

University of South Bohemia in České Budějovice
Faculty of Science

**Testing variation of locomotor activity and work within
families of a social African mole-rat (*Fukomys darlingi*), using the
Trovan technology electronic identification system**

Master's thesis

Bc. Terezie Englová

Supervisor: prof. doc. Mgr. Radim Šumbera, Ph.D.

Consultant: Mgr. Jan Šklíba, Ph. D.

České Budějovice 2019

Englová T. 2019. Testing variation of locomotor activity and work within families of a social African mole-rat (*Fukomys darlingi*), using the Trovan technology electronic identification system. Mgr. Thesis, in English- 58 p., Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic.

Annotation:

Activity of social mole-rats' and their contribution to work and cooperative tasks are explained by two different types of polyethism, caste and age polyethism. Behavioural castes, based on different contributions to cooperative tasks were defined in many mole-rats' species. However, additional experiments which would simulate higher working activity through manipulation with substrate or removal of some family members are rare. Contrary, in field and different laboratory studies, differences between individuals in the amount of work performed and task allocation were rather continual, and based on age (body mass). This study reveals locomotor and working activity of five families of *Fukomys darlingi* (43 individuals) which were obtained by radio-identification monitoring system and personal observation. Each family was tested twice, for the first time established families were tested and then after removal of most active workers. The activity is related to individuals' body mass, sex and reproductive status.

I hereby declare that I have worked on my diploma thesis independently and used only the sources listed in the bibliography. I hereby declare that, in accordance with Article 47b of Act No. 111/1998 in the valid wording, I agree with the publication of my master thesis, in full form resulting from deletion of indicated parts to be kept in the Faculty of Science archive, in electronic form in a publicly accessible part of the IS STAG database operated by the University of South Bohemia in České Budějovice accessible through its web pages. Further, I agree to the electronic publication of the comments of my supervisor and thesis opponents and the record of the proceedings and results of the thesis defence in accordance with aforementioned Act No. 111/1998. I also agree to the comparison of the text of my thesis with the Theses.cz thesis database operated by the National Registry of University Theses and a plagiarism detection system.

In České Budějovice, 11. 12. 2019

.....
Bc. Terezie Englová

Acknowledgements

I would like to thank my supervisor, Radim Šumbera, for all his advice he gave me since I started to study mole- rats, Jan Okrouhlík for helping me with developing methodology of experiments and for help with consequent statistical analyses, also Matěj Lövy for his involvement and finally, Jan Šklíba for his help and consultations of results and structure of the text. I would like to thank our whole group, including Radka Pešková, who also gave me valuable advice and help. Finally, I would like to thank my family, especially my brother who developed the programme for counting walked distances of individuals and time spent in the tunnel with barrier and all my friends and my boyfriend for their endless support.

Content

1	INTRODUCTION	1
1.1	Social behaviour.....	1
1.2	Cooperatively breeding societies	1
1.2.1	Helpers.....	3
1.3	Eusociality.....	4
1.3.1	Eusociality in vertebrates	4
1.3.1.1	Should mole-rats be considered eusocial?	5
1.3.2	Morphologically distinct castes in mole-rats.....	6
1.3.2.1	Reproductive caste	6
1.3.2.2	Dispersal caste	8
1.4	Age polyethism	9
1.4.1	The definition of age polyethism in invertebrates an mole-rats	9
1.4.2	Division of labour and specialization in tasks.....	10
1.4.2.1	Division of labour in mole-rats	11
1.4.2.2	Task switching	12
1.4.2.3	Task specialization.....	13
1.5	Mole-rats: Age or caste polyethism?	14
1.5.1	Failures to define behavioural castes which differ in body masses.....	14
1.5.2	Continual changes in activity	15
1.5.3	Contribution to cooperative tasks in males and females	16
1.5.4	Studies from field	17
1.6	Limits of laboratory experiments on mole-rats.....	19
1.7	Aims of my study.....	22
2	MATERIALS AND METHODS.....	24
2.1	Studied species.....	24
2.2	Experimental room, experimental burrow and the recording system	24
2.3	Sizes and characteristics of tested families	25
2.4	Tested animals and repetition	26
2.5	Experimental design.....	27
2.5.1	Experiment with digging substrate.....	27
2.5.2	Experiment with the barrier.....	28
2.6	Collected data and terminology	29
2.7	Statistical analysis.....	29
2.7.1	Experiment I. - Before removal.....	29
2.7.2	Experiment II. – After removal	30
3	RESULTS.....	31
4	DISCUSSION.....	44
5	CONCLUSION.....	48
6	BIBLIOGRAPHY.....	50

1 INTRODUCTION

1.1 Social behaviour

Social behaviour is defined by interactions between two or more individuals. It gave rise to many types of societies, ranges from simple societies, like temporal feeding aggregations to complex societies of cooperative breeders with division of labour and high levels of cooperation and with reproduction restricted to few individuals. In complex societies some individuals do not reproduce and perform tasks through which they help other individuals to breed. They behave altruistically. According to Darwin's theory of natural selection (Darwin 1859) individuals which increase direct fitness should be favoured over those which do not. However, individuals of highly social groups sometimes increase fitness of other members of a group to the detriment of their own fitness. (Davies et al. 2012).

When group members are genetically related, altruism can be explained by Hamilton's rule (Hamilton 1964). If an altruistic behaviour is directed towards a related individual, it can increase so called inclusive fitness. The cost of the donor must be overbalanced by the benefit of the recipient multiplied by the relatedness of the two. According to this rule it can be beneficial to help a closely relative individual.

1.2 Cooperatively breeding societies

High genetic relatedness of individuals in a group promotes evolution of complex societies in mammals, birds and insects (Lukas & Clutton-Brock, 2012), so-called cooperative breeding, where reproduction is restricted to few individuals and nonbreeders show alloparental care (Clutton-Brock 2009).

Cooperative breeding is highly associated with monogamy. It has been shown that the transition to cooperative breeding emerge from monogamy lineages and not from communal breeding societies characterized by polygynous mating systems.

So genetic relatedness has a strong influence on the evolution of cooperative breeding strategies(Lukas & Clutton-Brock, 2012).

According to phylogenetic reconstruction there are 14 evolutionary transitions to cooperative breeding: In rodents in *Fukomys*, *Heterocephalus*, *Microtus*, *Meriones*, *Rhabdomys*, *Castor*, *Atherurus* and two in *Peromyscus*; in carnivores in *Alopex*, *Canis*, *Lycaon*, in primates in Callitrichidae and in mongooses in Mungotinae. A model which assume that the evolution of cooperative breeding and social monogamy are tied is more likely than the model predicting the individual origin (Lukas & Clutton-Brock, 2012).

In **Cooperative breeding** societies subordinates care for offspring and sometimes perform other tasks related to colony maintenance. Individuals who stay in a group and help with taking care of pups are called helpers (Krebs and Davies 1997). Helpers contribute to different tasks which need to be done in order to maintain the colony and raise offspring successfully. Contributions to cooperative activities are much higher in helpers than breeding individuals.

Cooperative breeders can be divided into two categories, facultative and specialized cooperative breeders (Clutton Brock 2006). In **facultatively cooperative breeders**, parents and helpers from the previous litters share care juveniles, usually in unsuitable conditions. Representatives of this group are for example: golden and silver-backed jackals (Moehlman 1987), marmosets and tamarins (Goldizen 1988), and several marmots (Blumstein et al. 1999).

In facultatively cooperative breeders the possibility of rearing pups by a reproductive pair without helpers is higher than in specialized cooperatively breeding groups. **Specialized cooperative breeders** include African wild dogs (Creel & Creel 2002), Kalahari meerkats (Clutton-Brock et al. 1998) and many mole-rats' species: naked mole-rats (Lacey & Sherman 1991), Damaraland mole- rat (Bennett & Jarvis 1988) and Ansell's mole- rats (Patzenhaerová et al. 2013).

1.2.1 Helpers

Helpers can significantly lower costs of breeding to breeding female. Costs of breeding are lower with higher number of helpers in meerkats. Also interbirth intervals are shorter, resulting in increasing fitness of breeding female (Clutton-Brock et al. 1998). If helpers provide breeders with food they reduce fitness costs of breeding to dominant females even more by eliminating survival costs during foraging.

Positive relationships between the number of helpers and breeding success was found in facultatively cooperatively breeding carnivores, including golden and silver-backed jackals (Moehlman 1987), marmosets and tamarins (Goldizen 1988), as well as in most of the specialized cooperative breeders, including dwarf mongooses (Rood 1990), meerkats (Clutton-Brock et al. 1999) and African wild dogs (Creel & Creel 2002).

In cooperatively breeding societies staying in a group may seem to bring more costs for helpers with abandoning reproduction as the highest one. Short term costs of helping in meerkats are weight loss (Clutton-Brock et al. 1998) or a greater risk of injury or predation (Clutton-Brock et al. 2000). On the other hand, individuals can increase their indirect fitness by staying in a group, when risks of dispersal are too high (Danchin et al. 2008).

Individuals can also increase their chance for gaining a breeding status and increase their direct fitness by staying within a group (Lucas & Clutton-Brock 2012), which results in competition among nonbreeders and different helping effort among nonbreeders. Investing energy in helping may decrease helpers' future reproductive success. Therefore helpers should invest less energy in helping when the possibility of gaining breeding status is higher and they work less when the group is larger (Cant & Field, 2001, Russell et al. 2003). Societies of cooperative breeders are very complex; however eusocial insects are the most complicate social organizations, where individuals have many fold lower chance to reproduce and members of castes are even morphologically different from each other and from the queen.

1.3 Eusociality

The term eusociality was firstly used by Batra (1996) in bees, where “nest founding parents cooperate with mature daughters and division of labour occurs”. Michener (1969) defined eusociality in bees with three characteristics: morphological castes with division of labour, generation overlap of offspring and cooperative work on cells. Later Wilson (1971) generalized the definition for possible application to all invertebrates. Eusociality was defined as a social arrangement with the division of reproductive roles, cooperative care of offspring and overlapping generations of adults.

Eusociality has been described in several invertebrates, for instance in: aphids (Aoki 1982), Australian weevils (Kent & Simpson 1992), Australian thrips from Thysanoptera order (Crespi 1992) and shrimps (Duffy et al. 1996).

1.3.1 Eusociality in vertebrates

Jarvis (1981) compared vertebrates (the naked mole-rat *Heterocephalus glaber*) with eusocial invertebrate societies, and she described eusociality in this species. As the main feature of eusociality, Jarvis stated various castes of individuals. These castes were described to differ on the basis of the amount of work performed, such as digging or nesting material transporting. New terms which were lately often used to describe castes in other mole-rat species were proposed; **frequent** and **infrequent workers**. Members of these castes differed in body mass and also age. The caste of frequent workers consists of young individuals with lower body masses and infrequent workers contain older and heavier individuals. Breeding male and other individuals who did not contribute to maintaining tasks were described as the caste of **nonworkers** (Jarvis 1981). Later, this term was used only for breeding pair (Bennett & Jarvis 1988, Bennett et al. 1990, Bennett 1990).

Therefore, this study started a great interest in African mole-rats and more attempts to find eusociality among other mole-rats species were carried out. The attention was firstly focused on Damaraland mole rat because of their high numbers of colony members.

Recorded types of behaviour in studies of behavioural castes were food carrying, transporting of nesting material, burrow maintaining, digging and transporting of soil (Bennett & Jarvis 1988, Bennett 1990, Jacobs et al. 1991). Behavioural castes were also defined in *Fukomys damarensis* and eusociality was also described in this species (Bennett & Jarvis 1988, Bennett 1990, Jacobs et al. 1991). As in *Heterocephalus*, body masses of infrequent workers were higher in these studies (Bennett & Jarvis 1988, Bennett 1990, Jacobs et al. 1991). Since then Damaraland mole-rats and naked mole-rats, have been described as eusocial.

1.3.1.1 Should mole-rats be considered eusocial?

Description of eusociality among vertebrates was not fully supported by some authors. As mole-rats were considered eusocial, other cooperatively breeding species of birds and mammals could be defined as eusocial as well. For example meerkats *Suricata suricatta* (Clutton-Brock et al. 2000), common dwarf mongooses *Helogale parvula* (Rood 1990), African wild dogs *Lycaon pictus* (Creel & Creel 2002), marmosets and tamarins (Goldizen 1988), and Florida scrub Jay *Aphelocoma coerulescens* (Stallcup & Woolfenden 1978).

