*p*
Season	1	0.00459	0.054	0.82
Dispersed	1	0.1487	1.754	0.19
Weight difference	1	0.4105	4.841	**0.03**

Table S6. Body contact: Results of the ANOVA models to test whether dispersal and the season have an impact on the time spent in body contact in the bush Karoo rat.

Body contact = season + dispersed

Predictors	*df*	*Sum sqs*	F value	*p*
Season	1	0.0236	0.0014	0.97
Dispersed	1	7.647	0.4487	0.51
Weight difference	1	24.6384	1.4457	0.23

Table S7. Neighbor encounter tests - Fighting: Results of the linear mixed effects models to identify which factors influenced time spent in amicable and aggressive behaviours by bush Karoo rats during dyadic encounter tests with neighbours.

Fight = season + relatedness + body size difference + ID-focal (random).

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	-0.007	0.018	-0.043 – 0.029	0.692
Season (non-breeding)	0.016	0.013	-0.0098 – 0.042	0.220
Relatedness (non-kin)	0.009	0.015	-0.02 – -0.04	0.551
Weight difference	0.0003	0.001	-0009 – -0.001	0.641
Random effects σ2/ICC	0.000024/0.009			
Marginal *R*2/conditional *R*2	0.03/0.039			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S8. Neighbor encounter tests – Body contact: Results of the linear mixed effects models to identify which factors influenced time spent in amicable and aggressive behaviours by bush Karoo rats during dyadic encounter tests with neighbours. Factors in bold type indicate significant predictors.

Body contact = season + relatedness + body size difference + ID-focal (random).

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	4.27	1.496	1.26 – 7.29	< **0.01**
Season (non-breeding)	0.57	1.094	-1.63 – 2.80	0.60
Relatedness (non-kin)	-2.36	1.316	-5.04 – 0.31	0.08
Weight difference	-0.02	0.048	-0.12 – 0.08	0.69
Random effects σ2/ICC	5.078/0.26			
Marginal *R*2/conditional *R*2	0.06/0.305			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S9. Neighbor encounter tests - Grooming: Results of the linear mixed effects models to identify which factors influenced time spent in amicable and aggressive behaviours by bush Karoo rats during dyadic encounter tests with neighbours.

Grooms = season + relatedness + body size difference + ID-focal (random).

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	0.04	0.026	-0.0097 – 0.095	0.09
Season (non-breeding)	0.008	0.019	-0.0295 – 0.045	0.673
Relatedness (non-kin)	-0.02	0.022	-0.0645 – 0.025	0.356
Weight difference	-0.0002	0.001	-0.0019 – 0.002	0.793
Random effects σ2/ICC	0.000049/0.009			
Marginal	0.02/0.029			

*R*2/conditional *R*2				

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S10. Sniffing events during encounter tests: Results of the generalized linear mixed effects models to identify which factors influenced the frequency of sniffing events exhibited by bush Karoo rats during dyadic encounter tests towards neighbours. Factors in bold type indicate significant predictors.

*Sniff = season * relatedness + body size difference + ID-focal (random) + ID-stimulus(random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	0.42	0.35	-0.29 – 1.08	0.22
Season (non-breeding)	0.73	0.27	0.22 – 1.29	< **0.01**
Relatedness (non-kin)	0.54	0.36	-0.17 – 1.27	0.14
Weight difference	-0.02	0.008	-0.04 - -0.004	**0.02**
Season (non-breeding) : Relatedness (non-kin)	-0.63	0.43	-1.49 – 0.21	0.14
Random effects *σ*2/ICC	2.10/0.83			
Marginal *R*2/conditional *R*2	0.061/0.839			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S11. Latency to first aggression during intruder tests: Results of the linear mixed effects models to identify which factors influenced the latency to the first aggressive behaviour with a stimulus during intruder tests towards neighbours.

*Latency to first aggression = season * relatedness + body size difference + ID-focal (random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	4.286	1.69	0.08 – 7.67	< **0.01**
Season (non-breeding)	4.287	1.82	0.61 – 7.95	**0.02**

Relatedness (non-kin)	-3.06	1.64	-6.47 – 0.24	0.07
Weight difference	0.05	0.03	-0.0091 – 0.118	0.08
Season (non-breeding) : Relatedness (non-kin)	1.52		-2.59 – 5.65	0.46
Random effects σ2/ICC	11.07 /0.52			
Marginal *R*2/conditional *R*2	0.348/0.688			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S12. Latency to approach cage during intruder tests: Results of the linear mixed effects models to identify which factors influenced the latency to approach a cage with a stimulus during intruder tests towards neighbours.

*Latency to approach = season * relatedness + body size difference + ID-focal (random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	0.89	1.70	-2.68 – 4.32	0.60
Season (non-breeding)	2.88	1.82	-0.94 – 6.69	0.12
Relatedness (non-kin)	-0.98	1.61	-4.27 – 2.29	0.55
Weight difference	0.04	0.03	-0.02 – 0.01	0.17
Season (non-breeding) : Relatedness (non-kin)	3.02	1.97	-1.01 – 7.06	0.14
Random effects σ2/ICC	13.22/0.556			
Marginal *R*2/conditional *R*2	0.236/0.661			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S13. Charging events during intruder tests: Results of the generalized linear mixed effects models to identify which factors influenced the frequency of charging events exhibited by bush Karoo rats during intruder tests towards neighbours. Factors in bold type indicate significant predictors.

*Charging = season * relatedness + body size difference + ID-focal (random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	1.55	0.28	0.97 – 2.16	< **0.001**
Season (non-breeding)	-1.92	0.44	-2.899 - -1.10	< **0.001**
Relatedness (non-kin)	0.75	0.22	0.32 – 1.20	< **0.001**
Weight difference	-0.0056	0.0049	-0.016 – 0.004	0.26
Season (non-breeding) : Relatedness (non-kin)	0.54	0.39	-0.21 – 1.37	0.17
Random effects σ2/ICC	0.41/0.604			
Marginal *R*2/conditional *R*2	0.620/0.849			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S14. Number of trill sounds during intruder tests: Results of the linear mixed effects models to identify which factors influenced the number of trill sounds with a stimulus during intruder tests towards neighbours.

*Trills = season * relatedness + body size difference + ID-focal (random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	21.997	14.1	-4.73 – 48.83	0.13
Season (non-breeding)	-9.66	16.94	-41.62 – 22.76	0.57
Relatedness (non-kin)	9.53	18.2	-25.45 – 44.77	0.61
Weight difference	-0.38	0.27	-0.90 – 0.13	0.17
Season (non-breeding) : Relatedness (non-kin)	7.48	23.25	-37.60 – 52.53	0.75
Random effects σ2/ICC	101.6/0.65			

Marginal *R*2/conditional *R*2	0.121/0.178			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Table S15. Time spent at a cage during intruder tests: Results of the linear mixed effects models to identify which factors influenced the time spent at a cage of the stimulus during intruder tests. Factors in bold type indicate significant predictors.

*Time at cage = season * relatedness + body size difference + ID-focal (random).*

Predictors	*Estimates*	*SE*	95% CI	*p*
(Intercept)	5.08	1.25	2.54 – 7.59	**< 0.001**
Season (non-breeding)	-2.11	1.42	-4.95 – 0.75	0.14
Relatedness (non-kin)	4.63	1.37	1.76 – 7.39	**< 0.01**
Weight difference	-0.03	0.02	-0.07 – 0.024	0.30
Season (non-breeding) : Relatedness (non-kin)	-1.93	1.71	-5.43 – 1.59	0.27
Random effects *σ*2/ICC	4.38/0.354			
Marginal *R*2/conditional *R*2	0.381/0.600			

σ2, variance of the random effect "ID"; ICC, intraclass coefficient of variation.

The marginal R2 considers only the variance of the fixed effects, while the conditional R2 takes both the fixed and random effects into account.

Supplementary figures

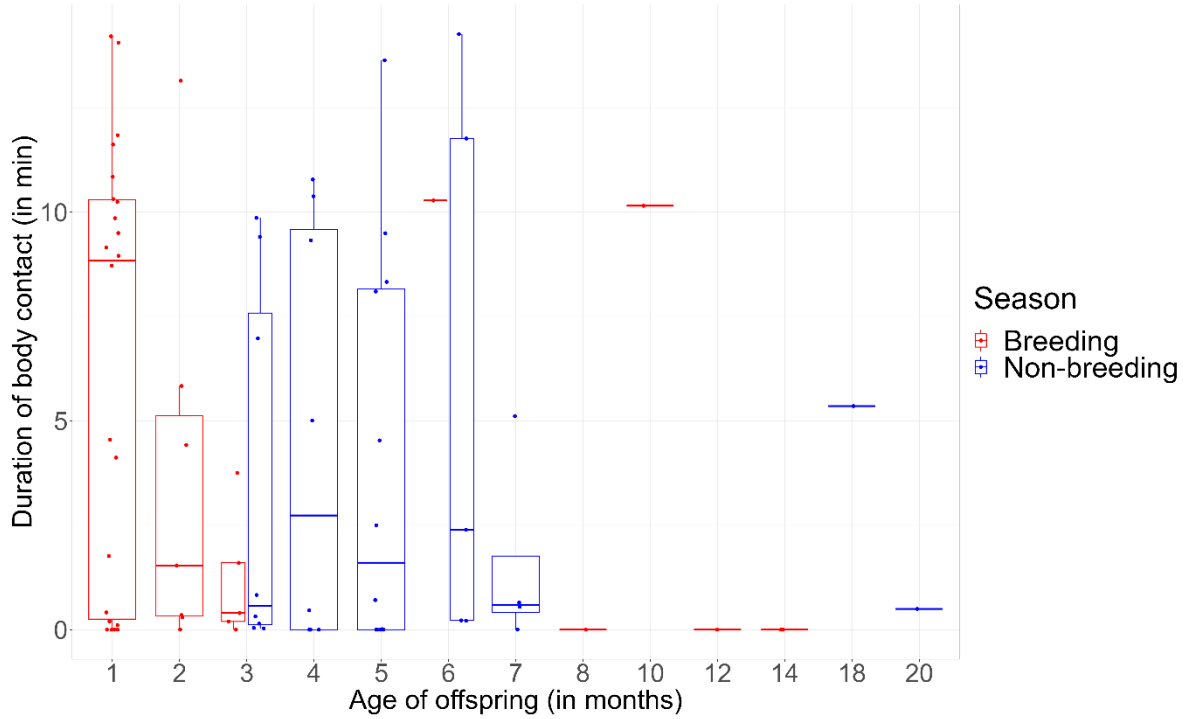

Figure S 1: The duration spent in body contact by female bush Karoo rats according to the age of the offspring, for both seasons. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values (breeding: n = 40, non-breeding: n = 39).

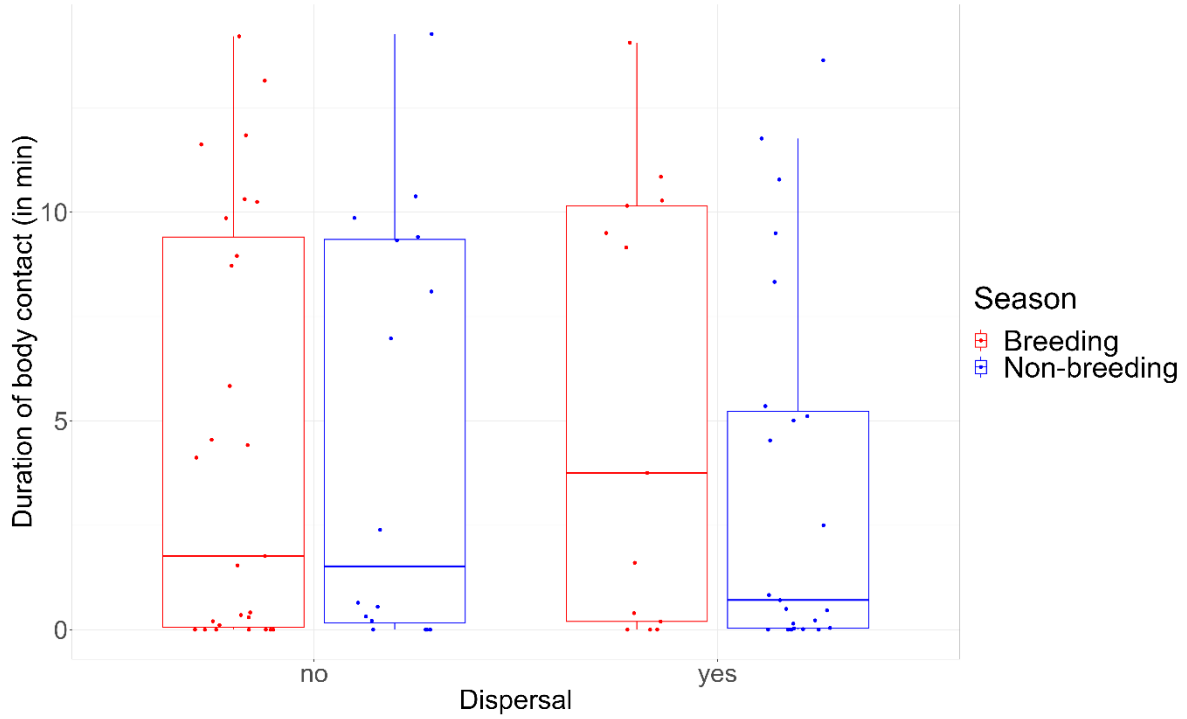

Figure S 2: The duration spent in body contact by female bush Karoo rats according to whether offspring had dispersed or not, for both seasons. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values (breeding: n = 40, non-breeding: n = 39).

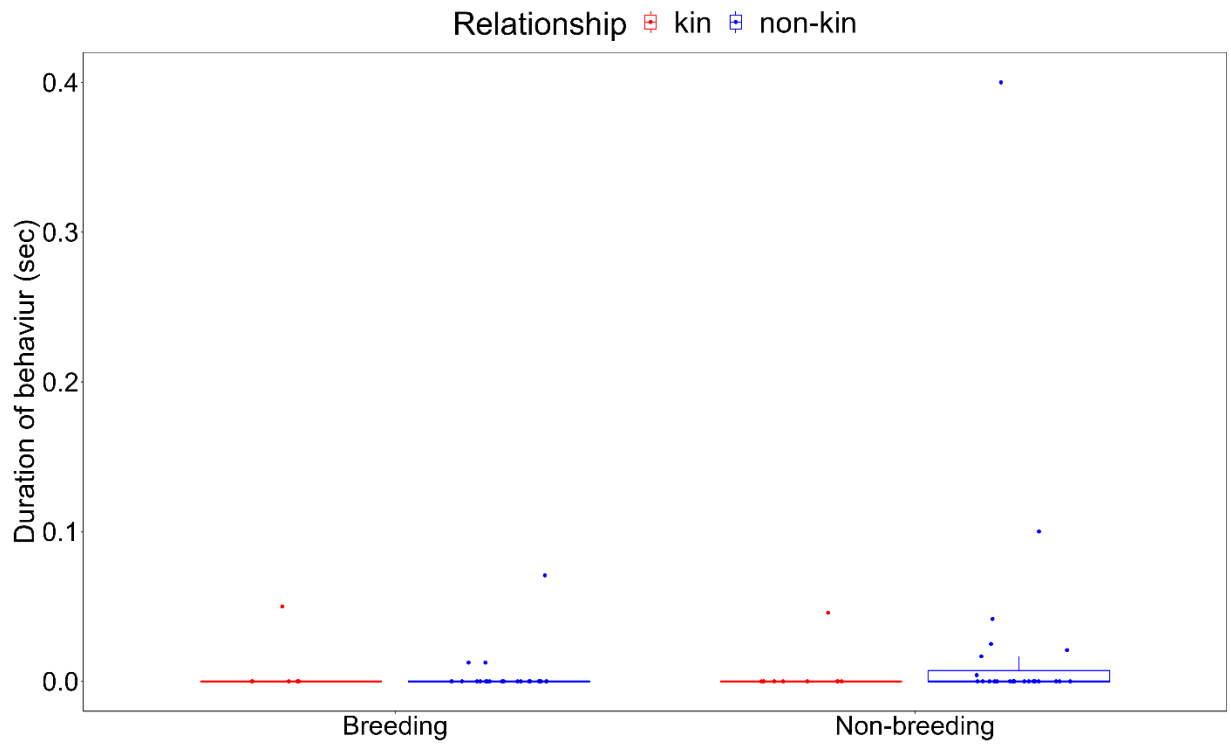

Figure S 3: The duration spent fighting by female bush Karoo rats for both seasons. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values (breeding: n = 40, non-breeding: n = 39)

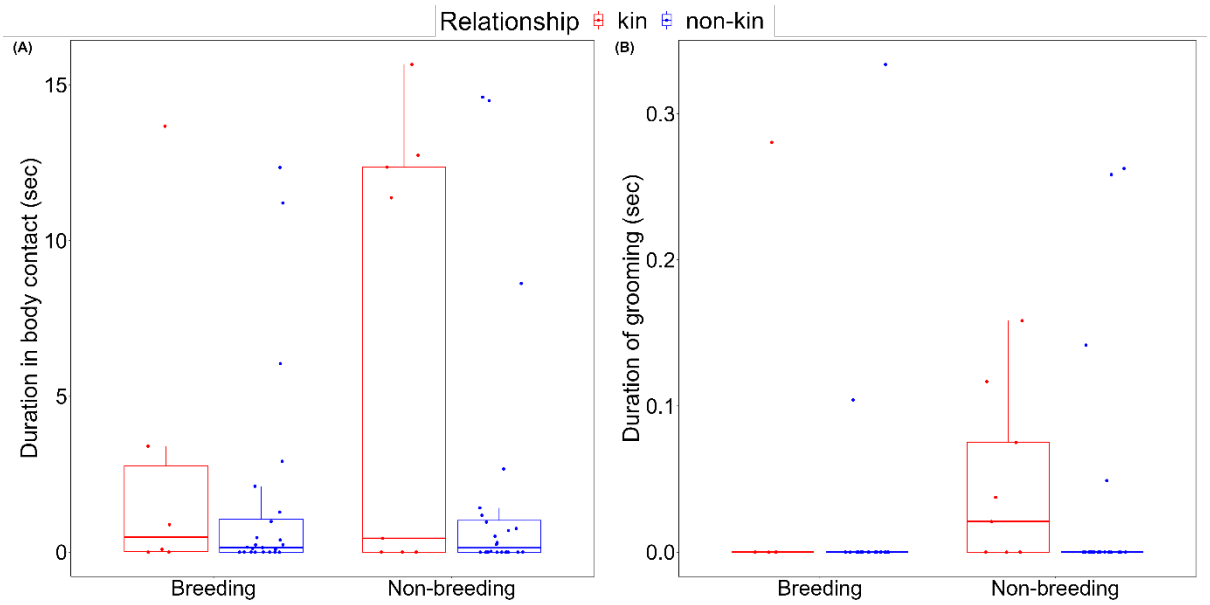

Figure S 4: The duration of body contact (A) and grooming (B) behaviours between neighbouring female bush Karoo rats during dyadic encounter tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values.

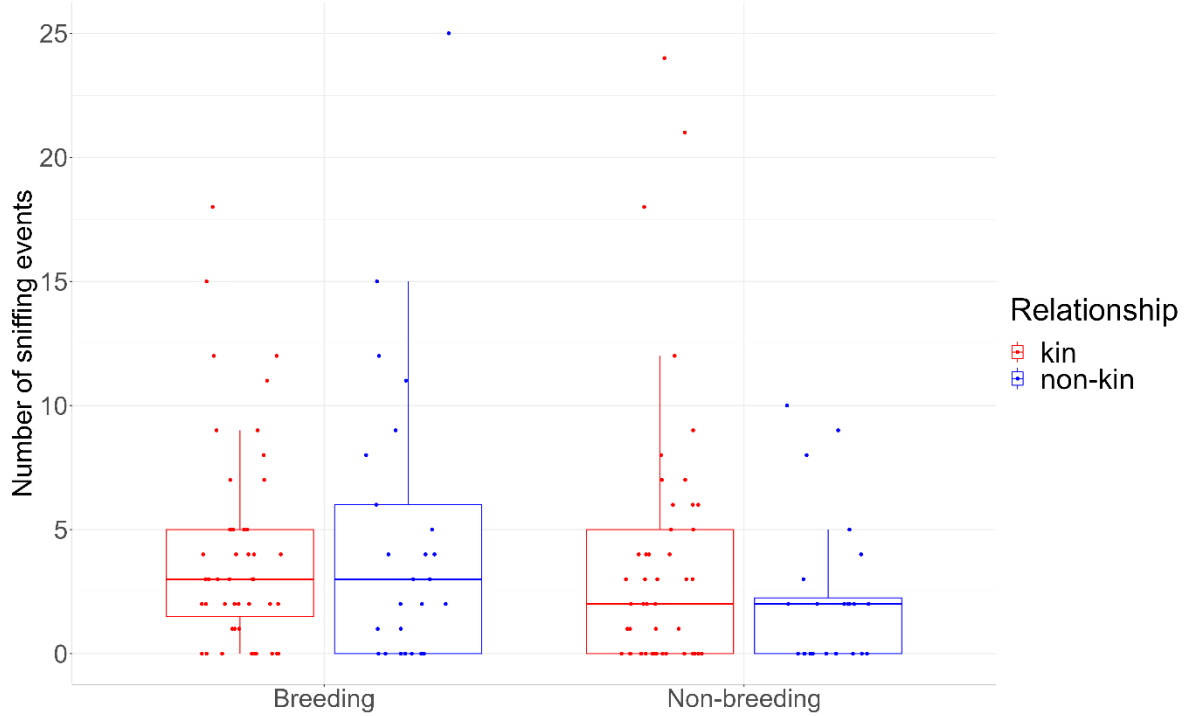

Figure S 5: The number of sniffing events between neighbouring female bush Karoo rats during dyadic encounter tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values.

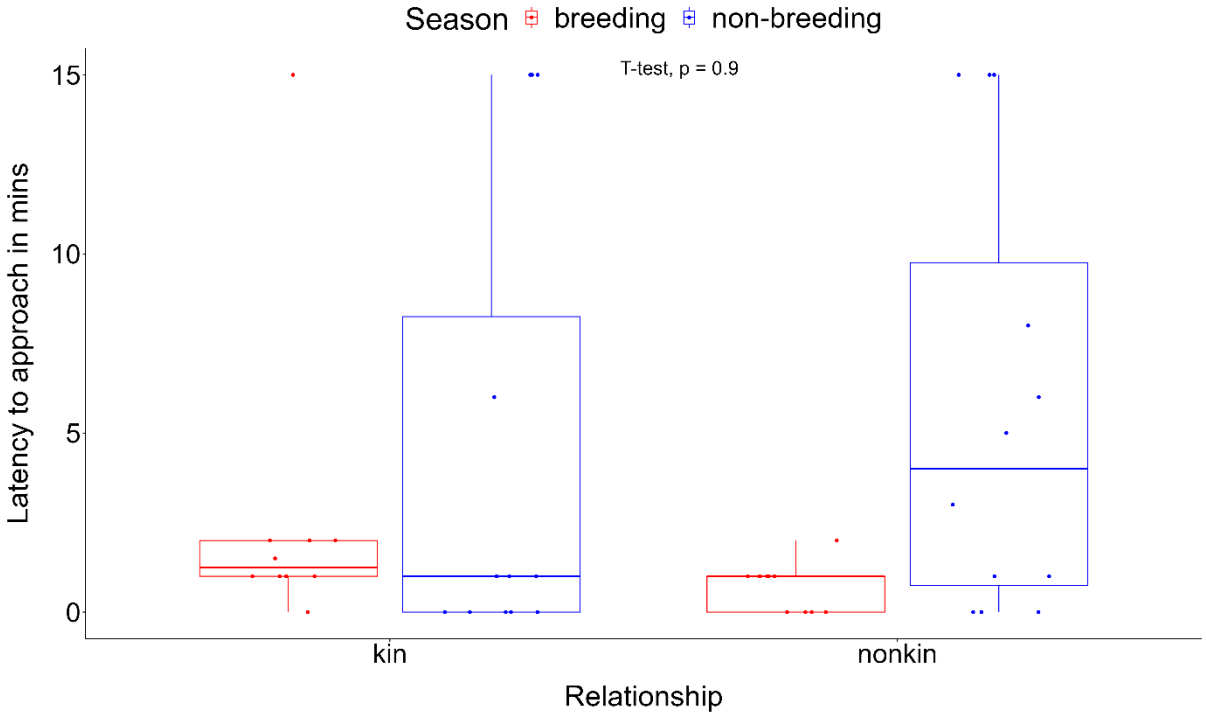

Figure S 6: A measure of the latency to approach a cage during intruder tests based on the relationship between individuals. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values.

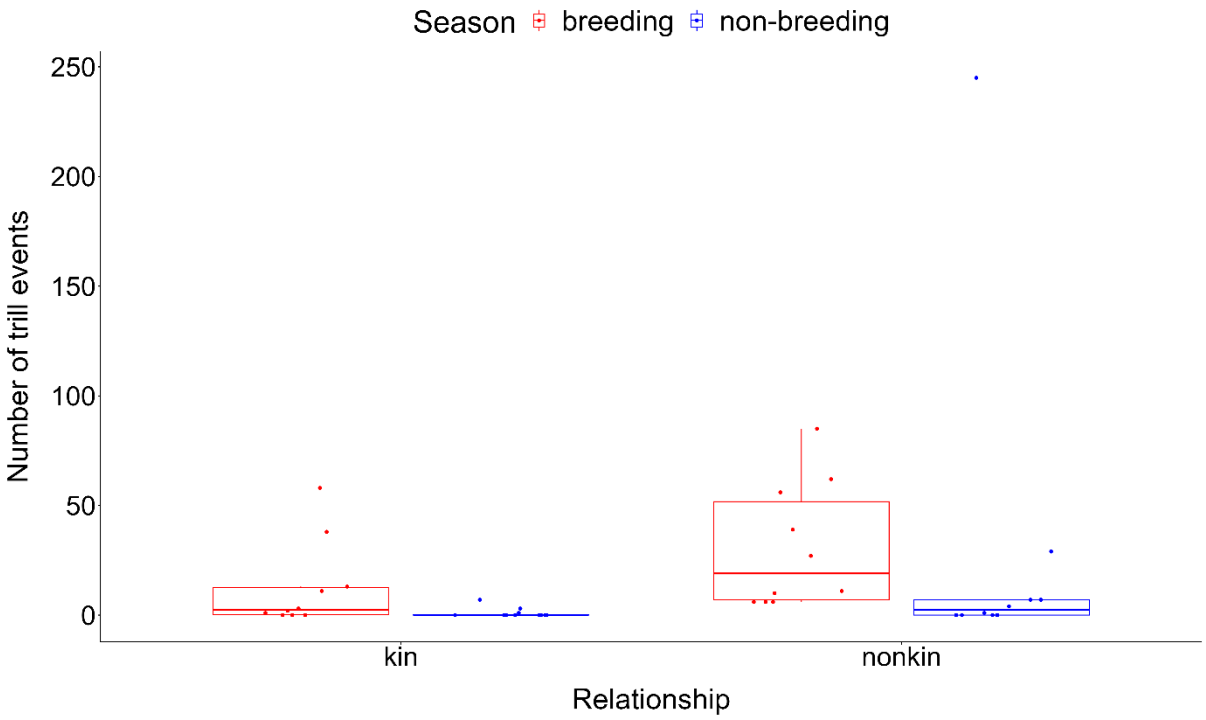

Figure S 7: The number of trilling events measured during intruder tests. Boxplots show median and 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values.