According some authors, societies should not be separated into distinct categories, rather they should be considered as a part of a continuum, the **eusocial continuum**. Position of each species could be identified by a degree of reproductive skew (breeding asymmetry) (Sherman et al. 1995). Values of this skew range from zero (for groups, where all individuals have a chance to breed) to one (for groups, where reproduction is completely monopolized by one breeding female and one breeding male per group/colony). The advantage of this approach is that all vertebrates and invertebrates can be arranged on one scale. Eusociality was later described in Ansell's mole-rat *Fukomys anelli* on the basis of reproductive skew proxy parameters (Patzenhauerová et al. 2013).

Some authors disagreed with the continuum and saw certain limits in this approach; therefore attempts of redefining eusociality emerged. Crespi & Yanega (1994) proposed to add a criterion of permanent castes as were describes for example in bees (Mitchener 1969), into the definition of eusociality.

Besides morphological castes, another additional criterion for eusociality was proposed by Crespi & Yanega (1994): the existence of groups of individuals who became “behaviourally distinct at some point prior to maturity”, thus forming thus certain irreversible behavioural castes. According to this approach, eusocial colonies of insects should be strictly divided from cooperatively breeding societies and social arrangement of mole rats *Heterocephalus glaber*, *Fukomys damarensis* and *Fukomys anselli* should be considered as semi social, not eusocial (Crespi & Yanega 1994). Another attempt to redefine eusociality was not so strict. It was proposed to add the criterion of permanent castes, but only to distinguish between highly eusocial species from primitively eusocial species, based on the presence or absence of morphological castes (Gadagkar 1994). Thus mole- rats can be considered as primitively eusocial.

1.3.2 Morphologically distinct castes in mole-rats

1.3.2.1 Reproductive caste

Attempts of redefining eusociality by means of adding the criterion of permanent castes (Crespi & Yanega 1994, Gadagkar 1994) started searching for irreversible castes among mole-rats.

One of these approaches was to compare mole- rats breeding female with a queen in eusocial insect societies. In many insect societies breeding queens are morphologically distinct (Holldobler & Wilson 1990). So called **reproductive caste** was described in eusocial mole-rats, with „irreversible changes linked with specialization” (Fig. 1) (Henry et al. 2007).

When Damaraland and naked mole-rat females acquire breeding position and start to breed, their lumbar vertebrae prolong. This phenomenon was used to prove the existence of castes of queens (O’Riain et al. 2000, Henry et al. 2007, Young & Bennett 2010).

Also the skull grow rates slowed down after gaining breeding position in naked mole-rat breeding female, which could be explained by the high energy investment in growing, which is not necessary when dominant females do not contribute to cooperative tasks like digging and burrow maintaining (Young & Bennett 2010).

Breeding females of Damaraland mole-rats are also compared to queens, due to longer life spans of breeding females. In eusocial insects queens live longer than nonbreeders of a group. Recapture data showed that breeding females persisted in colonies much longer than nonbreeding individuals (Schmidt et al. 2016). However, in natural conditions higher levels of mortality associated with foraging or disappearance because of dispersal must be taken into consideration. Differences in longevity were found between breeders and nonbreeders in *Fukomys anselli* also in laboratory conditions (Damman et al. 2011). It was found that breeders have evolved defences against combined oxidant and carbonyl stress compared to nonbreeders (Damman et al. 2011).

The justification of using the term morphological caste in mole rats is highly dubious. Slight changes of skeleton are common among vertebrates, as the mechanism of hormonal changes. For example in rats Bowman & Miller (1999) found expedited bone grow and mineralization after lactation. Vajda et al. (2001) confirmed that after lactation, losses in bone mineral density caused by pregnancy are compensated and bone formation rates increase. Consequently both, cortical and cancellous bone can be rebuilt even more effectively (Fig. 2). Changes in the bone structure can therefore be considered as common phenomenon among mammals (cf Burda 1999).

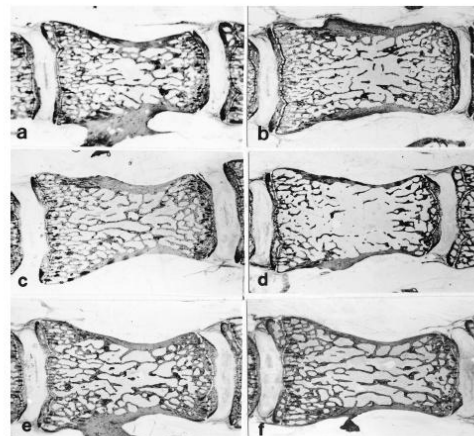


FIG. 2. Photomicrographs of lumbar vertebra from rats at parturition (b), weaning (d), and 8 wk after weaning (f) and their respective nulliparous controls (a, c, and e). Bone loss is apparent at weaning (d), but is restored 8 wk later (f). Von Kossa-stained sections; original magnification $\times 11$.

Fig. 1: X rays of a queen (C,D) Fig. 2: Lumbar vertebrae of a breeding rat (b,d,f)

1.3.2.2 Dispersal caste

Castes in mole-rats were also described on the basis of changes linked with dispersal. Some morphological, behavioural and physiological changes were found also among individuals who were about to disperse. Namely larger body fat reserves (O'Riain et al. 1996, Braude 2000, Hazell et al. 2000), higher levels of luteinizing hormone in blood and less working activity performed (O'Riain et al. 1996, Bennett & Faulkes 2000). However, larger fat reserves might be linked with the preparation for dispersal. In Belding's ground squirrels, *Spermophilus beldingi*, timing of dispersal is dependent on body masses and fat reserves in males (Nunes & Holekamp 1995).

In addition behavioural changes, like preferable mating with individuals from other colonies and greater levels of aggression towards conspecifics were supposed to be another distinct factor between naked mole-rat dispersers compared to other colony members (O'Riain et al. 1996).

In the study of Scantlebury et al. (2006), activity of predefined castes measured by DLW approach differ between periods of rainy and dry seasons. During dry periods, small individuals (frequent workers), were more active. Whereas during the period of rain, individuals with greater body masses (infrequent workers), increased their activity. It was concluded that heavier individuals are members of dispersing caste, which do not contribute to cooperative tasks but focus on building their own fat reserves and activate only under suitable conditions.

1.4 Age polyethism

Different levels of contribution to cooperative tasks and performance of various tasks among members of mole-rat families can be considered as a consequence of age polyethism and not the existence of castes. The variability in activity among individuals might be rather a continuum artificially categorized into castes.

1.4.1 The definition of age polyethism in invertebrates and mole-rats

Age polyethism was firstly proposed by Wilson (1963). He proposed to distinguish between caste and age polyethism. As a key example he provided age polyethism of honey bees, where younger bees stay in a hive as nurses and switch to foraging as they age (Wilson 1963). Later, age polyethism was defined as a phenomenon in societies where “the same individual passes through different forms of specialization as it grows older” (Wilson 1971). Specialization and switching between tasks as individuals age is the main sign of age polyethism. Similarly to caste polyethism, the assumptions for age polyethism is the presence of more litters in a group thus the possibility to substitute older individuals in performing a task and the possibility of older individuals to switch to another task.

Age polyethism had been already speculated to exist in mole-rats in the study of Jarvis (1981). However, it was supposed to be limited only to members of a group with higher growth rates and therefore the ability to join caste of nonworkers and possibly gain breeding status later. The caste of nonworkers were used only for breeding pair in consequent studies and age polyethism was not considered as an explanation for differences in individual activity and contribution to different cooperative tasks in mole-rats.

Lacey & Sherman (1991) proposed that age polyethism in mole-rats is not restricted to a subset of colony members, as Jarvis (1981) stated, but represents a flexible series of behavioural changes which occur as an individual age and grow.

Lacey & Sherman (1991) did not require all individuals to shift from colony maintenance to colony defence at the predetermined age. They suggested that the timing of transition between tasks is affected by ecological or demographic conditions.

1.4.2 Division of labour and specialization in tasks

In large colonies of socially living animals it is important to perform tasks linked with colony maintenance with the highest possible efficiency. Specialization in tasks can eliminate performing the work inefficiently and can be therefore considered as a product of the selection for increased effectivity of a colony (O’Riain & Jarvis 1998, Tofts & Franks 1992, Mooney et al 2015).

Hypothesis “Foraging for work” presumes that individuals start to perform a task which is the closest to them after birth. Consequently, these individuals switch to another task. The change of a specific task does not necessarily be depended on age andn the division of labour is therefore caused by the actual needs of a colony (Tofts & Franks 1992)

In invertebrates the task specialization and division of labour are common which is in contrast with only a few examples reported in vertebrates. Some of the most known specialization are coordinated hunts where individuals repeatedly adopt specific roles, as has been described in African lions, *Panthera leo* (Stander 1992), bottlenose dolphins, *Tursiops truncatus* (Cox & Bar 2005), and chimpanzees, *Pan troglodytes* (Boesch 2002).

Coordinated hunting is described in relatively stable groups where individuals recognize one another and interact repeatedly. This allows individuals to practise and perfect the specific motor controls for their role and it could be defined as a ‘team work’ (Anderson & Franks, 2001).

1.4.2.1 Division of labour in mole-rats

In mole-rats nonbreeding individuals can contribute to care of young directly by bringing food to them, carrying them, performing grooming, etc. They can also contribute indirectly by maintaining a burrow system and defending it.

In all social mole-rat species levels of the reproductive skew are high and the chance of gaining reproductive status is lower than in other cooperative breeding mammals. Only few individuals are considered to ever disperse. This fact, together with a relatively long lifespan, possibly results in performance of various cooperative tasks dependent on age. However the question is, if it can be considered as specialization.

In naked mole-rats, juveniles started to contribute to maintaining tasks soon after weaning. In age of around five weeks, they performed activities as **soil removing** or **food carrying** (Jarvis & Sherman 2002).

Nest-building behaviour was frequent around 9 months of age of helpers, later the levels of contribution decreased rapidly among nonbreeders of both sexes in Damaraland mole-rats. Increase in body mass resulted in reduction of nest building behaviour and increase of food carrying and digging (Thorley et al. 2018).

Another task performed by young members of colonies of naked mole-rats and Damaraland mole-rats is **babysitting** (Lacey & Sherman 1991, Bennett 1990, Mooney et al. 2015). Young helpers also cleaned and warmed up pups (Bennett & Jarvis 1988).

Pup carrying is an activity during which are newborns removed. In *Fukomys damarensis* contribution to pup carrying firstly increased rapidly and then decreased as individuals aged (Zotzl et al. 2018). Pup carrying behaviour was also sex dependent. Female breeders were more likely than female non-breeders to carry pups in *Fukomys damarensis*, whereas male breeders were less likely to carry pups than male non-breeders. When nonbreeders were compared, females tended to carry pups more than males (Zotzl et al. 2018).

Another specific task is **colony defence**. It is usually performed by individuals with a high body mass (Burda 1990, Lacey & Sherman 1991, Mooney et al. 2015). In naked mole-rats, the number of attacks on predator was higher in heavy individuals (Lacey & Sherman 1991). Mooney et al. (2015) found that aggression is positively correlated with body fat. Individuals who defended colony were not those who came into contact with the predator first (Lacey & Sherman (1991) but were summoned by a specific vocalization (Pepper 1991).

Volcanoing is one of the most dangerous tasks performed by social mole-rats. During the process of soil removing, last member of the working chain dig out the soil and is highly exposed to predation. This task was found to be performed by the largest individuals of the colony (Braude 1991).

Performing colony defence and volcanoing by large individuals is indirectly confirmed by capture order. Large individuals were one of the first who were caught (Lovegrove 1988, Brett 1991, Jacobs et al. 1991, Yeboah and Dakwa 2002).

1.4.2.2 Task switching

In meerkats, the division of labour change with age and individuals **switch** among tasks as they grow and age (Clutton-Brock et al. 2003). It had not been tested if subordinates of mole-rats perform the same tasks consistently across time or even if they switch tasks based on the changing demands of the colony. Mooney et al. (2015) conducted an experiment with naked mole-rats and removed most active performers of pup care and colony defence. It was found that mole-rats switch between tasks after removal and these changes are age dependent (Mooney et al. 2015). After removal, younger animals started to engage into digging and pup care and older animals were more aggressive towards conspecifics than before. Working behaviour (digging) remained the same before and after removal. Younger individuals therefore replaced removed diggers. It was important to reveal that mole-rats are able to adjust their behaviour to the removal of other colony members and switch between tasks when needed which deny the existence of permanent behavioural castes.

1.4.2.3 Task specialization

Division of labour, task specialization and even morphological specialization allowing performing specific tasks more effectively are common in nonvertebrates (Wilson 1971). In insect societies individuals perform different tasks such as brood care, colony defence or foraging (Holldobler & Wilson 1990). In some social insects, specialization is not permanent and task allocation changes with age and individuals can switch between tasks, like in honeybees, *Apis mellifera* (Johnson 2008) or some termites (Korb & Hartfelder 2008).