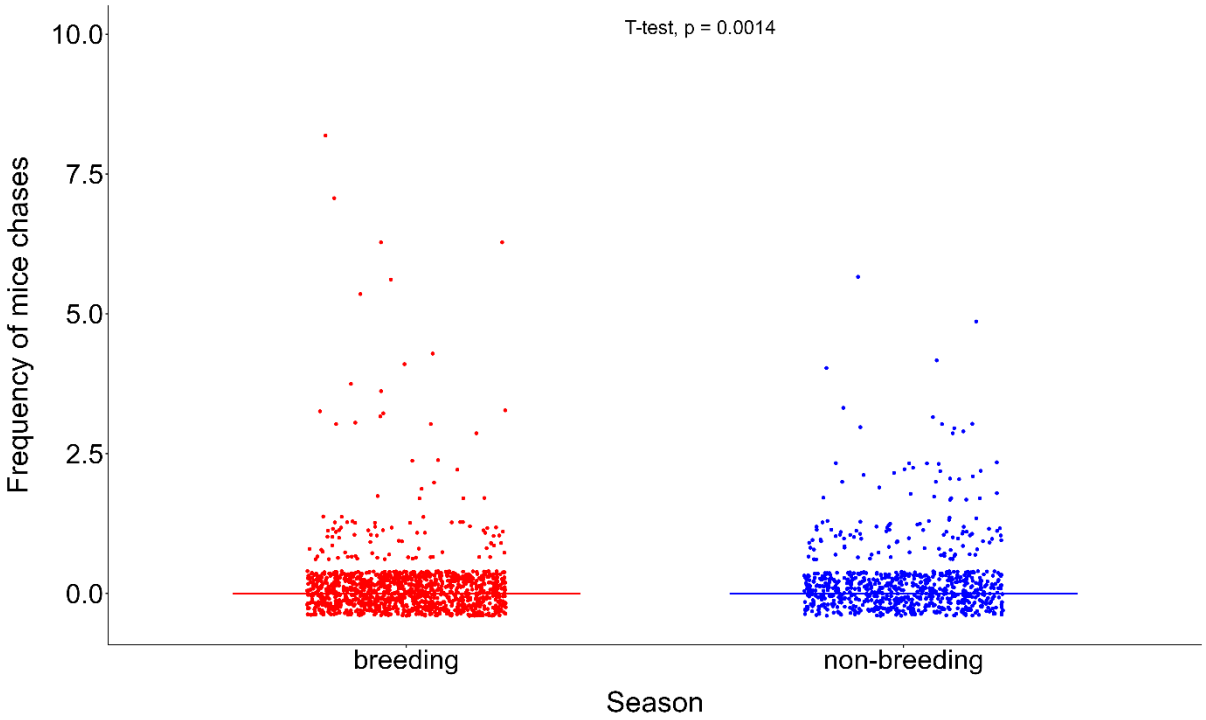

Figure S 7: The frequency of chasing events against striped mice.

Additional tables on models run using age of offspring as a variable.

Table S3.1. Sniffing: Results of the ANOVA models to test whether age of offspring and the season have an impact on the frequency of sniffing events in the bush Karoo rat.

Sniff = season + age of offspring

Predictors	*df*	F value	*p*
Season	1	0.711	0.402
Age offspring (months)	12	1.044	0.422
Season : Age offspring (months)	1	0.001	0.978

Table S4.1. Fighting: Results of the ANOVA model to test whether age of offspring and the season have an impact on the time spent fighting in the bush Karoo rat.

Fighting = season + age of offspring

Predictors	*df*	F value	*p*
Season	1	0.619	0.438

Age offspring (months)	12	0.418	0.953
Weight difference	1	1.101	0.298

Table S5.1 Grooming: Results of the ANOVA models to test whether age of offspring and the season have an impact

on the time spent grooming in the bush Karoo rat.

Grooming = season + age of offspring

Predictors	*df*	F value	*p*
Season	1	0.029	0.865
Age offspring (months)	12	0.105	0.9999
Weight difference	1	0.732	0.396

Table S6.1. Body contact: Results of the ANOVA models to test whether age of offspring and the season have an impact on the time spent in body contact in the bush Karoo rat.

Body contact = season + age of offspring

Predictors	*df*	F value	*p*
Season	1	0.171	0.68
Age offspring (months)	12	1.244	0.27
Weight difference	1	0.138	0.71

CHAPTER 6. Shared feeding grounds in a solitary rodent: an indication for cooperation by kin selection and mutualism?

Preprint available at: Makuya, L[1]., Pillay, N[1]., & Schradin, C[1,2]. (2024). Shared foraging grounds in a solitary rodent: an indication for cooperation by kin selection and mutualism? bioRxiv, 2024.2007.2007.602396. doi: 10.1101/2024.07.07.602396.

Authors' affiliations:

[1] School of Animal, Plant & Environmental Sciences, University of the Witwatersrand, Private Bag 3, WITS 2050, Johannesburg, South Africa

[2] IPHC, UNISTRA, CNRS, 23 rue du Loess, 67200 Strasbourg, France

Abstract

Kinship is important for understanding the evolution of social behaviour in group living species. However, even solitary living individuals differentiate between kin and non-kin neighbours, which could lead to some form of cooperation, defined as both partners benefitting from each other. A simple form of cooperation is mutualism, where both partners benefit simultaneously. Here we tested whether there is mutual tolerance by sharing foraging grounds between kin in a solitary species. This would indicate the possibility of kin selection and mutual cooperation. We used mini-GPS data loggers to investigate range overlap in the solitary bush Karoo rat (*Otomys unisulcatus*) between kin- and non-kin neighbours. Next, we quantified the extent to which individuals shared foraging grounds containing food plants within their overlapping ranges. Lastly, using step selection functions applied to GPS fixes collected every five minutes, we analysed how individuals moved relative to each other. Kin-neighbours had larger home range overlap than non-kin neighbours (70.4% vs 29.6%) and shared more of their foraging grounds (63% vs 37%). Temporal analysis of spatial data found no indication that neighbours avoided each other, independent of kinship. Instead, activity was synchronised. In sum, we found mutual tolerance between neighbours with regards to sharing foraging grounds, and kin shared nearly double as much of their foraging grounds than non-kin. These data can be interpreted as a simple way of mutual cooperation between kin in a solitary species, where both members benefit from sharing a considerable part of their foraging grounds.

Keywords: by-product mutualism; cooperation; fine-scale interaction; solitary; space use

Introduction

Solitary living species are not asocial, but often have non-random social interactions between neighbours which might even share resources (Makuya & Schradin, 2024b). Solitary living refers to the social organisation (Kappeler & Schaik, 2002) of a species where adult individuals live alone (Makuya & Schradin, 2024a). Historically, it has been assumed that solitary living is caused by intra-specific aggression, functioning to monopolise resources (e.g. European hamster; *Cricetus cricetus*) (Eibl-Eibesfeldt, 1953). However, a growing number of studies show that there are also amicable interactions between neighbours in solitary species, for example in mustelids (Twining et al., 2024), the African leopard (*Panthera pardus*) (Roex et al., 2022) and in estuarine crocodiles (*Crocodylus porosus*) (Baker et al., 2021). In the solitary puma (*Puma concolor*), individuals meeting at carrion respond in a non-random and consistent way, with some individuals always chasing each other aggressively, while other individuals commonly feed together (Elbroch et al., 2016; Elbroch & Quigley, 2016). Whether such amicable sharing of food sources also occurs in solitary herbivores has not been studied in detail, though studies on kin selection and overlap in the kangaroo rats (*Dipodomys ingens*) and Columbian ground squirrels (*Urocitellus columbianus*) suggest that sharing foraging grounds is possible (Dobson et al., 2012; Meshriy et al., 2011).

The distribution of resources, such as food, influences the spatial distribution of animals (Johnson et al., 2002). The resource dispersion hypothesis states that if rare resources rare and evenly distributed, animals will be less likely to aggregate because there is no benefit for resource defence (Johnson et al., 2002; Macdonald & Johnson, 2015; Macdonald, 1983). In contrast, rich patchily distributed resources favour the formation of groups that defend these resources. Food distribution has been used to explain spatial structure of solitary species. For example, small, patchy food patches resulted in within group scramble competition and solitary living in the lemur *Microcebus berthae* (Dammhahn & Kappeler, 2009). In contrast, the closely related lemur *M. murinus* relied on large patchily distributed food resources, thereby allowing females to overlap their home ranges and to form sleeping groups (Dammhahn & Kappeler, 2009). While large patchily distributed resources drive group living in rodents (Ebensperger, 2001), small patchily distributed food might drive solitary living to monopolize resources.

The distribution of resources influences social interactions between individuals (Halliwell et al., 2016). To understand the influence of resource distribution on social interactions, one must consider how individuals overlap spatially and temporally (Rodrigues, 2018; Tania et al., 2012). Using step-selection

functions on data obtained from GPS tracking, the movement of an individual can be related to the dynamic occurrence distribution of another individual, proving an estimation of fine-scale interactions (Schlägel et al., 2019; Ward et al., 2021). With this method one can determine whether individuals are attracted to conspecifics, avoid them or tolerate them. Fine-scale interactions have been quantified for bank voles (*Myodes glareolus*), where the males were always strongly attracted to females, while females were attracted to the males only when in oestrous but avoided them otherwise, maybe to avoid male infanticide (Schlägel et al., 2019). Similarly, the movements of male elongated tortoises (*Indotestudo elongata*) were driven by the presence of females (Ward et al., 2021). Both studies considered mates as a driver of the attraction of between individuals (Schlägel et al., 2019; Ward et al., 2021). Such fine-scale interactions between individuals could also provide insights into behavioural interactions underlying cooperation.

Cooperation between conspecifics has mainly been studied in group living species (Clutton-Brock, 2016). The most basic definition of cooperation is that it consists of an interaction between two individuals that benefits both (Bergmüller et al., 2007a). Cooperation can evolve easily when both individuals benefit at the same time, referred to as by-product mutualism (Bergmüller et al., 2007a). An example can be seen in group living animals who gain safety from predators by forming large groups (Taborsky et al., 2021). These animals cooperate by joint predator surveillance, whereby individuals take turns in watching for predators while the others forage, for example sentinels in meerkats (Clutton-Brock, 2016). In such cases, where one individual first has costs before it later benefits, cooperation is either based on mutual reciprocity (Trivers, 1971) or on kin selection leading to indirect fitness benefits (Hamilton, 1971). Simple forms of cooperation might occur in solitary species that have a kin-based spatial structure with overlap in home ranges and frequent non-random interactions (Makuya et al., 2024b; Meshriy et al., 2011). Mutual tolerance is a requirement for cooperation and might even occur in solitary species, especially between close kin, adding indirect fitness benefits.

Most solitary mammals are either nocturnal, small and live in dense habitats, or very large predators roaming over large areas. Thus, field-based studies of species on both extremes are challenging. In contrast, the solitary and folivorous bush Karoo rat (*Otomys unisulcatus*) can be easily observed in its natural habitat (Makuya et al., 2024b; Schradin, 2005). Its endemic to semi-arid regions of South Africa and builds stick-lodges inside shrubs. It is a central-place forager, foraging within a short distance (less than 25m) from its lodge to patches of leafy succulent plants and ephemeral herbs, called foraging grounds, and then returning to the lodge (Schradin, 2005). Runways lead from the lodge to the foraging grounds. Female bush Karoo rats form kin clusters, living closer to kin neighbours than to non-kin neighbours (Makuya et al., 2024b). This makes it likely that foraging grounds of close kin overlap. Social

interactions between close kin are characterized by tolerance while interactions between non-kin are more often aggressive, indicating a kin based amicable social structure (Makuya et al., 2024a). The bush Karoo rat is therefore a good model to test the extent to which a solitary species shares patches of food and whether there is indication of kin selection and mutualism.

Here we asked whether the kin based spatial structure (Makuya et al., 2024b) in combination with the amicable social structure (Makuya et al., 2024a) might enable some simple form of cooperation (by-product mutualism) in the solitary bush Karoo rat. Specifically, we predicted (i) that foraging grounds will overlap more between kin- than non-kin neighbours, (ii) that neighbouring bush Karoo rats show positive interaction at foraging grounds and a (iii) higher temporal overlap of kin- compared to non-kin neighbours, indicating that kin use the same food resource at the same time while non-kin avoid each other.

Methods

Study site

Our study was conducted on a 4.5-hecare field site next to the Succulent Karoo Research Station located in the Goegap Nature Reserve, Northern Cape, South Africa (29°41’41.17” S 17°53’34.91” E). The area is a biodiversity hotspot, characterized by Succulent Karoo vegetation (Cowling et al., 1999; Myers et al., 2000). The climate is arid with minimum temperatures of -3°C in winter and maximum temperatures up to 43°C in summer (Schradin, 2005). Mean annual precipitation at the field site is 160mm per annum. Seasons are characterised as the hot dry non-breeding season (November to April) and the cool moist breeding season (May to October).

Trapping

Our field site was divided into six areas. Two areas were trapped simultaneously for three days by two people, before moving on to the next two areas. We used a combination of foldable Sherman traps and locally produced heavy traps of the Sherman style. Traps were placed at lodges that showed signs of being occupied (presence of fresh plant material, fresh faeces and active runways) by bush Karoo rats. All trapped rats were processed to obtain information on their weight (to the nearest gram) and their reproductive status (vagina perforated or closed; males scrotal or not). We conducted experiments using females as male bush Karoo rats disperse in autumn. We determined the kinship of the rats from trapping data: juveniles and pups were assumed to be the offspring of the adult female occupying the lodge where

they were trapped; pups / juveniles were always trapped at the same lodge and during the breeding season only one adult female was ever trapped at a lodge at a time (Makuya et al., 2024b). Upon first capture, rats were marked with metal band ear tags that had a unique identification number (National Band and Tag Co., Newport, KY, U.S.A.) (Schoepf & Schradin, 2012a). Rats were additionally marked with non-toxic hair dye to aid in individual identification during focal animal observations (Inecto Rapido, Pinetown, South Africa).

Behavioural observations in the field

Focal observations were conducted five times a week at lodges with evidence of being occupied by bush Karoo rats to determine which individuals occupied the lodge (current study) and collect behavioural data for other studies (Makuya et al., 2024a). The morning observations started five minutes before the sun started shining on the lodges and lasted for 30 minutes; afternoon observations of 30 minutes started 25 minutes before the sun stopped shining on the lodges. Three to five lodges were observed on the same day by different observers. Each lodge was observed for at least two days a month.

Measuring daily ranges

Neighbouring adult female bush Karoo rats were simultaneously fitted with mini-GPS dataloggers (Gipsy 6 model from Technosmart, Italy https://www.technosmart.eu/gipsy-remote-copy/), from August 2021 to April 2023. We fitted 227 Gipsy collars on 43 days on a total of 125 females (on average, 4.6 ± 1.5 females were fitted simultaneously, range 2 to 7 females). The mini-GPS loggers weighed 4.5g including the protective coating, which is less than 5 % of the rats' body weight. These loggers were programmed to collect daily home range data according to methods validated by Makuya & Schradin (2023) and used by Makuya et al. (2024b). The mini-GPSs were fitted like a rucksack with a chest and a waist belt consisting of a wire running through a small rubber tube to avoid any skin irritation, fastened with a fishing crimp. The loggers collected the location (longitude, latitude, and altitude) of an individual and the date and time. We programmed the mini-GPS loggers to be off for the first two days (habituation) and start data collection on day 3 every 5 minutes; the battery life did not allow us to collect data for more than one day. We then re-trapped animals to remove the collars and to extract the data. Our method allowed us to assess whether movements of neighbouring rats are synchronised and to generate accurate estimate of distance travelled. A pilot study by Makuya showed that this method leads to similar home range sizes compared to using traditional method of collecting data every two hours for five days. The data were cleaned by

first removing waypoints that had low accuracy (> 50m) and outliers that were beyond 50m away from the centre.

Measuring home range quality

Bush Karoo rats have runways leading from their lodges to foraging areas. We measured food availability along runways used by all female bush Karoo rats from which we had determined home ranges from the mini-GPS data. We ran a measuring tape along every runway within the ranges (Figure 1). Every 20cm, we recorded plants growing at 25cm and 50cm at right angles from the runways, both to the left and to the right. We recorded whether these plants were dead or alive. We further determined whether the plants were food plants based on data from nest observations. When there was no plant, a zero was recorded. For each runway, we counted the total number of food plants and associated the number of food plants to the runways that were then shared between the rats as determined from the GPS data.

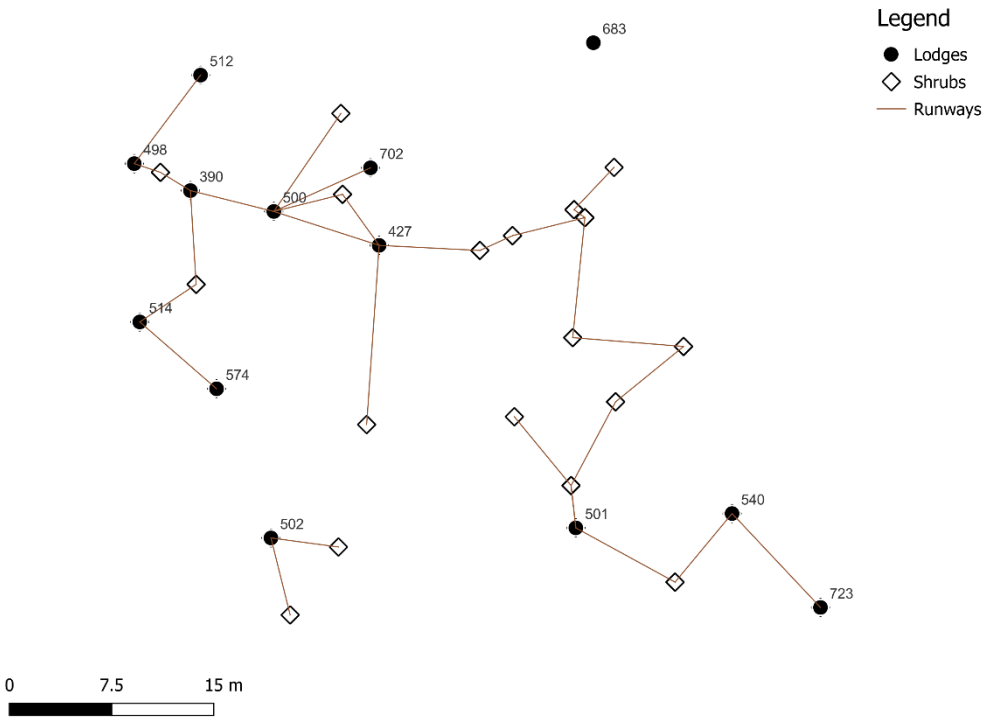

Figure 1. Example of lodges with runways. The hollow diamonds are the shrubs without lodges, black circles are lodges and brown lines are the runways between the lodges and shrubs.

Statistical analysis

To analyse the daily range sizes and overlap, we used the autocorrelated Kernel density estimation (AKDE) implemented in the R packages *ctmm* (Calabrese et al., 2016) and *ctmmweb* (Dong et al., 2018). Overlap was calculated for every pair of rats (N = 192) on the daily range data from the same day. For the pairs of individuals that overlapped (N = 148), we estimated the interactions between individuals based on step-selection functions (SSF) in a workflow code supplied by Schlägel et al. (2019). This method allowed us to investigate whether the movement of one rat was correlated with the movement of its neighbour with which it overlapped. From this, we obtained a coefficient of interaction and p-value indicating whether there was attraction (positive value and $P < 0.05$; the rat chose locations where its neighbour also occurred), avoidance (negative value and $P < 0.05$: the rat avoided locations where its neighbour occupies) and neutral (close to zero coefficient: the rats' choice of locations was independent of its neighbour's occurrence). We then ran a general linear model (GLM) to analyse the factors that influence the coefficient of interaction. For this, we fitted the model (equation 1 below) using the fixed effects estimated overlap, relationship of the neighbour (kin vs non-kin) and season (breeding vs non-breeding) to determine whether these also had an impact on the response.

1. *Coefficient of interaction = overlap + relationship + season*

We determined the amount of food shared for all pairs of bush Karoo rats that overlapped in their ranges. For each pair, we determined which runways they shared and then added up all food plants counted along these runways. To analyse the factors that influenced the total shared food, we fitted a generalised linear mixed model (GLMM) with a Poisson distribution and a square root link function, run in the *lme4 R* package using the g*lmer* function (Bates et al., 2015), with the relationship (kin vs non-kin), season (breeding vs non-breeding) and the percentage of overlap fit as fixed effects. We also fitted the ID of both bush Karoo rats as random effects (equation 2 below).

2. *Shared food = relationship + season + overlap+ (1|ID focal) + (1|ID neighbour)*

Results

The home ranges of bush Karoo rats were larger in the moist breeding season (649.5 ± 424.3 m^2, N = 82) than in the dry non-breeding season (447.6 ± 254.5 m^2, N = 67; estimate= -405.4, SE = 446.5, p = 0.37). Their home ranges overlapped more with kin neighbours than non-kin neighbours (dry season: 49.8 ± 6.2 % vs 26.9 ± 4.7 % and wet season: 43.8 ± 5.3 % vs 21.6 ± 4.2 %) (relationship; estimate = -22.5, SE 5.3, $P < 0.001$; season; estimate = -5.7, SE = 5.3, $P = 0.28$).

Overlap of foraging grounds

As predicted, we found higher overlap of foraging grounds between kin neighbours than non-kin neighbours (63% vs 37%, $P < 0.001$), especially during the dry, non-breeding season (dry season 66%, 38.5 ± 7.6 food plants for kin vs 33%, 26.04 ± 7.5 for non-kin; wet season 61%, 30.6 ± 6.9 vs 39%, 22.3 ± 6.1; $P = 0.001$, Table 1, Figure 2). The amount of shared food increased with increasing range overlap ($P < 0.001$; Figure 3).

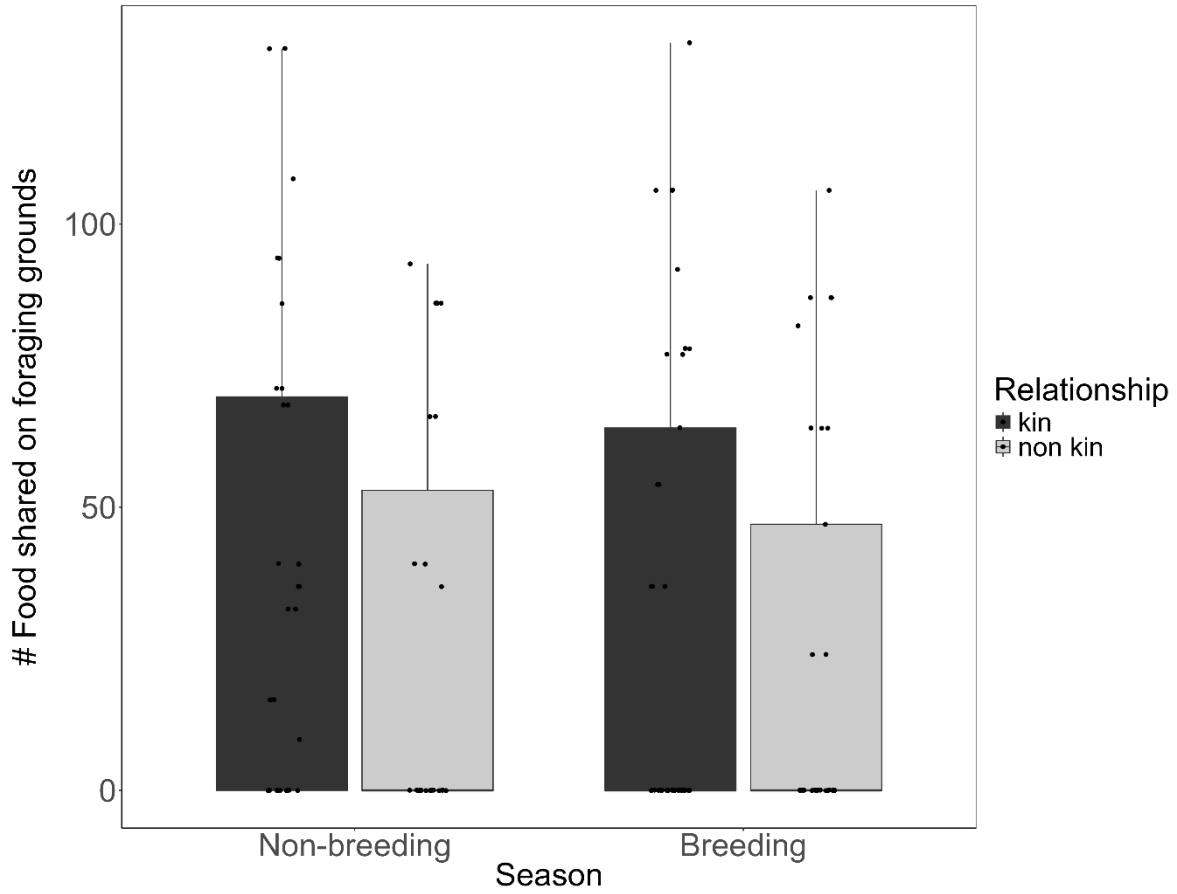

Figure 2. Seasonal comparison of total food shared between bush Karoo rats at foraging grounds by kinship. Boxplots show 1st and 3rd quartiles, the whiskers represent the minimum and maximum of the outlier data and points represent individual values.

Table 1. Summary of Poisson GLMM to model the number of food plants shared by neighbouring bush Karoo rats.

Model parameter	Estimate	SE	*P*
Intercept	-0.29	0.63	0.64
Overlap	0.069	0.004	**<0.001**
Relationship (non-kin)	1.85	0.33	**<0.001**

Season (breeding)	0.55	0.17	**0.001**

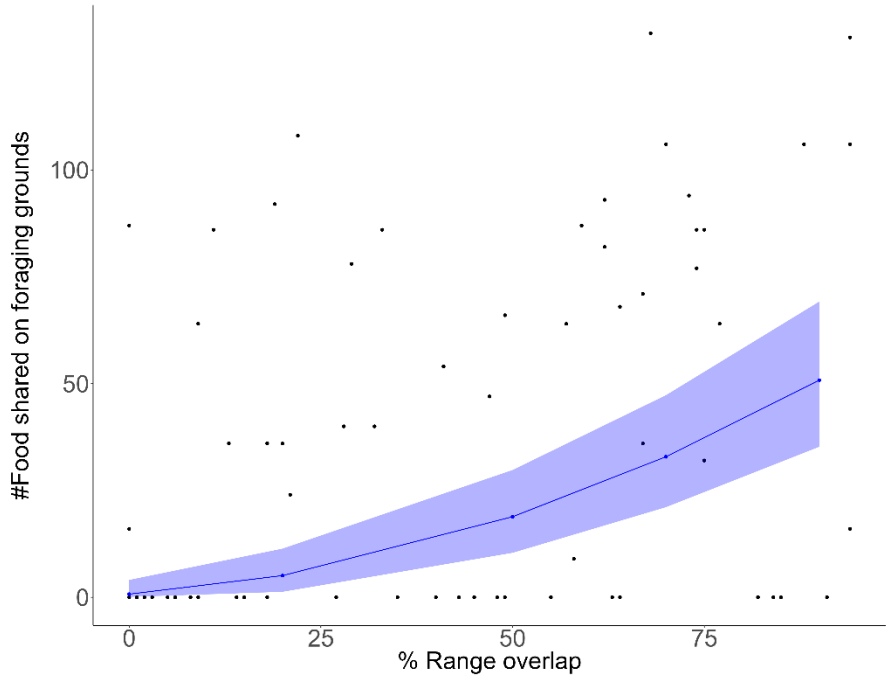

Figure 3. Mean fitted number of food plants within foraging grounds (solid line) and 95% confidence intervals (shaded area) against range overlap. Black dots are observed data.

Temporal overlap (estimated step-selection coefficient of interaction behaviour)

The ranges of 148 pairs of bush Karoo rats overlapped between 2022 and 2023. From 90 of these pairs, we obtained useable temporal overlap estimates; for the remaining 58 pairs, the models either did not converge, or the model failed to calculate the coefficient of interaction because individuals only marginally overlapped. A total of 82 pairs were attracted to each other (coefficients greater than one), with 64 of these pairs being significantly attracted to each other ($P < 0.05$). Only three pairs avoided each other (negative coefficients) but not significantly ($P > 0.05$). Five pairs showed neutrality which was significant for only one pair ($P < 0.05$). We removed four outliers that had more than double the mean value for the coefficient of interaction. We therefore ran the GLM model for 86 pairs.

The coefficient of interaction was significantly and negatively influenced by the degree of range overlap (GLM; $t = -5.60$, $P < 0.001$; Table 2 and Figure 4), i.e., rats which had a low overlap were more attracted to the same foraging grounds (higher coefficient) while rats which overlapped more were less or not attracted (neutral or negative attraction, coefficient close to zero). Unlike in our prediction, neither season

nor relationship had a significant effect on the coefficient of interaction (GLM; $t = 0.17$, $P = 0.869$ and $t = -0.33$, $P = 0.741$ respectively).

Table 2. Summary of Gaussian GLM to model factors influencing the estimated step-selection coefficient (relative selection strength) of interaction behaviour in bush Karoo rats.