Meerkats perform cooperative behaviours similar to those of mole-rats, including allolactation, babysitting and pup feeding as well as burrow digging and group defence. Males contribute more to colony defence and guarding, whereas females contribute more to babysitting and pup feeding (Clutton-Brock et al. 2002). But meerkat helpers also perform the full range of activities, and show no evidence of individual specialization in specific forms of cooperation (Clutton-Brock et al. 2002).

Mooney et al. (2015) stated that the performance of cooperative behaviours in naked mole-rats was stable across time, therefore it can be considered as the **task specialization**. However, Thorley et al. (2018) pointed out that this time was too short to claim specialization. Mooney et al. (2015) also added a disclaimer that “specialized colony defenders” may never abandon colony maintaining tasks. However, to define specialization investment in one cooperative behaviour must trade off against investment in other forms of cooperative behaviour (English et al. 2015).

Contrary to Mooney et al. (2015), Thorley et al. (2018) did not find any trade-offs between cooperative activities that would suggest specialization in Damaraland mole-rats. Surprisingly they found that helping in one activity correlates with helping in another activity, with age and body mass as determinants of overall contribution to all tasks. Level of contribution was positively correlated across activities so that some individuals are consistently ‘generous’ and others were consistently ‘mean’ (Clutton-Brock et al. 2003 for meerkats, Zottl et al. 2016, Thorley et al. 2018 for Damaraland mole-rats). It was also found that repeatability of cooperative behaviour was low; suggesting no specialization (Zottl et al. 2016).

Distribution of cooperative behaviour among individuals in Damaraland mole-rats resembles that in meerkats, *Suricata suricatta*. It was suggested that helpers of mole-rats and meerkats do not specialize in specific tasks but rather vary in activities performed. (Clutton-Brock et al. 2003, Zottl et al. 2016, Thorley et al. 2018).

1.5 Mole-rats: Age or caste polyethism?

There are several factors which may influence activity and contribution to specific tasks among individuals of a mole-rat colony. These are particularly **body mass, age, sex and reproductive status**. Existence of discrete behavioural castes differing in age and/or body mass was never really confirmed. On the other hand, activity level related to age and/or body mass and sex was detected repeatedly (Fritzsche & Gattermann 2002, Lacey & Sherman 1991, Lövy et al. 2013, Mooney et al. 2015 Schielke et al. 2012).

1.5.1 Failures to define behavioural castes which differ in body masses

Although persisting differences in the amount of work performed among different cohorts in mole-rat families were supposed, the existence of behavioural castes of frequent and infrequent workers was not confirmed in the following studies.

In *Fukomys mechowii*, the amount of work performed was not dependent on body mass, and most of work was performed by the two oldest individuals, despite this fact, terms frequent and infrequent workers remained (Wallace & Bennett 1998).

In several studies the amount of work was not related to body mass and new term, **casual workers**, was defined. Casual workers sometimes include lighter individuals which should belong to the caste of frequent workers (cf. Bennett 1989, Wallace & Bennett 1998). Casual workers were described in *Cryptomys hottentotus hottentotus*, (Bennett 1989, Bennett 1992) and in *Fukomys darlingi* (Gabathuler et al. 1996).

More inconsistent results were obtained in the study on *Fukomys damarensis*. Despite infrequent workers were characterised by higher body mass in many studies (Bennett 1990, Bennett & Jarvis 1988, Jacobs et al. 1991); frequent workers had greater body masses than infrequent workers in the study of Gaylard et al. (1998). All of those workers, including the breeding pair were furthermore socially dominant. Author explained this phenomenon by the fact that the particular colony was recently founded. After having several litters, dominant individuals decreased their activity. In recently founded colonies larger animals may constitute the initial workforce and participate in cooperative tasks more. Higher activity of bigger individuals was also found in *Fukomys anselli* Burda (1990), therefore other factors can be involved.

A similar situation can possibly happen when work demands of a colony increase, for instance after rains, when the opportunity for rapid burrow system extension comes (c.f. Scantlebury et al. 2006).

1.5.2 Continual changes in activity

The main difference between castes and age polyethism is that castes are discrete groups of individuals which differ in behaviour, physiology or morphology whereas in societies with age polyethism individuals remain plastic.

When activity was measured by radio frequency identification (i.e. the methodology used in the present thesis) in *Fukomys anselli*, it was found that activity of older members of the group was lower than activity of younger individuals. Authors described this trend as “general mammalian pattern”. No difference in activity between breeders and nonbreeders was found in this study (Schielke et al. 2012).

It was found (noteworthy, on the largest sample size ever tested) that investment into cooperation changes as individuals age and grow (Zotzl et al. 2016). Fast growing individuals with higher body masses invested more into cooperative activities (digging, nest building and food carrying) during the first year of observation, however, this situation changed the following year, when females with low body masses increased investment to cooperative behaviour.

Investment to cooperative behaviour did not depend on body mass after individuals were 600 days old and reached asymptotic body mass. Frequency distribution of total cooperation showed no bimodal distribution as would be in the presence of castes. Authors assumed that there is no evidence for castes in Damaraland mole-rats and age is the main predictor of performing cooperative behaviour. Individuals do not specialize in certain tasks but varied in their contribution to cooperative activities (Zotl et al. 2016). Total activity increased until individuals reached one year and then decreased (Thorley et al. 2018).

Similar trend in contribution to different tasks was found in meerkats, the contributions of helpers increase with age. Juveniles first began to make little contributions to babysitting and feeding subsequent litters of pups after they were 6-months-old and increased levels of helping up to the second year of life. Subsequently, their contributions decline before dispersal (Clutton-Brock et al. 2002).

According to Thorley et al. (2018) it is hard to distinguish castes from developmental processes leading to differences in performing a cooperative behaviour and contributions to it.

The greatest weakness of laboratory studies, where castes were described is the artificial criterion for the division into castes. Castes were determined on the basis of how many percent of working activities an individual performed within a family. The value of approximately 5% is the critical value for caste determination in many studies (Gaylard et al. 1998, Bennett & Jarvis 1988, Jacobs et al. 1991). The division into castes is therefore based on one critical artificial value and it is impossible to distinguish between castes and possible continuum in activity, which might occur (c.f. Tab. II.).

1.5.3 Contribution to cooperative tasks in males and females

Differences in helping effort may also differ between sexes. In many cooperative breeding societies, members of the philopatric sex generally contribute more to rearing young, whereas members of non philopatric sex contribute more to colony defence and guarding (Clutton-Brock et al. 2003).

For example, in meerkats and brown hyenas, females may remain and breed in their natal group, and females helpers typically contribute more to rearing young than males, whereas in African wild dogs, where males may remain and breed in their natal group, males generally contribute more than females to rearing young (Creel & Creel 2002).

In mole-rats differences in activity or contribution to different tasks also sometimes differ between sexes. The effect of sex was confirmed, with the tendency of females to be more active than males, in *Fukomys anselli* (Schielke et al. 2012, Fritzsche & Gattermann 2002). Females performed more digging and food carrying whereas males explored the surroundings more often. In Damaraland mole-rats, females invested more into nest building, digging and pup care than males (Zottl et al. 2016). Contribution of males and females to cooperative tasks can also be influenced by group size. In larger groups males contribute less (Thorley et al. 2018).

Differences in activity between sexes were also found in *Fukomys mechowii*. Females were more active than males (Dammann et al. 2011). However, this result is in conflict with another study of the same species, where males were more active than females (Wallace & Bennett 1998) but in accord with most studies on *F. anselli* as shown above.

Still, no differences between sexes in their activity and work levels were found in majority of studies (Jarvis 1981, Bennett 1990, Lacey & Sherman 1991, Jacobs et al. 1991, Gaylard et al. 1998, Lövy et al. 2013), which indicates that the effect of the sex is generally weaker than the effect of age/body mass.

1.5.4 Studies from field

Despite the fact that mole-rats are objects of many studies, only few of them were conducted under natural conditions, as it brings many difficulties. Mole-rats in the field are tracked by radio-telemetry, which do not provide specific information about task performed, however differences in spatial activity and time spent in a nest can be evaluated.

Different space-use patterns may be an indicator of the activity performed. For instance, if individuals use larger home-ranges it may signalize, that those individuals explore or patrol.

In all field studies, breeders were the least often detected outside their nest (Lovegrove 1988, Lövy et al. 2013, Šklíba et al. 2016). In *Fukomys mechowii*, the breeding male spent the most time in the nest, followed by the largest nonbreeders, and he was active only half of the time when compared to other family members. Additionally he used only 20% of the family home-range and was often located in food storages (Lövy et al. 2013). Similarly breeding males in *Fukomys anselli* used smaller home-ranges than the rest of the family (Šklíba et al. 2016).

Breeding female was caught only once during field studies. Breeding female was the most active in the area close to the nest. Surprisingly, space-use patterns of the breeding female were not very different from the patterns of other family members (Šklíba et al. 2016).

Differences in activity were also found between sexes in studies from field. Female nonbreeders in *Fukomys damarensis* and *Fukomys mechowii* tended to exhibit higher levels of outside-nest activity as compared with other family members (Lovegrove 1988, Lövy et al. 2014). Moreover, female nonbreeders in *Fukomys anselli* were located the furthest from the nest (Šklíba et al. 2016).

Studies from field are in concordance with studies from laboratories in several regards. Breeding individuals contributed the least to cooperative tasks related to colony maintenance (cf. Jarvis 1981, Wallace & Bennett 1988, Bennett & Jarvis 1988, Bennett 1990, Burda 1990, Fritzsche & Gattermann 2002). Large individuals were less detected outside of the nest (Lövy et al. 2013, Lovegrove 1988), which corresponds with lower activity of large individuals in captivity (Lacey & Sherman 1991, Fritzsche and Gattermann 2002, Dammann et al. 2011, Mooney et al. 2015, Schielke et al. 2012). Body mass of *Fukomys anselli* was positively correlated with time spent in the nest and negatively with the activity far from the nest (Šklíba et al. 2016).

Contrary to studies from laboratories, castes have not been described in field studies and Šklíba et al. (2016) suggested age polyethism as an explanation for different space-use patterns.

1.6 Limits of laboratory experiments on mole-rats

There are several limitations of studies from laboratories. Some of them are common for all, for instance space limitation and facilitation or even not enabling to perform specific tasks. In natural conditions mole-rats work every day on digging and repairing tunnels. However, many studies were conducted in stable small burrow systems (Bennett & Jarvis 1988, Bennett 1990, Jacobs et al. 1991, Bennett 1992, Gaylard et al. 1998,). In the first study, where eusociality and castes were described, the author even did not use any burrow system and observed family was studied in an aquarium (Jarvis 1981).

In many studies, mole-rats were placed into an artificial burrow system but no additional stimulation of work or extra space where digging and material transporting could be observed was provided (Bennett and Jarvis 1988, Bennett 1990, Jacobs et al. 1991, Moolman et al. 1998). In studies in which castes were described, only one used enlarged burrow system with a special space for digging, where soil could be also excavated out of the burrow system (Wallace & Bennett 1998). Desmet et al. (2012) confirmed that when a “digging unit” was connected mole-rats, *Fukomys micklemi*, increased their activity.

Some types of behaviour which are otherwise hardly observable under unnatural conditions can still been stimulated. For example, to stimulate colony defence, strange conspecifics were introduced to family members (Burda 1990, Mooney et al. 2015). Or even a snake was used in the study of Lacey & Sherman (1991) to stimulate anti-predator defence. However, stimulating of work behaviour was hardly ever done, even though working activity was studied. Interesting system was used in the study of Thorley et al. (2018). Several vertical tubes were connected to the system and substrate was added through these tubes, consequently mole-rats moved the substrate to a waste box.

One of the biggest problems of many studies is small sample sizes. Observations were often confined to one family with no repetition. (c.f. Tab.: I. and Tab.: II).

Tab. I: Differences in activity according to reproductive status, sex and body mass in several mole- rats species.

F-field studies, L- laboratory studies, N/A- not available, N.D. - no differences

Species	T	N _(groups) /	Activity of breeders		Activity according to sex	Correlation of activity and body mass	Source
	L	n	B.Female	B.Male			
<i>H. glaber</i>	L	/40	N/A	Low		Negative	1
<i>H. glaber</i>	L	1/31	N/A	N/A	N.D.	Negative	2
<i>H. glaber</i>	L	3/48	N/A	N/A	N/A	Negative	3
<i>F. mechowii</i>	T	1/5	N/A	Low	N.D.	Negative	4
<i>F. mechowii</i>	L	1/9	Low	Low	++ males	N.D.	5
<i>F. mechowii</i>	L	18	Average	Average	++ females	N/A	6
<i>F. damarensis</i>	T	1/5	N/A	Low	N.D.	Negative	7
<i>F. damarensis</i>	L	1/11	Average	Low	N.D.	Negative	8
<i>F. damarensis</i>	L	1/11	Average	Low	N.D.	Negative	9
<i>F. damarensis</i>	L	1/17	N/A	N/A	N.D.	Positive	10
<i>F. damarensis</i>	L	2/33	Low and high	Average	N.D.	Negative	11
<i>F. anelli</i>	T	5/17	Average	Low	N.D.	Negative	12
<i>F. anelli</i>	L	7/45	Low	Low	N/A	Positive	13
<i>F. anelli</i>	L	1/11	Low	Low	N/A	Negative	14
<i>F. anelli</i>	L	6/47	Low	Average	++ females	Negative	15

1: Jarvis 1981, 2: Lacey & Sherman 1991, 3: Mooney et al. 2015, 4: Lövy et al. 2013, 5: Wallace & Bennett 1988, 6: Dammann et al. 2011, 7: Lovegrove 1988, 8: Bennett & Jarvis 1988, 9: Bennett 1990, 10: Gaylard et al. 1998, 11: Jacobs et al. 1991, 12: Šklíba et al. 2016, 13: Burda 1990, 14: Fritzche & Gattermann 2002, 15: Schielke et al. 2012

Tab. II: Laboratory studies in which castes of frequent and infrequent workers were described

Species	N _(groups) / n _(individuals)	B. Female	B. Male	FW	IW	Differences in body masses (FW vs. IW)	Source
<i>H. glaber</i>	1/40	N/A	N/A	N/A	N/A	N/A	1
<i>F. mechowii</i>	1/9	3,4	3,4	15-22	7,11	273/236	2
<i>F. damarensis</i>	1/11	5,4-7,7	0,9- 1,5	12- 17,8	5	83/120	3
<i>F. damarensis</i>	1/11	8,2	1,1	13-20	6	80/120	4
<i>F. damarensis</i>	2/29	1,8-14	6,2-8,9	5,4- 13,2	0-5	93/127	5
<i>C.h. hottentotus</i>	1/8	30,4	3,4	12- 22,3	5,5-6,8	88/80	6

1: Jarvis 1981, 2: Wallace & Bennett 1998, 3: Bennett 1990, 4: Bennett & Jarvis 1988, 5: Jacobs et al. 1991, 6: Bennett 1992

Results can be also influenced by the type of activity recording. In several studies (Bennett & Jarvis 1988, Bennett 1990, Jacobs et al. 1991, Bennett 1992, Moolman et al. 1998) activity durations were not recorded. However this style of recording may hide the differences in individual's activity. Because some individuals, who work more often but for shorter periods of time can therefore, be considered as infrequent workers (cf. Desmet et al. 2012).

According to definition of (Mitchener 1974) castes should be permanent across time. However, the stability (persistence) of an individual's cooperative activity level was rarely tested. Only in few recent studies repeated observations of particular individuals were conducted. Importantly, it was found that mole-rats maintain behavioural plasticity through development or in changing conditions (Mooney et al. 2015, Zottl 2016, my study). They also found that individuals' contributions to different tasks were correlated positively, whereas in case of castes it should be correlated negatively.

1.7 Aims of my study

The aim of my study was to design and carry out a laboratory experiment which would reveal polyethism in locomotor activity and work within captive families of social mole-rat and enable to test the relation of activity and age, body mass, sex and reproductive status. Also to reveal how it can be affected by various stimulations of work behaviour and by change of the family composition. As the experimental subject I used five captive families of *Fukomys darlingi* individually marked with RFID (radio frequency identification) transponders. The experiment was conducted in an artificial Perspex burrow system equipped with six readers (antennas) automatically detecting each passage of each individual through a tunnel.

This method provides more accurate information about activity compared to the data collected by personal observations which were carried out only a few hours a day (Bennett and Jarvis 1988, Bennett 1990, Bennett et al. 1990, Jacobs et al. 1991 etc.). The disadvantage is that this type of activity recording does not provide specific information of activity performed. Therefore, I also combined this approach with video-recording mole-rats' working behaviour and subsequent analysis of the recordings.

The specific aims of my study were:

- 1) To reveal polyethism within mole-rat families as differences in individuals' overall locomotor activity and to relate these differences with age/body mass, sex and reproductive status. Based on the above literature review on mole-rat polyethism I predicted that in most families the amount of locomotor activity would be negatively related to body mass with little or no effect of sex and with breeding individuals to be the least active individuals in the families. Alternatively, activity of females could be higher than that of males, as females are more philopatric sex in mole-rats.

2) To test how the polyethism in overall locomotor activity within the mole-rat families will be affected by stimulating the activity by means of adding more substrate to the burrow system. We predicted that the general pattern of polyethism with smaller individuals being most active will either remain or be replaced by a situation when larger individuals become more active (such as is work demands of the family increase).

3) To test whether the working behaviour can be stimulated directly by adding a hard material for digging and whether differences in individuals' amounts of work done correspond with the differences in their overall locomotor activity detected before. I predicted that the amount of work detected will be correlated with the general locomotor activity under increased amount of substrate.

4) To test how removal of roughly $\frac{1}{4}$ of the most active workers out of each family change activity of remaining individuals when the same experimental design is repeated. I predicted that the removal of the most active/working individuals would increase the activity of individuals who had been less active previously substituting their role in the particular family.

5) To evaluate efficiency of the experimental design to study mole-rat polyethism and to propose its further improvement in order to simulate natural conditions potentially promoting polyethism.

2 MATERIALS AND METHODS

2.1 Studied species

I studied Mashona mole-rat, *Fukomys darlingi*, Bathyergidae, Rodentia. Mashona mole-rats inhabit scrub and open woodlands with mean annual rainfall exceeds 700 mm. It is a social species, which members live in small colonies with one breeding pair and about three successive litters. The gestation period of a breeding female is from 56 to 61 days and litters are small and usually contain up one or two newborns. Mashona mole-rats feed on geophytes and roots and do not show signs of sexual dimorphism (Bennett et al. 1994).

2.2 Experimental room, experimental burrow and the recording system

Experiments were conducted in an experimental room at the Department of Zoology, University of South Bohemia. The period of light was set from 7 a.m. to 7 p.m. The temperature in the room ranged from 24 to 25 °C. All tested individuals were weighted before and after each experiment. Tested families were fed twice during the experiment. For the first time, food was placed inside the food chamber before each family was placed into the experimental burrow system. For the second time, food was placed inside food chamber during substrate addition. More specifically constant amount of carrots, potatoes, small pieces of apples, beetroots and sweet potatoes and dried fodder- Darwin was added each time.

Mole-rats families are standardly housed in glass aquariums and were placed into the experimental burrow system only for the time the experiments were conducted. I constructed the system to simulate ideal conditions for digging in tunnels similar to natural conditions. It was decided to use a burrow system in the shape of infinity eight so substrate could be transported around.

The system was 4.3 meters long. It contained one nest (20x20x20 cm) and one food chamber (28x28x28 cm) (Fig. : 3). The nest chamber was smaller in size and it was always used as the nest by the whole family.

Activity of all family members was recorded by Trovan technology electronic identification system. This system contains one reader and six antennas (units). These antennas recognise each family member thanks to microchip ID-162B ISO FDX-B applied under individual's skin. Data were continuously saved to reader's inner memory. The saved data were namely: a unit number, a date and time of passage and a transponder (microchip) code. The arrangement of antennas is shown in figure (Fig. 3)

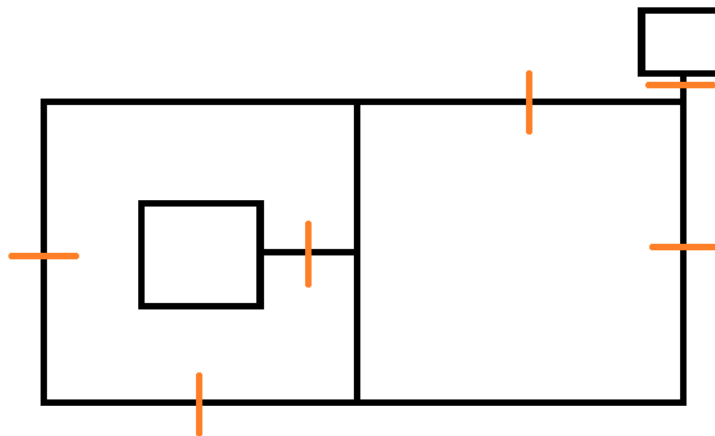


Fig. 3: The scheme of experimental burrow.

Antennas are represented by six orange lines; the larger square represents food chamber and the smaller square in the nest.

2.3 Sizes and characteristics of tested families

Five mole-rat families were tested which, except one which does not contain breeding female, consisted of reproductive pair and their offspring from different litters.

Numbers of family members are recorded in the table (Tab. 3). Family size ranged from 7 to 10 members during the first round (mean size 8.6 +- 1,342) and from 5 to 7 during the second round (mean size 6.2+- 0,837). Together 43 individuals were tested (17 males and 26 females).

Tab. III: Number of mole- rats in five families during the first experiments before removal and the second experiment after removal.

Family ID	Experiment I (n)	Experiment II (n)
1	8	6
2	10	7
3	7	5
4	8	6
5	10	7

2.4 Tested animals and repetition

Each family was tested twice. Firstly, complete families were tested, but for the second time the most active family members were removed to test if they will be replaced by remaining family members in changing colony demands. Animals which were removed from families were chosen on the basis of their performed working activity at the barrier in the Experiment I. Those individuals who performed the highest working activity during removing the barrier (this treatment is described lower in detail) and were more active in the situation with more substrate and preferably from one cohort, were denoted as potential frequent workers and separated from a family.

The percentage of removed animals were kept to ranged from 25% to 30% of the family size, because it was not possible to remove the same number of individuals from each family due to different family sizes. The repetition was carried after 30 days after the reduction of established families.

2.5 Experimental design

2.5.1 Experiment with digging substrate

The constant amount of peat was used as a substrate during all experiments. Peat is used as bedding in home aquariums as well. At the beginning of an experiment the experimental maze was loosely filled with five kilograms of peat. After 24 hours long habituation and 48 hours of measuring the locomotor activity, another five kilograms were added. This situation was followed by another 24 hours long habituation and 48 hours of measurement in more substrate.

More substrate was added in order to intensify working demands, to see possible changes in activity and to test those changes according to age/body mass, reproductive status and sex.

Tab. IV: The time schedule of experiments.

Day	Experimental situations
1	Habituation 1 (HA1)
2	Less substrate (LS)
3	Less substrate (LS)
4	Habituation 2 (HA2)
5	More substrate (MS)
6	More substrate (MS)
7	Barrier

2.5.2 Experiment with the barrier

After six days of recording locomotor activity in different amounts of substrate, I connected a new segment with a tunnel separated by a removable plastic barrier. Antennas on the experimental system were rearranged within a minute (Fig. 4). Then the two removed antennas were placed on the new tunnel allowing measuring time spent at the barrier. Finally, at the end of this tunnel, a briquette made of pressed wood sawdust was placed to stimulate work (biting).

During this experiment, mole-rats were filmed with a camera (Panasonic HC-V260) for forty- five minutes. The time spent in the tunnel with the barrier was counted for every individual using a programme created for this purpose in JAVA. Working activity was thus expressed in two ways: time biting the barrier (s) and the time spent in the tunnel with the barrier (s).

The experiment with the barrier serves for stimulating work on enlarging the burrow system. Consequently, it can be evaluated which individuals are the most active workers because this experimental set up provides direct information about the working activity.

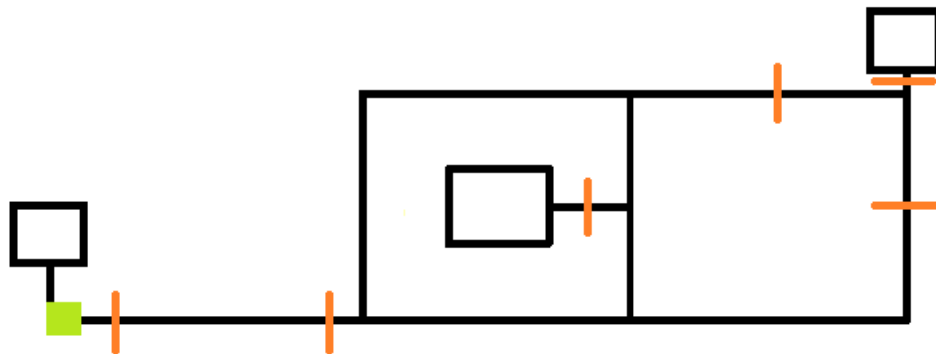


Fig. 4: The burrow system after adding the barrier (the green square).

Black lines represent tunnels. The large rectangle represents the food chamber. The smaller black square on the right is the nest and the one on the left side is only a final part of a system, which was never reached during experiments. Orange lines show the arrangement of antennas.