Model parameter	Estimate	SE	*P*
Intercept	8.7442	1.1378	**<0.001**
Overlap	-8.3224	1..4862	**<0.001**
Relationship (non-kin)	-0.2914	0.8779	0.741
Season (breeding)	0.1446	0.8757	0.869

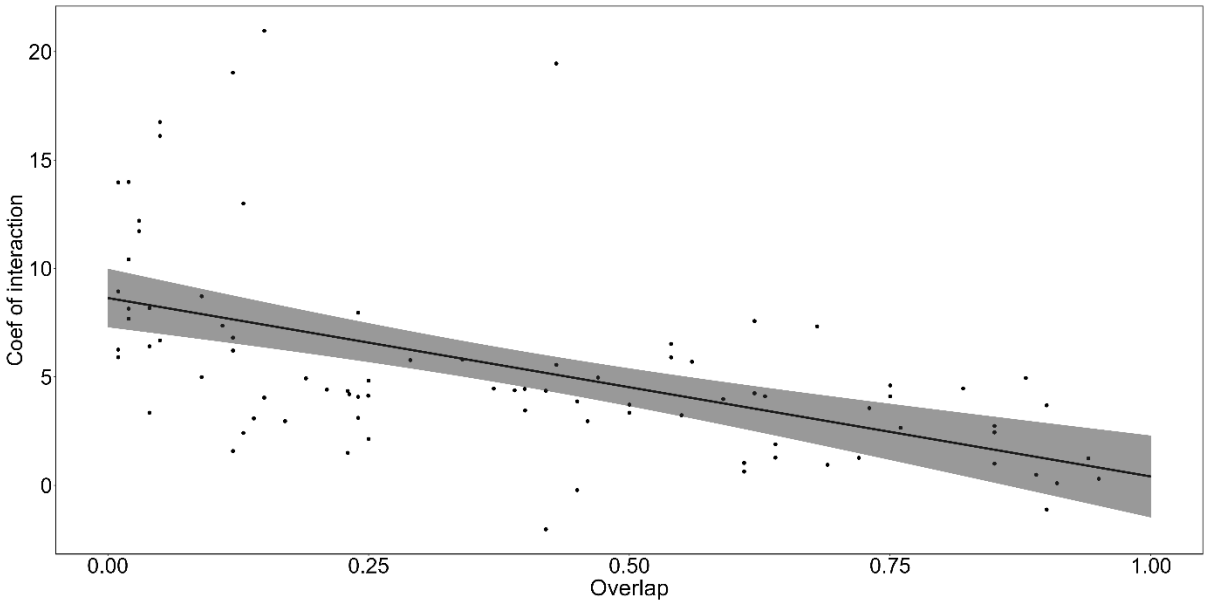

Figure 4. Mean fitted coefficient of interaction (solid line) and 95% confidence intervals (shaded area) against estimated range overlap. Black dots are observed data.

Discussion

We tested whether closely related females of the solitary bush Karoo rat shared foraging grounds in a mutualistic way. Female bush Karoo rat shared more foraging grounds with their kin neighbours than their non-kin neighbours, and this especially in the food restricted dry season. They were attracted to each other at the foraging grounds, and we found no indication that they avoided each other. Therefore, they shared the restricted food resource mutualistically and especially between close kin, which can be interpreted as a simple form of cooperation.

The home ranges of female bush Karoo rats overlapped and so did their foraging grounds. We showed in a previous study that the overlap in home ranges was mostly driven by kinship (Makuya et al., 2024b).

Here we replicated this result: female kin shared nearly 50% of their home ranges, while overlap with non-kin was only half as much. Overlap in home ranges also lead to a significant overlap in foraging grounds. Female bush Karoo rat shared 61% of their food with their kin neighbours but only 39% with their non-kin neighbours in the moist season. In the food restricted dry season, when one might expect individuals to monopolize food and overlap thus to decrease, this difference was even stronger, and females shared 66% of their food with close kin but only 33% with non-kin.

Individuals can use the same resources without sharing them, for example by using them at different times of the day, or by avoiding each other and only visiting a resource when it its free. Therefore, we calculated interaction scores between dyads to assess whether they avoided each other. Interactions scores were positive for 90% of dyads independently of kinship, which could mean one of two things: either they were attracted to each other or they were attracted to the foraging grounds, an essential resource (Spiegel et al., 2016). Bush Karoo rats are central place foragers, which forage a short distance from their lodges to the foraging grounds. Previously we reported that they have as many kin as non-kin neighbours (Makuya et al., 2024b) but behave differently to each of them. Female bush Karoo rats react with tolerance or are even amicable towards close kin, but are aggressive towards non-kin (Makuya et al., 2024a). Together these results indicate that the positive interaction scores were rather due to attraction to the food within the foraging grounds (local enhancement) than due to social attraction. If it would have been social attraction, we would have expected positive association between kin and avoidance between non-kin; also, in encounter tests we did not find animals were attracted by each other (Makuya et al., 2024a). However, they did not avoid each other but visited the foraging grounds during the same time of the day, leading to positive interaction coefficients.

Bush Karoo rats shared more foraging grounds the more their home ranges overlapped, but at the same time their interactions scores declined, even though remaining positive in most cases. This indicates that they increased the distance between individuals when sharing larger areas. Thus, this effect might not indicate reduced competition but simply a statistical effect: when they share larger areas, by chance they are more often far away from each other.

Season had no influence on the interaction score, but on the number of shared food in the foraging grounds. If sharing of food is simply a consequence of overlapping areas with high food availability (food being an abundant resource that does not need to be defended), one would have expected them to share more foraging grounds in the food rich moist season and to monopolise food when it becomes rare by decreasing overlap. Surprisingly, we found the opposite: more overlapping foraging grounds in the food restricted dry season. This further indicates that tolerance between female bush Karoo rats did not decrease when food became scarce. Indeed, in another study, we observed that aggression between

neighbouring females is higher in the food rich breeding season compared to the dry season, probably to avoid reproductive competition such as female infanticide (Makuya et al., 2024a). This indicates that food is only a minor driver of competition and explains why foraging grounds can be shared in a mutualistic way.

By-product mutualism is a form of cooperation where both parties benefit at the same time (Bergmüller et al., 2007b). However, it is only regarded as a form of cooperation if it includes an interaction between the two parties (Bergmüller et al., 2007b), especially if the behaviour involved was under selection for this purpose (West et al., 2007). Whether mutual sharing of foraging grounds in bush Karoo rats represents a simple form of cooperation is not completely evident: while both parties potentially benefit from having access to the foraging grounds, this is not characterised by interactions but rather by social tolerance allowing both individuals to exploit this resource. Importantly, tolerance is not a random response but represent an absence of aggressive interactions. This indicates that tolerance is an evolved trait that avoids aggressive interactions and could thus also be regarded as a kind of neutral interaction. For example, in solitary carnivores, some individuals do regularly tolerate each other when foraging at carcasses, but aggressively chase away other individuals from the same food source (Elbroch & Quigley, 2016; Roex et al., 2022; Twining et al., 2024). Some degree of tolerance is necessary for all group-living species, which do not automatically classify as cooperative because they share foraging grounds, but typically only when they directly interact to share food (Clutton-Brock, 2002). Thus, in several solitary mammals, individual-specific tolerance for mutual sharing of food resources has evolved, often based on kinship, indicating early forms of by-product mutualism

Conclusion

In conclusion, the spatial structure has an influence on the social interactions between individuals. Specifically, solitary bush Karoo rats overlapped more with their kin neighbours and therefore shared more foraging grounds, without avoiding each other. This adds to the growing number of studies that indicate that solitary species can have non-random social interactions with their neighbours. Attraction to the same foraging grounds leads to the sharing of food, especially between kin, and the mutual tolerance of each other together with the synchronised behaviour could be interpreted as an interaction that benefits both, or at least as the absence of an aggressive interaction to monopolise food, and as such as a simple form of cooperation. Such forms of cooperation could be the starting point for the evolution of more complex forms of cooperation including amicable interactions.

References

Baker, C. J., Frère, C. H., Franklin, C. E., Campbell, H. A., Irwin, T. R., & Dwyer, R. G. (2021). Crocodile social environments dictated by male philopatry. *Behavioral Ecology, 33*(1), 156-166. doi: 10.1093/beheco/arab120

Bates, D., Maechler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. . *Journal of Statistical Software, 67*(1), 1-48. doi: 10.18637/jss.v067.i01.

Bergmüller, R., Johnstone, R. A., Russell, A. F., & Bshary, R. (2007a). Integrating cooperative breeding into theoretical concepts of cooperation. *Behavioural Processes, 76*, 61- 72.

Bergmüller, R., Russell, A. F., Johnstone, R. A., & Bshary, R. (2007b). On the further integration of cooperative breeding and cooperation theory. *Behavioural Processes, 76*, 170- 181.

Calabrese, J. M., Fleming, C. H., & Gurarie, E. (2016). *ctmm*: An r package for analyzing animal relocation data as a continuous-time stochastic process. *Methods in Ecology and Evolution, 7*(9), 1124-1132. doi: https://doi.org/10.1111/2041-210x.12559

Clutton-Brock, T. (2002). Breeding together: kin selection and mutualism in cooperative vertebrates. *Science, 296*, 69-72.

Clutton-Brock, T. (2016). *Mammal societies*. Chichester: Sons.

Cowling, R. M., Esler, J. J., & Rundel, P. W. (1999). Namaqualand, South Africa - an overview of a unique winter-rainfall desert ecosystem. *Plant Ecology, 142*, 3-21.

Dammhahn, M., & Kappeler, P. M. (2009). Females go where the food is: does the socio-ecological model explain variation in social organisation of solitary foragers? *Behavioral Ecology and Sociobiology, 63*, 939–952. doi: 10.1007/s00265-009-0737-2

Dobson, F. S., Viblanc, V. A., Arnaud, C. M., & Murie, J. O. (2012). Kin selection in Columbian ground squirrels: direct and indirect fitness benefits. *Molecular ecology, 21*(3), 524-531.

Dong, X., Fleming, C., Noonan, M., & Calabrese, J. (2018). Ctmmweb: A Shiny Web App for the Ctmm Movement Analysis Package.: https://github.com/ctmm-initiative/ctmmweb.

Ebensperger, L. A. (2001). A review of the evolutionary causes of rodent group-living. *Acta Theriologica, 46*, 115-144.

Eibl-Eibesfeldt, I. (1953). Zur Ethologie des Hamsters (*Cricetus cricetus* L.). *Zeitschrift für Tierpsychologie, 10*(2), 204-254. doi: https://doi.org/10.1111/j.1439-0310.1953.tb00392.x

Elbroch, L. M., Lendrum, P. E., Quigley, H., & Caragiulo, A. (2016). Spatial overlap in a solitary carnivore: support for the land tenure, kinship or resource dispersion hypotheses? *Journal of Animal Ecology, 85*(2), 487-496. doi: https://doi.org/10.1111/1365-2656.12447

Elbroch, L. M., & Quigley, H. (2016). Social interactions in a solitary carnivore. *Current Zoology, 63*, zow080. doi: 10.1093/cz/zow080

Halliwell, B., Uller, T., Wapstra, E., & While, G. M. (2016). Resource distribution mediates social and mating behavior in a family living lizard. *Behavioral Ecology, 28*(1), 145-153. doi: 10.1093/beheco/arw134

Hamilton, W. D. (1971). Geometry for the selfish herd. *J Theor Biol, 31*, 295-311.

Johnson, D. D. P., Kays, R., Blackwell, P. G., & Macdonald, D. W. (2002). Does the resource dispersion hypothesis explain group living? *TRENDS in Ecology and Evolution, 17*, 563-570.

Kappeler, P. M., & Schaik, C. P. v. (2002). Evolution of primate social systems. *International Journal of Primatology, 23*, 707-740.

Macdonald, D., & Johnson, D. (2015). Patchwork planet: the resource dispersion hypothesis, society, and the ecology of life. *Journal of Zoology, 295*(2), 75-107.

Macdonald, D. W. (1983). The ecology of carnivore social behaviour. *Nature, 301*(5899), 379-384.

Makuya, L., Pillay, N., Sangweni, S. P., & Schradin, C. (2024a). Tolerant mothers: aggression does not explain solitary living in the bush Karoo rat. *bioRxiv*, 2024.2006. 2024.600450. doi: https://doi.org/10.1101/2024.06.24.600450

Makuya, L., Pillay, N., & Schradin, C. (2024b). Kin based spatial structure in a solitary small mammal as indicated by GPS dataloggers. *Animal Behaviour, 215*, 45-54. doi: https://doi.org/10.1016/j.anbehav.2024.06.022

Makuya, L., & Schradin, C. (2023). Measuring range sizes in a 100-g rodent: mini-GPS are more reliable than transmitters, but the location error reduces reliability. *Mammalian Biology, 103*(5), 455-465. doi: https://doi.org/10.1007/s42991-023-00365-4

Makuya, L., & Schradin, C. (2024a). Costs and benefits of solitary living in mammals. *Journal of Zoology, 323*, 9-18. doi: https://doi.org/10.1111/jzo.13145

Makuya, L., & Schradin, C. (2024b). The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences, 121*(13). doi: https://doi.org/10.1073/pnas.2402871121

Meshriy, M. G., Randall, J. A., & Parra, L. (2011). Kinship associations of a solitary rodent, Dipodomys ingens, at fluctuating population densities. *Animal Behaviour, 82*(4), 643-650.

Myers, N., Mittermeier, R. A., Mittermeier, C. G., Fonseca, G. A. B. D., & Kent, J. (2000). Biodiversity hotspots for conservation priorities. *Nature, 403*, 853-858.

Rodrigues, A. M. M. (2018). Resource availability and adjustment of social behaviour influence patterns of inequality and productivity across societies. *PeerJ, 6*, e5488. doi: 10.7717/peerj.5488

Roex, N. l., Mann, G. K. H., Hunter, L. T. B., & Balme, G. A. (2022). Relaxed territoriality amid female trickery in a solitary carnivore. *Animal Behaviour, 194*, 225 - 231. doi: https://doi.org/10.1016/j.anbehav.2022.09.022

Schlägel, U. E., Signer, J., Herde, A., Eden, S., Jeltsch, F., Eccard, J. A., & Dammhahn, M. (2019). Estimating interactions between individuals from concurrent animal movements. *Methods in Ecology and Evolution, 10*(8), 1234-1245.

Schoepf, I., & Schradin, C. (2012a). Better off alone! Reproductive competition and ecological constraints determine sociality in the African striped mouse (*Rhabdomys pumilio*). *Journal of Animal Ecology, 81*(3), 649-656. doi: 10.1111/j.1365-2656.2011.01939.x

Schradin, C. (2005). Nest-Site Competition in Two Diurnal Rodents from the Succulent Karoo of South Africa. *Journal of Mammalogy, 86*(4), 757-762. doi: 10.1644/1545-1542(2005)086[0757:Ncitdr]2.0.Co;2

Spiegel, O., Leu, S. T., Sih, A., & Bull, C. M. (2016). Socially interacting or indifferent neighbours? Randomization of movement paths to tease apart social preference and spatial constraints. *Methods in Ecology and Evolution, 7*(8), 971-979.

Taborsky, M., Cant, M. A., & Komdeur, J. (2021). *The evolution of social behaviour*: Cambridge University Press.

Tania, N., Vanderlei, B., Heath, J. P., & Edelstein-Keshet, L. (2012). Role of social interactions in dynamic patterns of resource patches and forager aggregation. *Proceedings of the National Academy of Sciences, 109*(28), 11228-11233. doi: doi:10.1073/pnas.1201739109

Trivers, R. L. (1971). The evolution of reciprocal altruism. *The Quarterly Review of Biology, 46*, 35- 57.

Twining, J. P., Sutherland, C., Zalewski, A., Cove, M. V., Birks, J., Wearn, O. R., Haysom, J., Wereszczuk, A., Manzo, E., & Bartolommei, P. (2024). Using global remote camera data of a solitary species complex to evaluate the drivers of group formation. *Proceedings of the National Academy of Sciences, 121*(12), e2312252121.

Ward, M., Marshall, B. M., Hodges, C. W., Montano, Y., Artchawakom, T., Waengsothorn, S., & Strine, C. T. (2021). Nonchalant neighbors: Space use and overlap of the critically endangered Elongated Tortoise. *Biotropica, 53*(5), 1342-1355.

West, S. A., Griffin, A. S., & Gardner, A. (2007). Social semantics: altruism, cooperation, mutualism, strong reciprocity and group selection. *J Evol Biol, 20*, 415- 432.

CHAPTER 7. General Discussion

Animal societies are described by considering four components of the social system (Kappeler, 2019; Kappeler & van Schaik, 2002): (1) their social organisation, which is the composition of social units within a population or species; (2) the social structure that describes social interactions between individuals; (3) the mating system (who mates and reproduces with whom); and (4) the care system (who takes care of the offspring). All four components are inter-related (Figure 1) and each can have multiple categories (Kappeler, 2019). For example, animals can live in six different forms of social organisation (Figure 1). This includes solitary (e.g. Cape dune mole-rat, *Bathyergus suillus* (Bray et al., 2012)), pair (e.g. Etendeka round-eared elephant shrew, *Macroscelides micus* (Rathbun & Dumbacher, 2015)), one male and multiple females (e.g. western lowland gorilla, *Gorilla gorilla* (Parnell, 2002)), multiple males and one female (e.g. pheasant-tailed jacana, *Hydrophasianus chirurgus* (Fresneau et al., 2021)), multiple males and multiple females (e.g. African wild ass, *Equus africanus* (Moehlman, 1998)) and sex specific (e.g. impala, *Aepyceros melampus*, where females live in herds, males solitarily (Rduch, 2016)). The social structure describing social interactions is more complex and has itself several components. For example, animals can have pair bonds or not, be teritorial or not, show dominace hierarchies or not and have cooperation or not (hence the number of options is multiplied by 2 in Figure 1). Note that Figure 1 does not list all potential aspects of social structure. The mating system considers who mates with whom (observations) and especially who reproduces with whom, which and can be investigated through genetic studies (Kappeler & van Schaik, 2002). Four catergories of the mating system are recognised: monogamy, polygyny, polyandry or polygynandry, and these can be found in all the social organisations; for example, many pair-living species show extra pair paternity (Dobson et al., 2018). The final component of the social system is the care system that describes who takes care of the dependent young and the categorises are no care, paternal care, maternal care, biparental care, communal care and cooperative care (Kappeler, 2019).

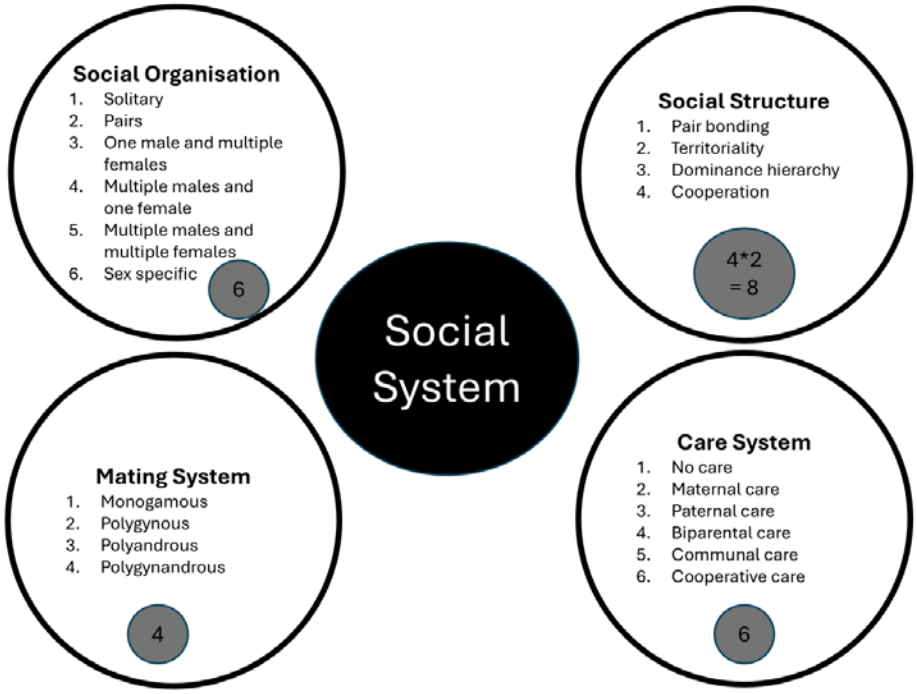

Figure 1. Components of social systems. Social systems consist of four components, indicated by the large white circles. Each component can have mutiple categories, for example there are 6 different categories of social organisation. Indicated in grey circles is for each component how many different categories are possible. For the social structure, the components are multiplied by two because they can each either occur or not (e.g., animals can have pair bonds or not, be territorial or not etc.), which provides the minimum number of possibilities. If the four components are seen categorically, then at least 1152 different forms of social systems are possible. In reality, most components are continous, not categorical (for example not dominance hierachy yes or no, but how hierarchical the dominance hierarchy is), leading to even a higher biodiversity of social systems. Apdapted from Kappeler (2019), published in Makuya & Schradin (2024b).

Regarding the four components of the social system as categories leads to multiple possible combinations. Using such a categorical classification of components of social systems indicates that more than 1000 (6*8*4*6) different forms of social systems are possible (Figure 1). Moreover, the components can vary continuously but not categorically (for example the degree of territoriality; the frequncy of extra-pair young) and are variable (a species can show intra-specific variation in social organisation). This creates the potential for more combinations to be possible (Makuya & Schradin, 2024b). For example, a more realistic measure of mammalian mating systems is using the degree of multiple paternity in litters (Dobson et al., 2018) than placing it into categories (monogamy, polygyny, polyandry or polygynandry). Furthermore, most species are not restricted to one form of social organization but show intra-specific variation in social organization (Schradin et al., 2018b).

In solitary mammals, the social organisation comprises of social units of a single adult individual. Solitary living is determined by the social organisation, where adult males and females live alone, although there may or may not be kinship spatial structure. Solitary species also have social interactions, where neighbours mainly interact amicably or aggressively, and the mating system can involve one male mating with multiple

females with which he overlaps (polygyny) or that both males and females have several partners, with which they either mate randomly (promiscuity) or non-randomly based on mate choice (polygynandry). In mammals, the care system of solitary species is always maternal care only. When the different components of social systems in solitary species are scored categorically, at least 12 different social systems of solitary mammals are possible (Figure 2). However, most components are continuous, for example the degree of aggression between neighbours.

To date, mainly group living species have been studied (Krause & Ruxton, 2002; Rubenstein & Abbot, 2017) with solitary living species receiving less attention. However, solitary mammals are expected to benefit from their form of social organisation while missing out on the benefits of group living. With approximately 22% of studied mammals being solitary living, research needs to be better focussed on understanding the evolution of social behaviour in these species.

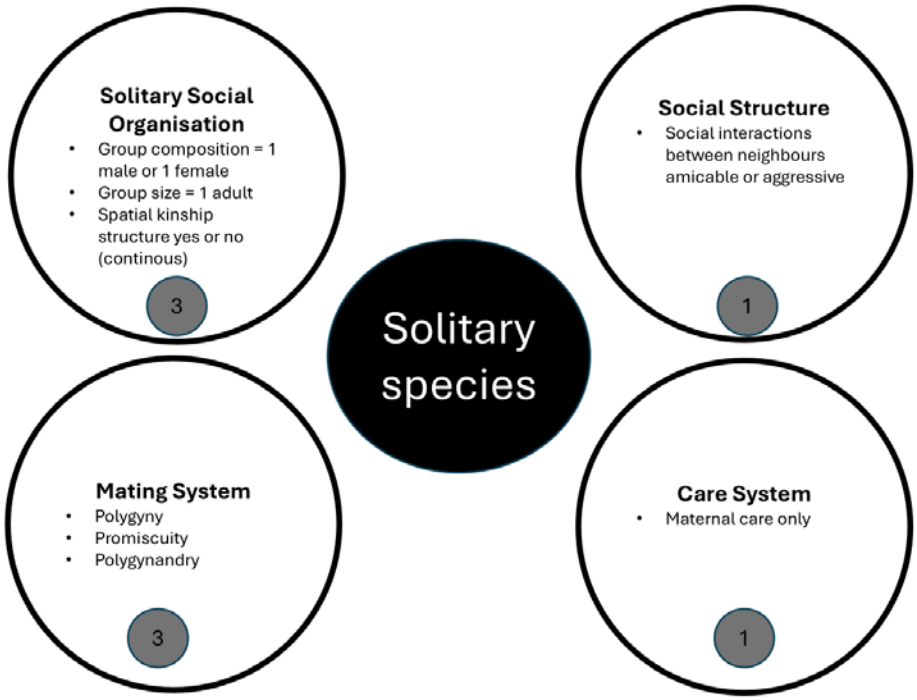

Figure 2. Components of social systems in solitary mammals. The social organisation is made up of three aspects which include the group composition (1 male or 1 female), the group size (1 adult), and the spatial structure (kinship structure or not) which can be continuos. The social structure is only made up of one asepect, the social interactions between neighbours which can be amicable or aggressive. The mating system is made up of three aspects, polygyny, promiscuity or polygynandry. And finally, the care sytem is such that only maternal care is possible (adapted from Kappeler, 2019).

In this thesis, I studied the social and spatial organisation of a solitary rodent, the bush Karoo rat (*Otomys unisulcatus*). The bush Karoo rat was a good model to use because it occupies an open habitat which allowed for easy focal animal observations to be conducted. Previous studies on the bush Karoo rat have provided conflicting conclusions regarding its social organisation, stating that it was group living and in particular that large groups formed (Babu et al., 2011; Do Linh San et al., 2016; Hoepfl, 2013; Xalu et al., 2009) and this required clarification. My thesis generated empirical data to better understand aspects of the social system for this species in the Succulent Karoo. I will first summarise my own results before adding them to the broader concept of solitary living. I will discuss all four components of social systems and how they are related to solitary living species with particular emphasis on the bush Karoo rat results.

Summary of main results

The main aim of my study was to improve our understanding of the costs and benefits of solitary living. My approach was to study the social organisation, spatial structure and social structure of the bush Karoo rat, and further to investigated how these depend on kinship, and whether this could be indicative of some cooperation.

Objective 1. Review costs and benefits of solitary living

In Chapter 2, I reviewed the literature on studies about how common is the occurrence of solitary living in mammals and when it evolved. I then focussed on the theoretical costs and benefits and the empirical evidence exists for these costs and benefits. Solitary living occurs in 22% of all studied mammals. Although solitary living has not been studied as an evolved trait, the fossil record and results from comparative studies show that solitary living is not ancestral in mammals, and indicating that it is probably an adaptation to prevailing environments (Makuya & Schradin, 2024a). However, there was a paucity of data on the costs and benefits of solitary living. The socio-ecological model for group living shows that the key factor that are important to understand the distribution of females in a population is resource distribution as well as the risk of predation and infanticide, which together determine the net benefits of group versus solitary living. In turn, the males distribute themselves according to the distribution of the females (Clutton-Brock, 1989). Solitary living can be an adaptive consequence of the same factors used to explain group living. Therefore, I modified the socio-ecological model developed for group living to that of solitary living (Figure 1 in Chapter 2). My model shows how the costs of group living are the benefits of solitary living, and the benefits of group living are the costs of solitary living (Table 3 in Chapter 2). Chapter 2 provides the theoretical foundation for empirical studies on the costs and benefits of solitary living.

Objective 2. Social organization

Validating the mini-GPS method. To understand solitary living – or any other form of social organisation- one must understand how socio-ecological factors influence the distribution of individuals. I used mini-GPS dataloggers to study the distribution and range size of individuals. This is the first study of its kind on a small rodent. In Chapter 3, I validated the use of mini-GPS dataloggers to measure range sizes compared to the method of using radio tracking in bush Karoo rats. Specifically, I tested whether there was a difference in the range size obtained using the two different methods. Radio tracking resulted in smaller range size estimates that were only representative of the location of the lodges of the rats and not their foraging grounds compared with the larger range size estimates from using mini-GPS dataloggers. Radio tracking has been extensively used to measure range sizes in mammals. However, it has the risk of the radio-collared animal being disturbed by the person tracking it. This is especially problematical for central place foragers. Bush Karoo rats forage away from their main lodges to nearby foraging grounds via runways that interconnect multiple shrubs. When disturbed, they withdraw into their lodges. This was particularly a problem when using the homing in technique during radio tracking, resulting in smaller ranges estimated. Mini-GPS on the other hand provided a more accurate estimation of the range use by not only revealing the lodges the rats occupied, but also their foraging grounds.