2.6 Collected data and terminology

We know certain information about each individual, namely sex, age and body mass. Age is known for all studied mole-rats, except two pair of breeders which were captured in field. All the other tested mole-rats were born in captivity.

Thanks to data collected by antennas placed on the burrow system electronic identification system, minimal walked distance of each individual during each experiment (with less and more substrate) could be counted. Minimal walked distance in this study represents **locomotor activity**. This term was used in the study of Schielke et al. (2012) for the same variable; therefore I decided to use the same terminology. Another data obtained during the barrier treatment is the time spent on barrier removing and time spent in the tunnel with the barrier.

2.7 Statistical analysis

2.7.1 Experiment I. - Before removal

Statistical analyses are presented according to the aims of my study. I used the similar process through the whole statistical analyzing. I plotted variables on scatter graphs, I used linear regression models to test relationships from scatter graphs and I use two- factor ANOVA to test the effect of sex or two- factor ANOVA for repeated measures to test changes caused by the removal of most active workers.

1) I plotted the initial locomotor activity to visualize differences in activity according to body mass. Individuals were differentiated in the scatter plot according to sex and reproductive status. Consequently, I tested the relationship between the initial locomotor activity and body mass using a linear regression model. The effect of sex was analyzed using a two- factor ANOVA model with the added effect of family.

2) I used two-factor ANOVA to test if individuals increased their activity in more substrate when compared to the initial locomotor activity in less substrate; family member ship was used as categorical predictor.

Then I plotted values of locomotor activities in both types of substrate amount to compare the situation in each family. I tested the differences in activity using linear regression models.

3) I used scatter graphs and plotted initial locomotor activities and times spent by removing the barrier to see, if individuals whose general locomotor activity was higher are also more active in barrier removing. Linear regression models were used to test the dependence of initial locomotor activity on barrier removing. These models were also used to test barrier removing according to body mass.

2.7.2 Experiment II. – After removal

The most active workers were removed to reveal responses in overall and working activity on individual and family level. I tested for locomotor and working activity increase caused by higher working demands to each individual due to removal.

4) Firstly, I tested for an increase in general locomotor activity after removal. The effects of removal on general locomotor activity, was evaluated using a two factor ANOVA model for repeated measures, with locomotor activities from Ex. I and Ex. II as dependent variables, and family membership as a categorical predictor.

I plot a scatter graphs to investigate whether there are the same increasing trend in activity after removal the most active workers across families and these relation were verified by linear regression models.

Secondly, I tested if remained individuals also increased their contribution to barrier removing. Two factor ANOVA model for repeated measure was used, with times spent by barrier removing in Ex I. and times spent by barrier removing in Ex II. as dependent variables and the factor of family membership as a categorical predictor.

I plotted scatter graphs to see changes in contributions to barrier removing in individual families. The relationship between times spent by barrier removing before and after removal, was consequently tested by linear regression models to statistically describe this relationship.

3 RESULTS

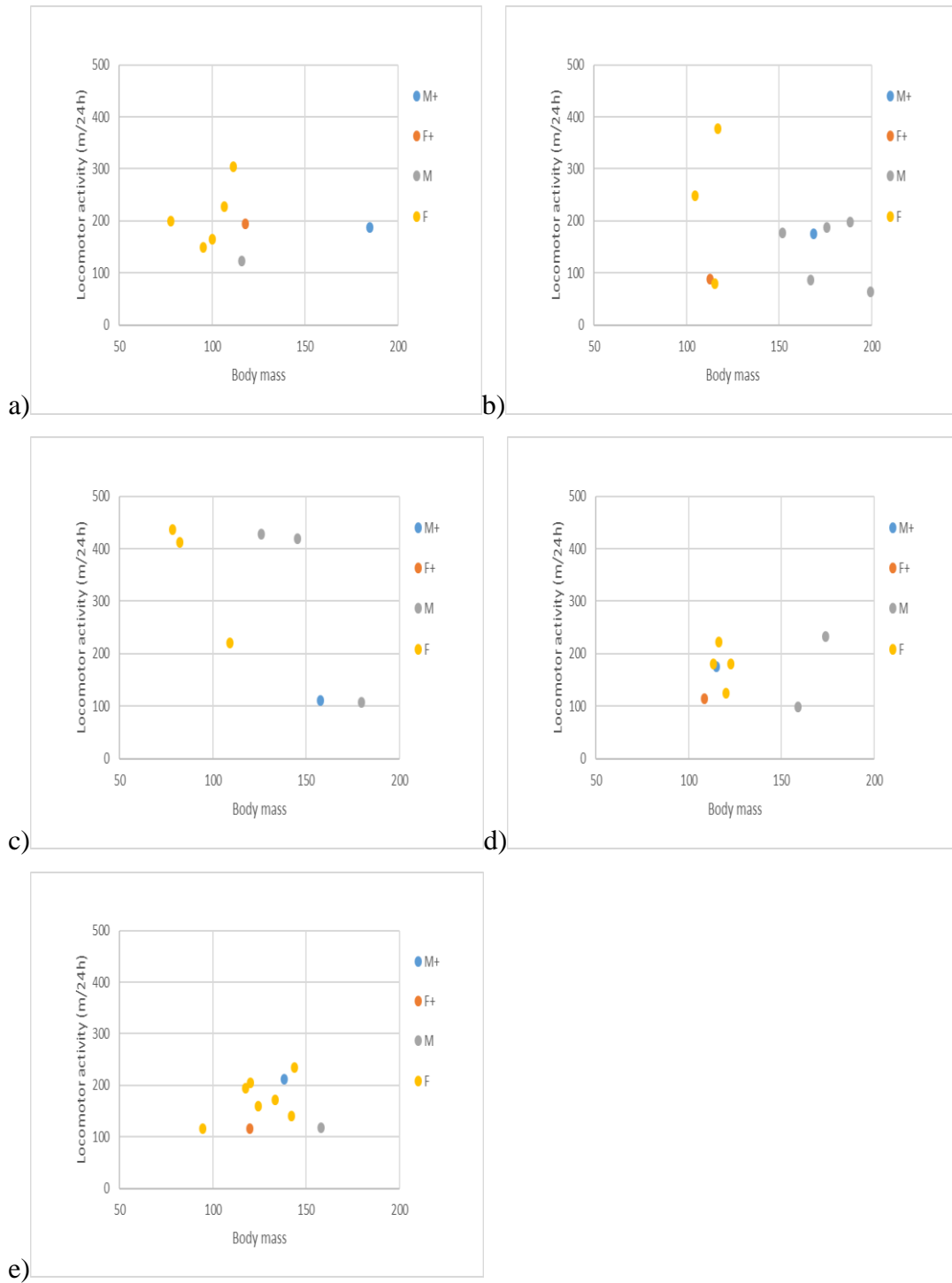


Fig. 5: Relationship of locomotor activity and body mass of individuals in each family in the initial situation with less amount of substrate.

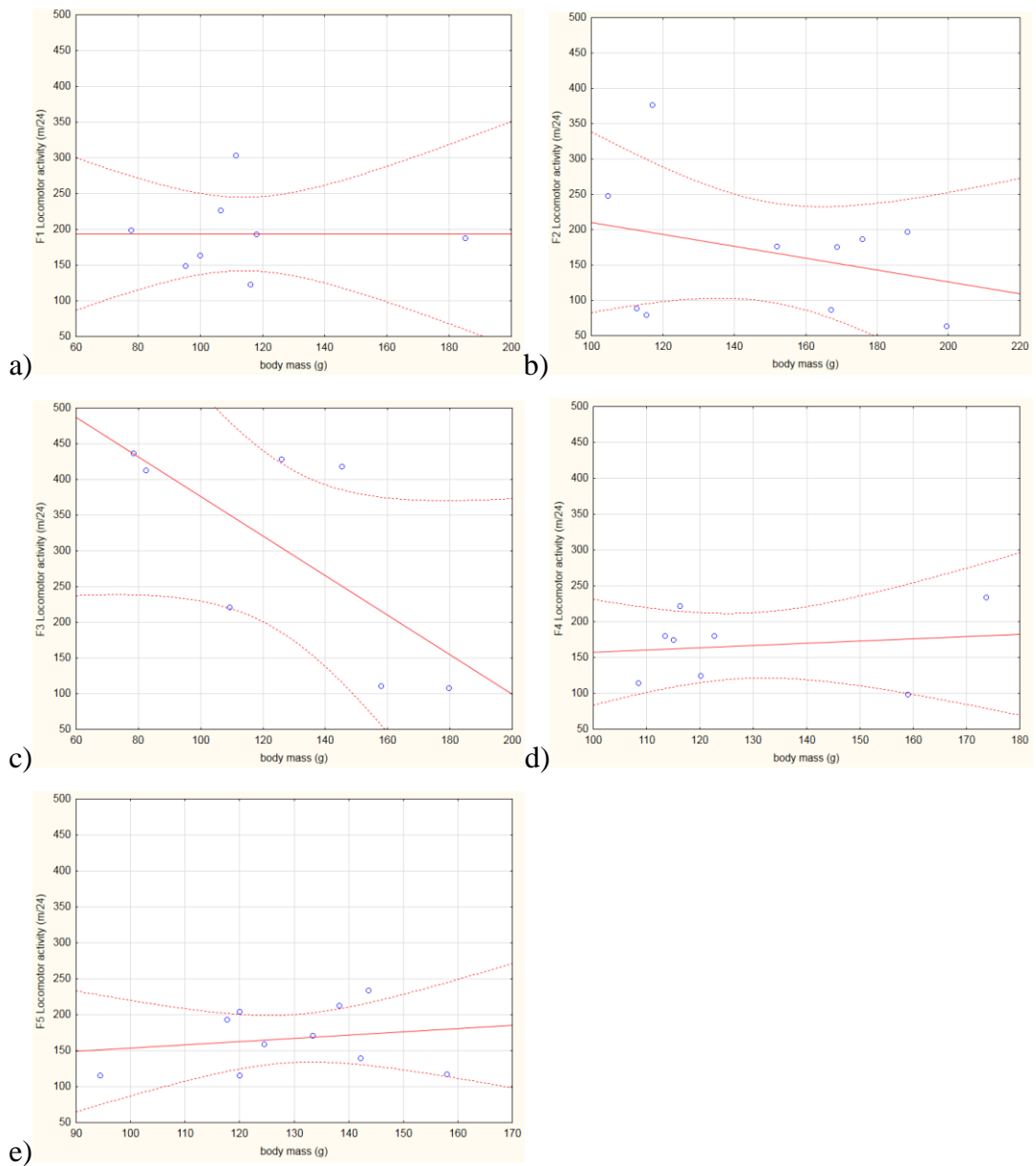
Sex and reproductive status is indicated by a different symbol colour.

a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5

1) In order to explore initial variability in locomotor activity within each family, I present several scatter plots of the locomotor activity and body mass with sex and reproductive status indicated by a different symbol (Fig. 4). I considered this as a “reference state” for further comparisons.

Figure 4 shows that the effect of sex cannot be clearly distinguished from the effect of body mass, because males had invariably greater body mass than females in each family. Another phenomenon shown in Figure 4 is that male and female breeders are not larger than nonbreeders of the same family, with the only exception of the male breeder in family 1. Similarly, initial locomotor activity of breeders in each family was within the range of nonbreeders (Fig. 4). In most of the further analyses I therefore neglect the effect of sex and reproductive status.

There was no clear relationship between the initial locomotor activity and body mass in each family although in the family 2 and 3 there was a weak (and nonsignificant) tendency to a negative relationship (linear regression; F1: $R^2=-0.167$, $F_{1,6}=0.000008$, $p=0.998$, F2: $R^2=-0.018$, $F_{1,8}=0.837$, $p=0.387$, F3: $R^2=0.369$, $F_{1,5}=4.510$, $p=0.087$, F4: $R^2=-0.139$, $F_{1,6}=1.147$, $p=0.715$, F5: $R^2=-0.088$, $F_{1,8}=0.271$, $p=0.616$).



*Fig.6: The linear regressions of locomotor activity in the initial situation with less amount of substrate according to body mass
a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5*

Sex had no significant effect on initial locomotor activity (Two-way ANOVA with Family as an additional factor: $F_{1,37}=1.828$, $p=0.184$).

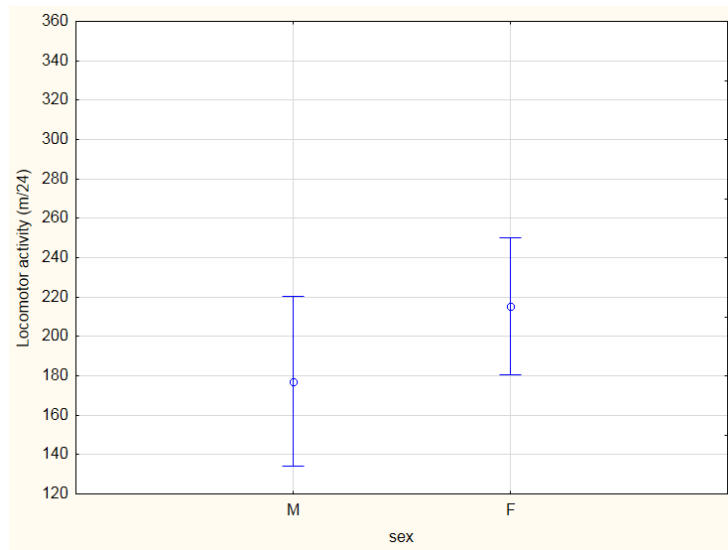


Fig. 7: ANOVA graph- difference in initial locomotor activity in males (left bar) and (right bar) females

2) I tested how locomotor activity changed after the addition of substrate. Surprisingly, locomotor activity did not change significantly after substrate addition. There was even a tendency towards decreased locomotor activity approaching significance (repeated-measure ANOVA with Family as additional factor; $F_{1,38}=2.911$, $p=0.096$).

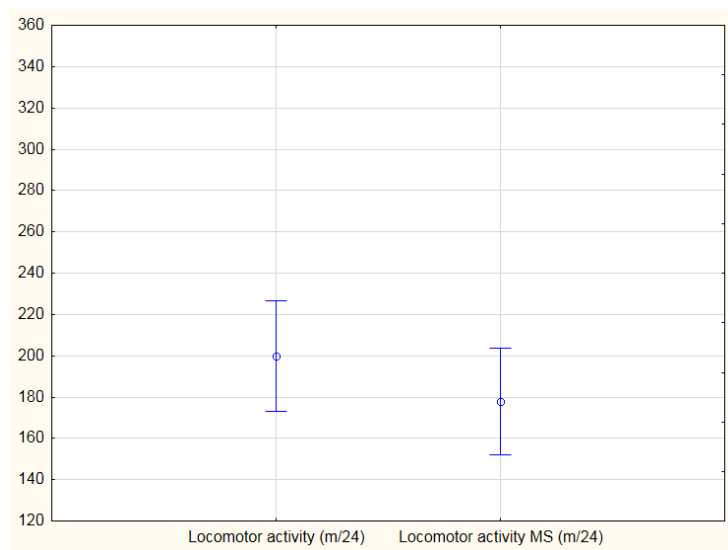


Fig. 8: ANOVA graph- locomotor activity in less substrate (left bar) to more substrate (right bar)

To explore if differences in locomotor activity between members of a family after adding more substrate remained the same or changed, I used scatter plots of the locomotor activity before and after adding the substrate (Fig. 9).

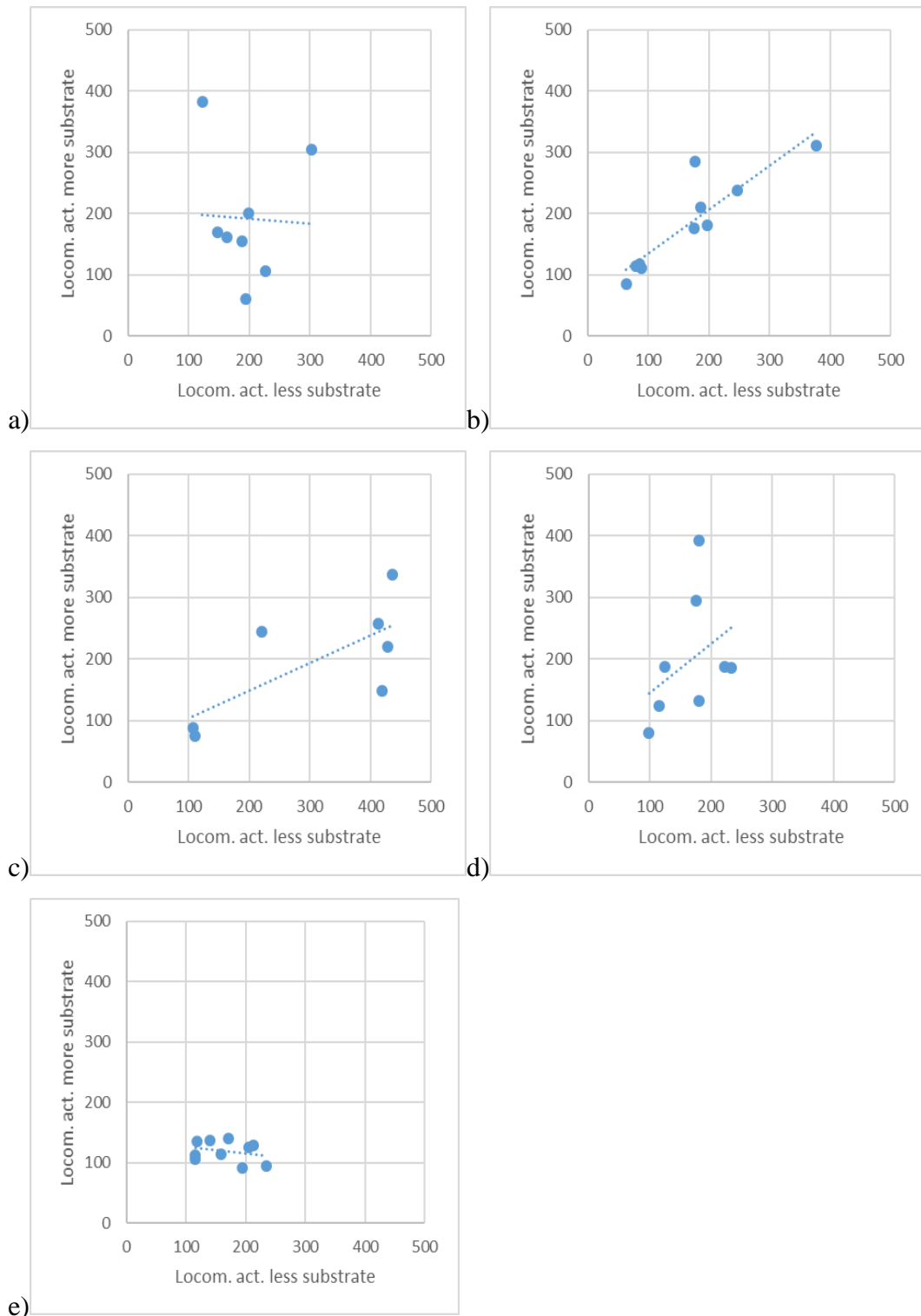


Fig. 9: The dependence of locomotor activity in the initial situation with less amount of substrate to the situation after substrate addition.

a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5

The relation was significantly only in family 2 (Linear regression, F2: $R^2=0.765$, $F_{1,8}=30.369$, $p=0.0006$) and approached significance in family 3 (F3: $R^2=0.421$, $F_{1,5}=5.360$, $p=0.068$). Positive, but not significant relationship between locomotor activity in less substrate and more substrate was also found in family 4 (F4: $R^2=0.009$, $F_{1,6}=1.061$, $p=0.343$). On the other hand, negative relationship, although not significant was found in families 1 and 5 (F1: $R^2=-0.165$, $F_{1,6}=0.010$, $p=0.922$, F5: $R^2=-0.043$, $F_{1,8}=0.628$, $p=0.451$). Interestingly, the tightest correlations were found in the same families which had the tightest correlations between initial locomotor activity and body mass (Fig. 6: b,c).

Manyfold increase of locomotor activity was detected only in two individuals of families 1 and 5, which explains the decreasing trend (Fig. 9: a,e). More specifically, it was the only nonbreeding male in family 1 which was previously the least active individual of the family, and one of the smallest nonbreeding females of the family 4 whose activity was relatively high even before adding substrate

3) To evaluate the barrier removing I started again with simple scatterplots to visualize if the most active individuals were also more active in barrier removing. The time spent by biting (removing) the barrier was log-transformed because the differences among individuals in barrier removing were even hundredfold.

Time spent by barrier removing tended to be positively related to the initial locomotor activity in four families but the relation was nearly significant in family 5 only (Linear regression, F5: $R^2=0.319$, $F_{1,8}=5.215$, $p=0.0517$). Results were not significant in families 1, 2 and 4 (Linear regression, F1: $R^2=-0.085$, $F_{1,6}=0.449$, $p=0.528$, F2: $R^2=-0.109$, $F_{1,8}=0.114$, $p=0.744$, F4: $R^2=0.108$, $F_{1,6}=1.845$, $p=0.223$) and family 3 as the only one showed opposite trend (F3: $R^2=-0.013$, $F_{1,5}=0.922$, $p=0.380$).

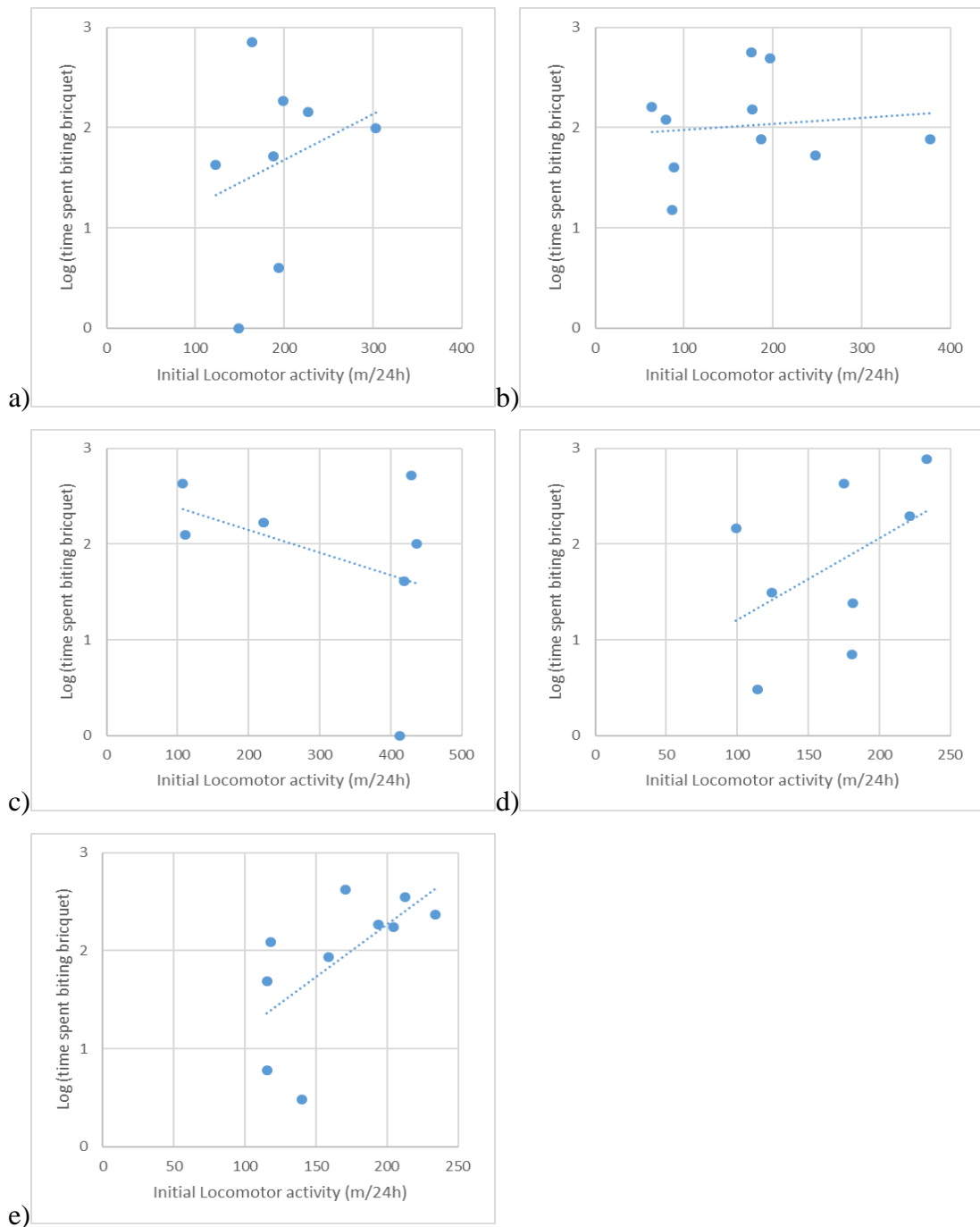


Fig. 10: The initial locomotor activity in the initial situation with less amount of substrate (space for free movement) according to the time spent by barrier removing
 a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5

Interestingly, in 4 out of the 5 families there was also a tendency towards positive relation between barrier removing and body mass (Linear regression, F1: $R^2 = -0.162$, $F_{1,6} = 0.024$, $p = 0.88$, F2: $R^2 = 0.064$, $F_{1,8} = 1.624$, $p = 0.238$, F3: $R^2 = 0.143$, $F_{1,5} = 2.003$, $p = 0.216$, F4: $R^2 = 0.221$, $F_{1,6} = 2.991$, F5: $R^2 = -0.028$, $F_{1,8} = 0.751$, $p = 0.411$). In four families there was a tendency to positive relation of barrier removing and body mass, which can signalize that barriers may served as an enrichment although it si probable that large individuals partially monopolize this activity.