Social and spatial organisation. I investigated the social and spatial organization of the bush Karoo rat in Chapter 4. I analysed long-term trapping and observation data of individually marked bush Karoo rats from 2018-2023. I found that bush Karoo rats were mainly solitary living. However, small social groups also occurred which all consisted of close kin, indicating intraspecific variation in social organization. Further, female bush Karoo rats lived closer to kin neighbours and therefore had a spatial structure characterised by kinship. Females also overlapped more with their kin neighbours than with non-kin neighbours. Although this overlap could be interpreted because of living closer to kin, females bush Karoo rats had a similar number of kin and non-kin neighbours. Kinship influences patterns of individual movement (Lacey et al., 2000) and the spatial structures of some solitary felids and rodents has been described as dependent on kinship (Elbroch & Quigley, 2016; Meshriy et al., 2011). In the bush Karoo rat, the spatial kin structure seems to be due to females dispersing for short distances to nearby lodges that sometimes belong to their mother. This is similar to red squirrels (*Tamiascuirus hudsonicus*) which also form kin based spatial structures when offspring disperse only within 100m and inheriting part or all of their mother's territory (Berteaux & Boutin, 2000). In my study, bush Karoo rats were predominantly solitary with very few kin groups and their spatial structure was kin based.

Objective 3. Social structure

Solitary mammals are characterised by males and females mainly meeting for mating and therefore having a few social interactions (Dammhahn & Kappeler, 2009; Schlägel et al., 2019). However, a growing number of studies show that solitary mammals are not asocial but can have non-random and complex social interactions that are sometimes characterised by amicability between interacting individuals (Elbroch & Quigley, 2016; Twining et al., 2024). Although living and interacting with kin has benefits in group living species (Brent et al., 2017), little is known about interactions among kin in solitary species. Therefore, in Chapter 5, I investigated social interactions between neighbouring bush Karoo rats depending on their kinship. I did this through experimental tests both in the laboratory and in the field. I measured aggression and amicability in dyadic encounter tests and field intruder tests between kin and non-kin neighbours. Maternal aggression towards offspring reaching adulthood has been argued as a mechanism driving dispersal in solitary species and in turn leading to solitary living (Eibl-Eibesfeldt, 1953). In contrast, I found that mothers were not aggressive towards their offspring in the dual encounter tests, even after the adult offspring had dispersed. In tests conducted in a neutral arena, aggression was low between adult neighbours, independent of the season and the kinship. However, bush Karoo rats build extensive stick lodges that are a valuable resource because they offer refuge against the harsh environment and protection from predators. I therefore expected the bush Karoo rats to defend their lodges against intruders because it is believed that female territoriality occurs to defend valuable resources (Sinn et al., 2008; Wolff, 2007). I found that bush Karoo rats were more aggressive during the field intruder tests directly in front of the lodges, especially towards non-kin neighbours. These results suggest that the absence of social attraction and the internal motivation of individuals reaching sexual maturity to disperse are the mechanisms driving solitary living in the bush Karoo rats.

Objective 4. Cooperation in a solitary mammal?

In Chapter 6, following up from the result of the range overlap from Chapter 5, I quantified the extent of shared foraging grounds in overlapping ranges. To achieve this, I used the daily ranges estimated from mini-GPS dataloggers to calculate the spatial range overlap between neighbouring bush Karoo rats. Next, I analysed the temporal range overlap using step selection functions to investigate how individual bush Karoo rats moved relative to each other and whether they were attracted to the same or avoided foraging grounds used by other bush Karoo rats with which they overlapped. I found that bush Karoo rats ranges overlapped more with kin than with non-kin neighbours, and they also shared more feeding grounds with close kin. Furthermore, there was mutual tolerance at shared foraging grounds as indicated by attraction towards the same feeding grounds. This can be interpreted as the starting point for the evolution of by-product mutualism, a simple form of cooperation (Bergmüller et al., 2007b). I broadly define cooperation

without taking into consideration how it evolved, following n the definition of Bergmüller et al. (2007b). In by-product mutualism, both interacting individuals benefit at the same time, such that no cheating can take place (Bergmüller et al., 2007b). In some solitary felids, social interactions were classified as social tolerance and not true cooperation within social groups (Jędrzejewski et al., 2022). This included sharing of food resources as observed in pumas (Elbroch & Quigley, 2016). This contrasts with group living felids such as lions (*Panthera leo*) that hunt cooperatively (Stander, 1992). While solitary species were previously believed to be uncooperative with conspecifics and avoid overlapping in their ranges (Sandell, 1989), my study shows that this is not always the case.

What do objectives 1-4 mean when taken together?

The socio-ecological model for solitary living suggested in Figure 1 in Chapter 2 can be applied to the bush Karoo rats using the evidence gathered in my thesis. First, I showed that more than 96% of female and male bush Karoo rats are solitary. Second, bush Karoo rats form kin based spatial structure with small home ranges that they can easily defend from potential intruders. Furthermore, home ranges and associated feeding grounds are partly shared with kin neighbours. This reduces competition. Their home ranges are small and likely determined by the distribution of shrubs and lodges. Bush Karoo rats do not invest much time and energy in aggressively defending the home ranges. The bush Karoo rats showed a simple form of cooperation through mutual tolerance between kin at shared feeding grounds. In sum, the bush Karoo rats display a specific form of solitary social system (Figure 2): Solitary living with a kinship based spatial structure, a non-aggressive social structure with extreme tolerance between solitary females that are close kin, and maternal care only. The mating system is so far unknown, though the fact that there are very few breeding males per breeding females (personal observation) predicts low levels of multiple paternities within litters. This solitary social system contrasts with the old believes that solitary mammals are randomly distributed and highly aggressive towards conspecifics including their adult offspring.

What we still do not know (future studies)

Social organisation and social structure of solitary living species. My study demonstrated that bush Karoo rats formed kin based spatial structures along matrilines. Living near their kin offers the benefit of sharing food resources (Figure 3). However, dispersal strategies in bush Karoo rats still requires additional investigation as has been studied in detail in group living species such as naked mole-rats (Burland et al., 2002). Future studies could focus on aspects such as predation pressure in which the bush Karoo rats are

expected to be solitary living further due to the high predation risk (Figure 3). More future studies could be on investigating social bonding in choice experiments and how these changes as offspring become older. In the few groups observed, future studies could further investigate whether there is any dominance hierarchy and whether bush Karoo rats have any forms of submissive behaviours.

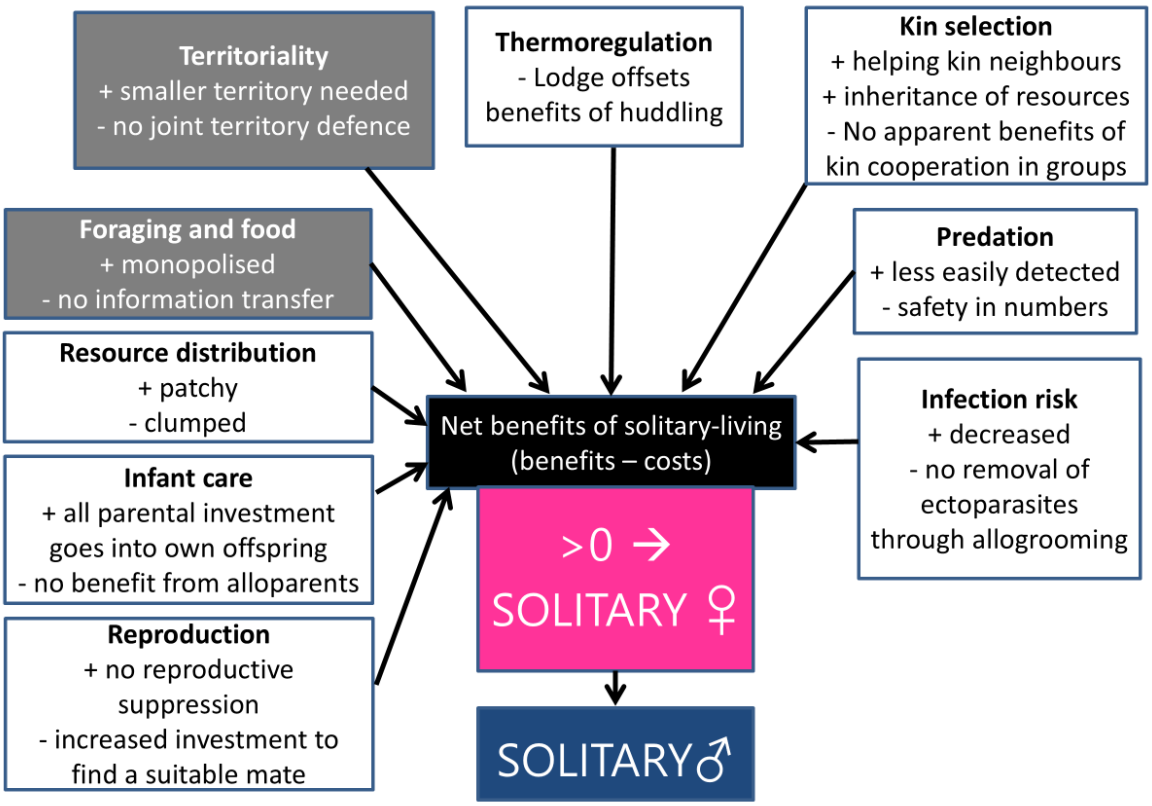

Figure 3. Socio-ecological model to explain solitary living in the bush Karoo rats**.** Nine factors that influence the social organisation of a species are shown. The plus (+) indicates that the described factor favours solitary living while the minus (-) describes costs of solitary living. When these factors are greater than 0, they influence whether solitary living leads to a net benefit for females. In mammals, social organisation is mainly determined by how females distribute themselves, i.e. males follow female social organisation. Factors that have already been studied are indicated in grey and factors that still need investigation in white.

Mating system of solitary living species. The mating system can be inferred from data of focal animal observations as well as genetic analyses (Kappeler, 2019). This information can be used for the classification of animals into different mating system categories, although this has been criticised as being unrealistic (Dobson et al., 2018). The social organisation correlates with the mating system (Kappeler, 2019), with many solitary species having a mating system characterised by multiple paternity (Eberle & Kappeler, 2004). Anecdotal results indicate that bush Karoo rat males roam over large areas during the breeding season, visiting several females. Most males disappear during the breeding season, leading to a

strongly female biased sex ratio. Additional analyses are still needed to understand whether multiple paternity occurs in the bush Karoo rats. Currently, we only understand the female kin structure, but do females with the same mother also same share the same father?

Care system in solitary living species. Paternal care in solitary mammals is usually absent (Hawkins & Racey, 2009). Similarly in the bush Karoo rats, the care system consisted of only mothers providing care for the offspring, similar to the solitary tamarisk gerbil (*Meriones tamariscinus*) (Gromov, 2024). However, some female kin groups of bush Karoo rats occur during the breeding season (Chapter 3). This display of intra-specific variation in social organisation (IVSO) could lead to interesting studies about the evolution of allo-parental care (Box 1).

Box 1. Anecdotal observation of allo-nursing in the normally solitary bush Karoo rat

Allo-nursing is defined as nursing young that are not one's own. It is a special form of allo-parenting that has been observed only in communally breeding animals (Roulin, 2002). It is not expected to occur in mainly solitary living species, but only in highly sociable ones (Solomon & French, 1997), such as communal breeders (several females breed at the same time and do not only nurse their own but also other young; Astúa et al., 2015; Ebensperger et al., 2016; König, 1994; Tardif et al., 1993) and cooperative breeders (subordinate females nurse young of the dominant female; MacLeod et al., 2015; MacLeod et al., 2013). It is assumed to have evolved to increase inclusive fitness and is a "complex", highly derived and special behaviour (MacLeod & Lukas, 2014; Roulin, 2002).

I observed an instance of allo-nursing in the predominantly solitary bush Karoo rat at a lodge that was inhabited by a mother and two adult daughters. The lodge was extremely large (diameter >3m) with many entrances, inside a euphorbia shrub, and it was not clear whether the mother and daughters in fact used the same parts of the lodge or lived in separate areas of the lodge. The mother and one daughter had pups simultaneously, but the mother's pups were several days older and thus much heavier than her daughter's pups. Birth date had been determined by two females losing considerable body mass, 9 days apart from each other. On one occasion, I trapped the mother who was suckling one of her daughter's pups, which at that stage was about 17 days old, while the mother's pups were 26 days old.

Why I think studying allo-nursing in BKRS can be exciting opportunity for further study. Observing allo-nursing in bush Karoo rats, indicates that allo-nursing could evolve when 1. the species is solitary with little aggression (as in the bush Karoo rats) and not solitary and aggressive (such as in the European hamster), and 2. the environment changes such that kin groups form and 3. allo-nursing could increase

inclusive fitness. Being less aggressive might allow for more tolerance between individuals and therefore enable indirect fitness benefits via kin selection.

Solitary living in mammals

Many mammals have intraspecific variation in social organisation (IVSO) which is influenced by population density (Schradin, 2013; Schradin et al., 2018b). Solitary species also display IVSO as seen in the bush Karoo rats in this study. Studying IVSO in solitary species is important to understand the evolutionary reasons for the switch between solitary living to group-living (Schradin et al., 2018b).

My study demonstrated that bush Karoo rats formed kin based spatial structures along matrilines. Associating with kin can in turn help to offset the cost of solitary living through inclusive fitness benefits. Thus, dispersal strategies in solitary species need attention in future studies. The bush Karoo rats have small home ranges which they can easily defend from intruders, with little investment into aggression. This indicates that solitary species in general could benefit from reducing the costs of foraging by having small home ranges.

In many solitary species, the males roam over large areas to gain access to mating opportunities with multiple females and thereby increasing their reproductive success (Eberle & Kappeler, 2004; Randall et al., 2002; Stockley et al., 1994). Thus, solitary mammals often have a mating system characterised by multiple paternity (Eberle & Kappeler, 2004), but more studies are needed, especially to understand variation in multiple paternity between solitary species (Dobson et al., 2018).

In solitary mammals, it seems obvious that the care system consists always of maternal care only. However, as for social organisation, there might be intra-specific variation with some deviation from maternal care only (Box 1). This potential variation needs more exploration as it could offer an avenue of research to understand evolutionary transitions from maternal care only to biparental care systems and care systems with helpers.