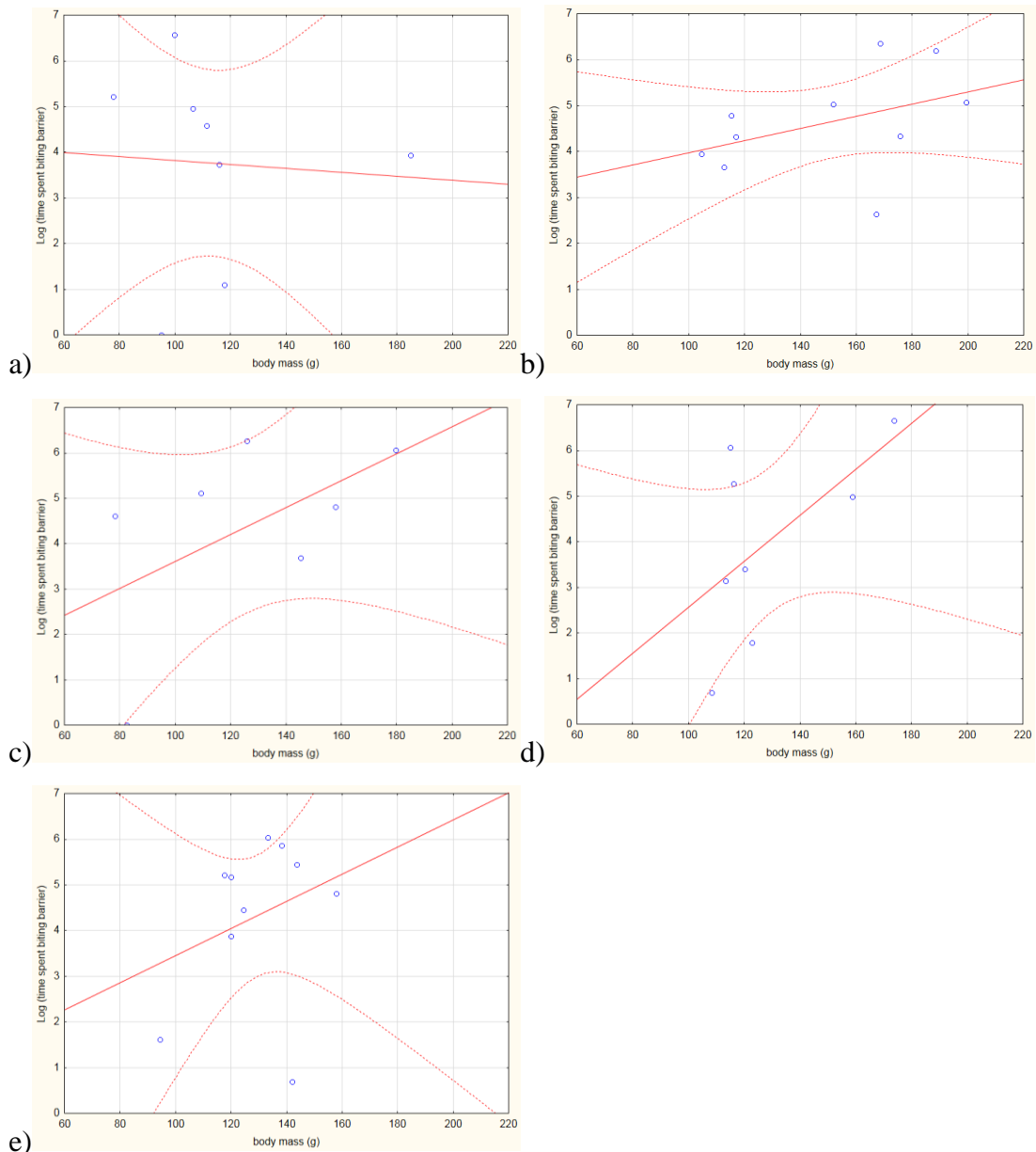


Fig.11: The linear regressions of times spent by barrier removing according to body mass

a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5

4) To reveal changes in locomotor activity before and after removal of the most active workers, repeated-measure ANOVA with Family as an additional factor was used. It was found that the locomotor activity of individuals in less substrate increased after removal of the most active workers ($F_{1,26} = 34.699$, $p = 0.000003$).

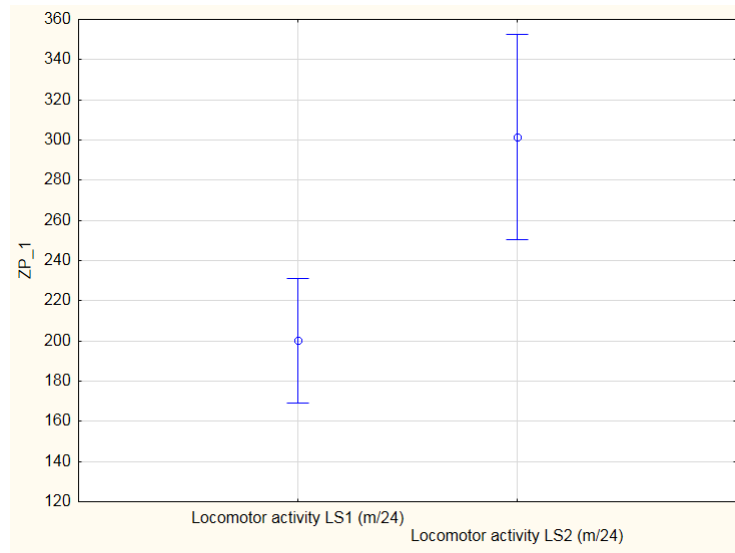


Fig. 12: ANOVA graph- locomotor activity in less substrate before removal (left bar) to less substrate after removal (right bar)

To explore which individuals increased their activity the most, I used scatter graphs (Fig. 13).

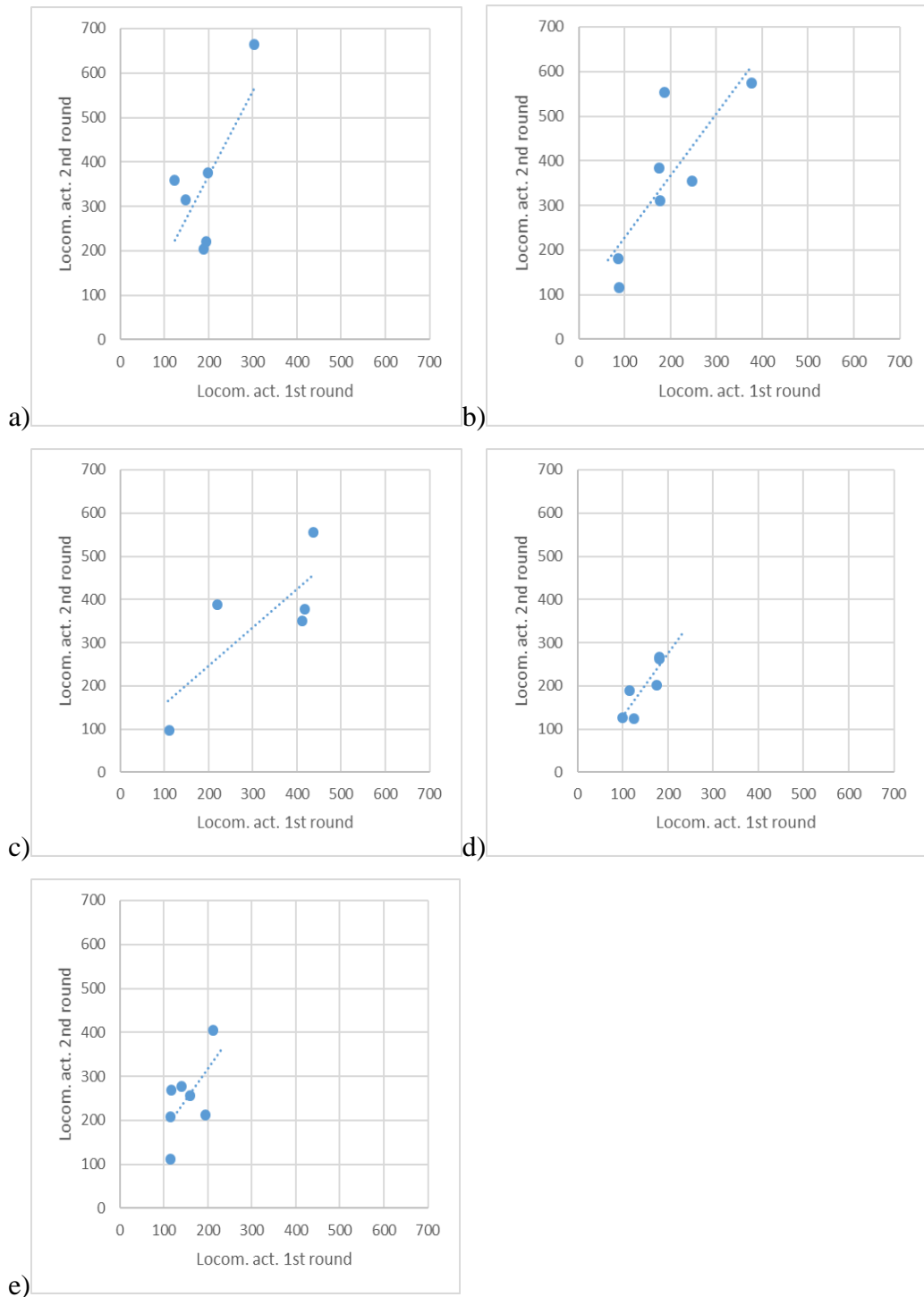


Fig. 13: The locomotor activity in the situation with less amount of substrate before removal according to the locomotor activity in less substrate after removal

a) Family 1 b) Family 2 c) Family 3 d) Family 4 e) Family 5

Locomotor activity of individuals in all families relatively tightly correlates with their activity before removal of the most active workers (Linear regression, F1: $R^2=0.380$, $F_{1,4}=4.062$, $p=0.114$, F2: $R^2=0.574$, $F_{1,5}=9.074$, $p=0.030$, F3: $R^2=0.493$, $F_{1,3}=4.885$, $p=0.114$, F4: $R^2=0.674$, $F_{1,4}=11.339$, $p=0.028$, F5: $R^2=0.286$, $F_{1,5}=3.399$, $p=0.125$). Manyfold increase of the locomotor activity following removal of some individuals was observed in six individuals from three families. Some of these animals were relatively less active before removal and some of them relatively more active.

To compare the difference in time spent by barrier removing before and after removal of the most active individuals, I used repeated-measure ANOVA with Family as an additional factor. There was a strong increase of the time spent by barrier removing ($F_{1,26}=18.561$, $p=0.0002$).

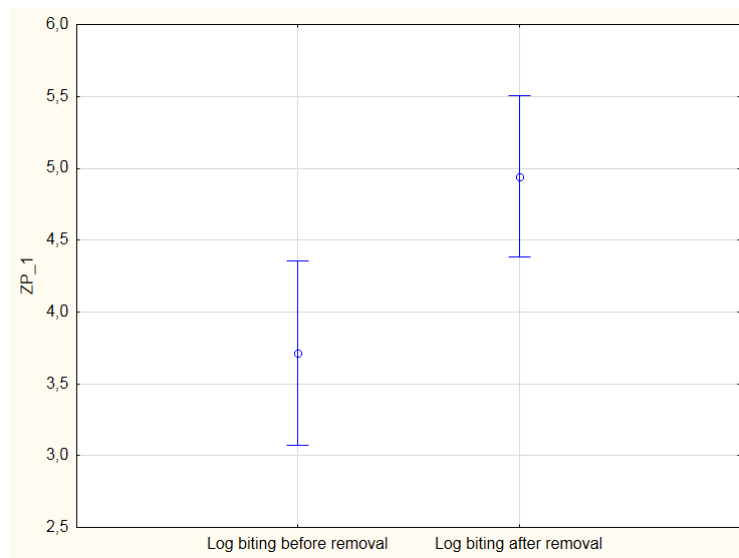


Fig. 14: ANOVA graph- time spent by barrier removing before removal (left bar) to time spent by barrier removing after removal (right bar)

Again, I used scatterplots to explore the changes of barrier removal times before and after the removal of the most active workers in individual families.