Conclusions

The social systems of solitary species are poorly studied, and my study provides foundational information on the occurrence, characteristics and benefits of one solitary species living in a harsh environment. To be solitary living does not mean to be aggressive and asocial. Bush Karoo rats, although being solitary living, showed very low levels of aggression. It had a spatial organisation comprising of close female kin living in close proximity to each other, indicating the important role kin selection could play in solitary

species. My study adds to the growing number of studies that indicate that solitary species have non-random social interactions with their neighbours and that kinship influences social interactions. A simple form of cooperation based on kin selection in combination with by-product mutualism making cheating impossible could be the beginning of more complex forms of cooperation where tolerance is replaced by amicable interactions. My thesis demonstrates that studying solitary species can help us to reach a better understanding of social evolution. It furthers shows how important it is to realise that there are different forms of solitary living (Figure 2).

References

Astúa, D., Carvalho, R. A., Maia, P. F., Magalhães, A. R., & Loretto, D. (2015). *First evidence of gregarious denning in opossums (Didelphimorphia, Didelphidae), with notes on their social behaviour* (Vol. 11).

Babu, N., Xalu, M., & San, E. D. L. (2011). Socio-spatial organization of bush Karoo rats (Otomys unisulcatus) in the Albany Thicket Biome: preliminary results.

Bergmüller, R., Russell, A. F., Johnstone, R. A., & Bshary, R. (2007b). On the further integration of cooperative breeding and cooperation theory. *Behavioural Processes, 76*, 170- 181.

Berteaux, D., & Boutin, S. (2000). Breeding dispersal in female North American red squirrels. *Ecology, 81*(5), 1311-1326. doi: https://doi.org/10.1890/0012-9658(2000)081[1311:BDIFNA]2.0.CO;2

Bray, T. C., Bloomer, P., O'Riain, M. J., & Bennett, N. C. (2012). How attractive is the girl next door? An assessment of spatial mate acquisition and paternity in the solitary Cape dune mole-rat, Bathyergus suillus. *PloS one, 7*(6), e39866.

Brent, L. J. N., Ruiz-Lambides, A., & Platt, M. L. (2017). Family network size and survival across the lifespan of female macaques. *Proceedings of the Royal Society B: Biological Sciences, 284*(1854), 20170515. doi: doi:10.1098/rspb.2017.0515

Burland, T. M., Bennett, N. C., Jarvis, J. U. M., & Faulkes, C. G. (2002). Eusociality in African mole-rats: new insights from patterns of genetic relatedness in the Damaraland mole-rat (*Cryptomys damarensis*). *Proc R Soc Lond B, 269*, 1025-1030.

Clutton-Brock, T. H. (1989). Mammalian mating systems. *Proceedings of the Royal Society B: Biological Sciences, 236*, 339-372.

Dammhahn, M., & Kappeler, P. M. (2009). Females go where the food is: does the socio-ecological model explain variation in social organisation of solitary foragers? *Behavioral Ecology and Sociobiology, 63*, 939–952. doi: 10.1007/s00265-009-0737-2

Do Linh San, E., Babu, N., Xalu, M., Le Gars, S., Perquin, J., & Baxter, R. (2016). A conservation assessment of Otomys unisulcatus. *The red list of mammals of South Africa, Swaziland and Lesotho*.

Dobson, F. S., Abebe, A., Correia, H. E., Kasumo, C., & Zinner, B. (2018). Multiple paternity and number of offspring in mammals. *Proceedings of the Royal Society B: Biological Sciences, 285*(1891). doi: 10.1098/rspb.2018.2042

Ebensperger, L. A., Correa, L. A., León, C., Ramírez-Estrada, J., Abades, S., Villegas, Á., & Hayes, L. D. (2016). The modulating role of group stability on fitness effects of group size is different in females and males of a communally rearing rodent. *Journal of Animal Ecology, 85*(6), 1502-1515. doi: 10.1111/1365-2656.12566

Eberle, M., & Kappeler, P. M. (2004). Sex in the dark: determinants and consequences of mixed male mating tactics in Microcebus murinus, a small solitary nocturnal primate. *Behavioral Ecology and Sociobiology, 57*, 77-90.

Eibl-Eibesfeldt, I. (1953). Zur Ethologie des Hamsters (*Cricetus cricetus* L.). *Zeitschrift für Tierpsychologie, 10*(2), 204-254. doi: https://doi.org/10.1111/j.1439-0310.1953.tb00392.x

Elbroch, L. M., & Quigley, H. (2016). Social interactions in a solitary carnivore. *Current Zoology, 63*, zow080. doi: 10.1093/cz/zow080

Fresneau, N., Lee, Y.-F., Lee, W.-C., Kosztolányi, A., Székely, T., & Liker, A. (2021). Sex role reversal and high frequency of social polyandry in the pheasant-tailed jacana (Hydrophasianus chirurgus). *Frontiers in Ecology and Evolution, 9*, 742588.

Gromov, V. S. (2024). Ecology and social behavior of the tamarisk gerbil Meriones tamariscinus: Insights from long-term research in the wild and seminatural environments. *Current Zoology*, zoae004.

Hawkins, C. E., & Racey, P. A. (2009). A novel mating system in a solitary carnivore: the fossa. *Journal of Zoology, 277*(3), 196-204. doi: https://doi.org/10.1111/j.1469-7998.2008.00517.x

Hoepfl, J. (2013). Aspects of the biology and behaviour of the Bush Karoo Rat (Otomys unisulcatus) in the Central Karoo, South Africa.

Jędrzejewski, W., Hoogesteijn, R., Devlin, A. L., Tortato, F., Concone, H. V. B., Azevedo, F., Eriksson, C. E., Fragoso, C. E., Abarca, M., Morato, R. G., Giordano, A. J., Hoogesteijn, A., Rampim, L., Gamarra, G., Carreño, R., Velásquez, G., & Schmidt, K. (2022). Collaborative behaviour and coalitions in male jaguars (Panthera onca)—evidence and comparison with other felids. *Behavioral Ecology and Sociobiology, 76*(9), 121. doi: 10.1007/s00265-022-03232-3

Kappeler, P. M. (2019). A framework for studying social complexity. [journal article]. *Behavioral Ecology and Sociobiology, 73*(1), 13. doi: 10.1007/s00265-018-2601-8

Kappeler, P. M., & van Schaik, C. P. (2002). Evolution of primate social systems. *International journal of primatology*, *23*(4), 707-740.

König, B. (1994). Components of lifetime reproductive success in communally and solitarily nursing house mice - a laboratory study. *Behav Ecol Sociobiol*, *34*, 275-283.

Krause, J., & Ruxton, G. D. (2002). *Living in Groups*. Oxford: Oxford University Press.

Lacey, E. A., Patton, J., & Cameron, G. (2000). Spatial and social systems of subterranean rodents. *Life underground: the biology of subterranean rodents (EA Lacey, JL Patton, and GN Cameron, eds.). University of Chicago Press, Chicago, Illinois*, 257-296.

MacLeod, K., & Lukas, D. (2014). Revisiting non-offspring nursing: allonursing evolves when the costs are low. *Biology letters*, *10*(6), 20140378.

MacLeod, K. J., McGhee, K. E., & Clutton-Brock, T. H. (2015). No apparent benefits of allonursing for recipient offspring and mothers in the cooperatively breeding meerkat. *Journal of Animal Ecology*, *84*(4), 1050-1058. doi: 10.1111/1365-2656.12343

MacLeod, K. J., Nielsen, J. F., & Clutton-Brock, T. H. (2013). Factors predicting the frequency, likelihood and duration of allonursing in the cooperatively breeding meerkat. *Animal Behaviour*, *86*. doi: 10.1016/j.anbehav.2013.09.012

Makuya, L., & Schradin, C. (2024a). Costs and benefits of solitary living in mammals. *Journal of Zoology*, *323*, 9-18. doi: https://doi.org/10.1111/jzo.13145

Makuya, L., & Schradin, C. (2024b). The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences*, *121*(13). doi: https://doi.org/10.1073/pnas.2402871121

Meshriy, M. G., Randall, J. A., & Parra, L. (2011). Kinship associations of a solitary rodent, Dipodomys ingens, at fluctuating population densities. *Animal Behaviour*, *82*(4), 643-650.

Moehlman, P. D. (1998). Feral asses (Equus africanus): intraspecific variation in social organization in arid and mesic habitats. *Applied Animal Behaviour Science*, *60*(2-3), 171-195.

Parnell, R. J. (2002). Group size and structure in western lowland gorillas (Gorilla gorilla gorilla) at Mbeli Bai, Republic of Congo. *American Journal of Primatology: Official Journal of the American Society of Primatologists*, *56*(4), 193-206.

Randall, J. A., Hekkala, E. R., Cooper, L. D., & Barfield, J. (2002). Familiarity and flexible mating strategies of a solitary rodent, *Dipodomys ingens*. *Anim Behav*, *64*, 11-21.

Rathbun, G. B., & Dumbacher, J. P. (2015). Home range and use of diurnal shelters by the Etendeka round-eared sengi, a newly discovered Namibian endemic desert mammal. *PeerJ*, *3*, e1302.

Rduch, V. (2016). Population characteristics and coexistence of puku (Kobus vardonii) and impala (Aepyceros melampus) in and around Kafue National Park, Zambia. *Mammalian Biology*, *81*(4), 350-360.

Roulin, A. (2002). Why do lactating females nurse alien offspring? A review of hypotheses and empirical evidence. *Animal Behaviour, 63*, 201-208.

Rubenstein, D. R., & Abbot, P. (2017). The Evolution of Social Evolution. In D. R. Rubenstein & P. Abbot (Eds.), *Comparative Social Evolution* (pp. 1-18). Cambridge: Cambridge University Press.

Sandell, M. (1989). The mating tactics and spacing patterns of solitary carnivores *Carnivore behavior, ecology, and evolution* (pp. 164-182): Springer.

Schlägel, U. E., Signer, J., Herde, A., Eden, S., Jeltsch, F., Eccard, J. A., & Dammhahn, M. (2019). Estimating interactions between individuals from concurrent animal movements. *Methods in Ecology and Evolution, 10*(8), 1234-1245.

Schradin, C. (2013). Intraspecific variation in social organization by genetic variation, developmental plasticity, social flexibility or entirely extrinsic factors. [Review]. *Philosophical Transactions of the Royal Society B-Biological Sciences, 368*(1618). doi: 10.1098/rstb.2012.0346

Schradin, C., Hayes, L. D., Pillay, N., & Bertelsmeier, C. (2018b). The evolution of intraspecific variation in social organization. *Ethology, 124*(8), 527-536.

Sinn, D. L., While, G. M., & Wapstra, E. (2008). Maternal care in a social lizard: links between female aggression and offspring fitness. *Animal Behaviour, 76*(4), 1249-1257. doi: https://doi.org/10.1016/j.anbehav.2008.06.009

Solomon, N. G., & French, J. A. (1997). *Cooperative Breeding in Mammals*. Cambridge: Cambridge University Press.

Stander, P. E. (1992). Cooperative hunting in lions: the role of the individual. *Behavioral ecology and sociobiology, 29*, 445-454.

Stockley, P., Searle, J. B., Macdonald, D. W., & Jones, C. S. (1994). Alternative reproductive tactics in male common shrews: relationships between mate-searching behavior, sperm production, and reproductive success as revealed by DNA fingerprinting. [Article]. *Behavioral Ecology and Sociobiology, 34*(1), 71-78.

Tardif, S. D., Harrison, M. L., & Simek, M. A. (1993). Communal infant care in marmosets and tamarins: relation to energetics, ecology, and social organization. In A. B. Rylands (Ed.), *Marmosets and Tamarins: Systematics, Behaviour and Ecology* (pp. 220-234). London: Oxford University Press.

Twining, J. P., Sutherland, C., Zalewski, A., Cove, M. V., Birks, J., Wearn, O. R., Haysom, J., Wereszczuk, A., Manzo, E., & Bartolommei, P. (2024). Using global remote camera data of a solitary species complex to evaluate the drivers of group formation. *Proceedings of the National Academy of Sciences, 121*(12), e2312252121.

Wolff, J. O. (2007). Social biology of rodents. *Integrative Zoology, 2*(4), 193-204. doi: 10.1111/j.1749-4877.2007.00062.x

Xalu, M., Babu, N., & San, E. D. L. (2009). Activity patterns of bush Karoo rats (Otomys unisulcatus) in the Thicket Biome of South Africa: preliminary results.

APPENDIX

List of publications I co-authored during my PhD. PDFs of publications related directly to my PhD topic of solitary living are attached (marked as *).

Peer-reviewed

***Makuya, L**., & Schradin, C. 2024. The secret social life of solitary mammals. *Proceedings of the National Academy of Sciences,* 121(13). doi: https://doi.org/10.1073/pnas.2402871121

Mulvey, J., Pillay, N., **Makuya, L**., Rödel, H.G. & Schradin, C., 2023. African striped mice have relatively smaller brains in the food deprived dry season than in the wet season. Mammalian Biology, pp.1-10. https://doi.org/10.1007/s42991-023-00383-2

Makuya, L, Pillay, N, Rimbach, R. & Schradin, C. 2023. Suppression of the physiological stress response is not stress. Trends in Ecology & Evolution. https://doi.org/10.1016/j.tree.2023.06.007

Schradin, C, **Makuya, L**, Pillay, N. & Rimbach, R. 2023. Harshness is not stress. Trends in Ecology & Evolution. http://doi.org/10.1016/j.tree.2022.12.005.

Makuya, L, Olivier, C.A. & Schradin, C. 2022. Field studies need to report essential information on social organization - independent of the study focus. Ethology. 128: 268-274.

*Olivier, C.A., Schradin, C. & **Makuya, L**. 2022. Global Change and Conservation of Solitary Mammals. Frontiers in Ecology and Evolution 10:906446. doi: 10.3389/fevo.2022.906446.

Popular science

Makuya, L. 2024. Some solitary mammals have surprisingly social lives: what I learned from a tiny South African rodent. *The Conversation* https://theconversation.com/some-solitary-mammals-have-surprisingly-social-lives-what-ive-learned-from-a-tiny-south-african-rodent-231510.

Makuya, L. & Schradin, C. 2024. Living solitary is neither easy nor primitive. Journal of Zoology Blog. https://jzoblog.wordpress.com/2024/02/16/living-solitary-is-neither-easy-nor-primitive/.