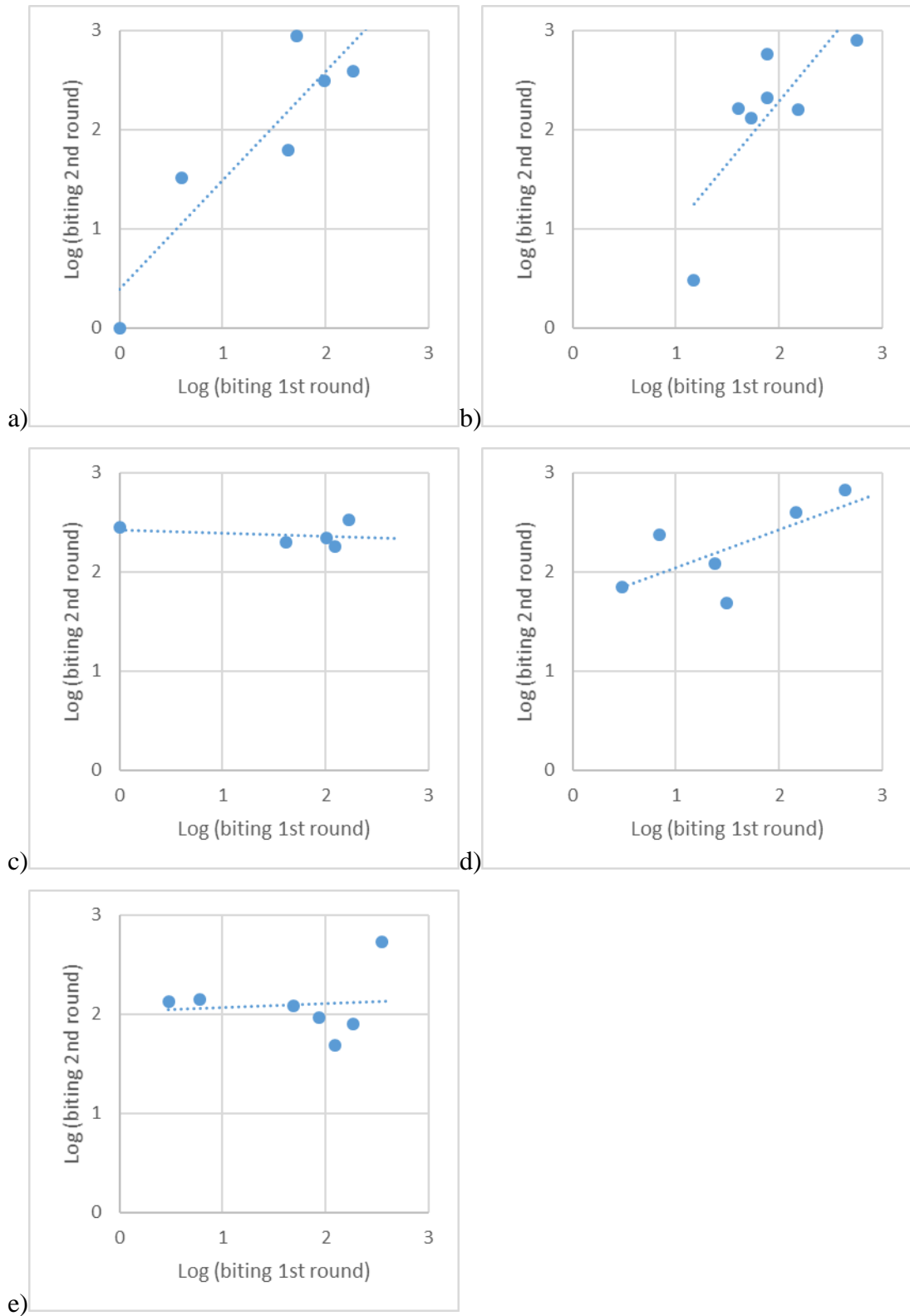


Fig. 15: Time spent by barrier removing before the removal of the most active workers to time spent by barrier removing after removal.

a) Family 1, b) Family 2, c) Family 3, d) Family 4, e) Family 5

It can be seen that times spent by barrier removing are tightly correlated in families 1 and 2 (Linear regression, F1: $R^2=0.735$, $F_{1,4}=14.852$, $p=0.018$, F2: $R^2=0.548$, $F_{1,5}=8.281$, $p=0.035$). Nonsignificant results were found in the rest of the tested families (F3: $R^2=-0.262$, $F_{1,3}=0.169$, $p=0.708$, F4: $R^2=0.350$, $F_{1,4}=3.691$, $p=0.127$, F5: $R^2=-0.191$, $F_{1,5}=0.038$, $p=0.853$). Results are not significant probably because of the fact that some individuals who spent little time by barrier removing before removal increased their investment into this task markedly after removal of the most active workers.

Many fold increase was found in six individuals, included two breeding females. However, in general individuals with low body mass tended to increase their activity the most.

4 DISCUSSION

1. Is there an evidence for polyethism in Mashona mole-rat families?

I found remarkable differences in locomotor activity within members of each family tested, but only in two of them I found the expected negative relation of the locomotor activity and body mass, even though it was weak and statistically insignificant. This is surprising, because in most of the comparable laboratory studies conducted on the genus *Fukomys*, a negative relationship was detected (Jacobs et al. 1991, Fritzsche and Gattermann 2002, Schielke et al. 2012), while only two studies reported positive relationship (Gaylard et al. 1989, Burda 1990). An unclear relationship as detected in my experiments was also detected by Bennett and Jarvis (1988), Bennett (1990) and Wallace and Bennett (1989). Interestingly, all available field radio-telemetry studies revealed a negative relationship between body mass and activity (Lovegrove 1988, Lövy et al. 2013, Šklíba et al. 2016).

In my study, the evidence for any sort of body mass-based polyethism in Mashona mole-rat is therefore unconvincing. However, the differences found between the groups members in their initial locomotor activity persisted even under situation when some of the most active members of the families were removed. This may suggest that the locomotor activity still reflects a sort of behavioural polyethism, though it might be related to body mass.

I found no clear effect of sex on the initial locomotor activity. This is in accord with most of the comparable studies. In the only studies where some effect of sex on the activity was found, the most active sex was either males (Wallace and Bennett 1998) or females (Dammann et al. 2011, Schielke et al. 2012).

In my experimental families males were invariably larger than females, which is in conflict with all literature data on *Fukomys darlingi* (Bennett et al. 1994) and other species of the genus (c.f. Grzimek 2004). I can speculate that this is a consequence of long time spent under captive condition with *ad libitum* access to highly nutritious food and relatively low work demands.

I found no clear effect of reproductive status on the initial locomotor activity. This is surprising, because in most laboratory studies, breeding males were the least active individuals of their families (Bennett and Jarvis 1988, Bennett 1990, Wallace and Bennett 1998, Burda 1990, Fritzsche and Gattermann 2002). Studies from field brought the same results; male breeders were less active nonbreeders (Lovegrove 1988, Lövy et al. 2013, Šklíba et al. 2016)

Activity of breeding females is usually lower than in other family members (Wallace and Bennett 1998, Burda 1990), but it apparently depends on the phase of the reproduction in a given family. I have to stress that in none of the families tested in my thesis the breeding female was highly pregnant or taking care of pups. On the other hand no differences in activity between breeders and nonbreeders were also found in *Fukomys anselli* (Schielke 2012, Damman and Burda 2006).

2. The effect of adding substrate into the artificial burrow system

Substrate addition did not cause expected increase in overall locomotor activity. In some families the activity of their members remained roughly similar, but in other families strong changes in the locomotor activity occurred but no predictable pattern was identified in these changes. The only significant relation between locomotor activity in less substrate and more substrate was found only in family 2 and family 3 (approaching significance). Noteworthy, negative relation between body mass and activity was also detected only in these two families.

Manyfold increase of locomotor activity was detected only in four individuals (one from family 1 and three individuals from family 5, which explains the decreasing trend (Fig. 9: a,e).

One reason for that activity did not increased, is the fact that the additional substrate was processed by the animals within a relatively short time after which part of the burrow system (including antennas) was permanently blocked. To stimulate work constantly, substrate should be continuously added and removed, however this would require a sophisticated technical solution, as already mentioned before (Wallace & Bennett 1998, Desmet et al. 2012, Thorley et al. 2018).

3. The effect of adding hard substrate

Working activity was stimulated when the new segment, with barrier at the end, was connected. Individuals started to explore the new segment immediately after a plastic wall separating the new segment was removed. They started to bite (remove) the barrier and so called digging chains, with individuals replace one another were observed.

It was found that individuals who were more active also contributed more to barrier removing, but these results were significant only in family 5. On the other hand, I found opposite trend in family 3, where individuals who were less active before, worked on the barrier removing for longer time.

Consequently I found positive relation between barrier removing and body mass, suggesting that heavier individuals were more active in barrier removing. It may be caused by an attractiveness of a new object and barrier could partly serve as enrichment. As I already mentioned I observed many types of behaviour during barrier removing, also several fights between the largest individuals and many cases of leg or tail pulling (synonym for tail tugging). All individuals who were removing the barrier were frequently pulled by other individuals. According to the recent study, tail tugging is used to monopolize work in a specific place (Kutsukake et al. 2019). Tail tugging was not an aggressive behaviour and was performed by all individuals regardless their sex or body mass, however tail tugging does not always resulted in monopolization of work and pulled individuals often did not react to this behaviour (Kutsukake et al. 2019, my observations). I agree that tail tugging was not a really aggressive behaviour; however I assume that it was a stronger signal than leg pulling.

4. The effect of the removal of the most active individuals

The reduction of number of individuals in families resulted in increase of locomotor activities of remaining individuals and the total locomotor activity (walked distance) in each family remained the same as before removal. These results can indicate that some of remained family members replace the removed individuals (cf Mooney et al. 2015).

Remained individuals also increased their contributions to barrier removing and barriers were being removed even for longer time. Individuals who contributed to barrier removing little (before removal) increased their activity. However, it may be caused by lower competition on the working place, therefore longer times of barrier removing can be a by- product of less animals alternating in this activity. Individuals who were less active in barrier removing before increased their activity however; I am not able to clearly distinguish between the increases in contribution to barrier removing as a response to removal of workers resulting in higher working demands and between less competitions resulting in increase of times spent by barrier removing. Consequently, I suggest improving the design of this experiment.

According to my results total activity of tested families differ as do responses for higher working demands on family and mainly on individual levels. Despite most individuals decrease their activity after substrate addition, four individuals increased their activity rapidly, and after removal six individuals showed much higher levels of locomotor activity than before removal. However, no clear pattern describing these changes was found. I assume that some intra family factors are involved. Activity of an individual mole-rat can be influenced by many factors. For example colony size, how is it established, age structure, sex ratio, its position in the hierarchy structure and dispersal tendencies should definitely contribute to explain the variability unexplained by age/body mass or sex. For example in Damaraland mole- rat the difference between newly formed and established colonies, with heavier individuals more active in a new established family (Gaylard 1998), the effect of dominance hierarchy was considered in this study and also in Jacobs et al. (1991), Gabathuler et al. (1996), Moolman et al. (1998), Wallace & Bennett (1998).

5 CONCLUSION

My experimental design revealed individual differences in locomotor activity within each of the tested family of Mashona mole-rats that were persistent even under reduced number of individuals and can therefore be considered as polyethism. However, the polyethism was not related to body mass as had been predicted. The differences in locomotor activity were mostly maintained when a segment with a hard substrate (briquette) was introduced and time each individual spend biting the barrier was measured. The experimental design proved to be potent to address several biologically interesting questions concerning mole-rat social behaviour, polyethism and cooperation; however some improvements need to be done to avoid problems identified in my thesis.

Suggested improvements of the experimental setting:

I) I would suggest use the same system as in the study of Thorley et al. (2018) with vertical tubes for substrate addition and I would also reconsider a waste box (cf Wallace & Bennett 1998, Desmet et al. 2012, Thorley et al. 2018)

II) The barrier is a great way to stimulate work, we considered putting some food behind the barrier, but we neglected this variation because of possible enrichment. Even though barrier was obviously attractive for almost all tested mole rats (except breeding females). Hence more tunnels with barriers should be connected at once to be available to all family member regardless their position in dominance hierarchy.

III) Also I should considered habituation even for the barrier treatment. Therefore to simulate the system blocking, which was my goal, I should put more briquettes in a row. I would propose to perform this experiment separately and observe each family for more frequently and for longer time and to record these initial activities but also to repeat recording after at least one hour.

V) Another challenge can be to create an ethogram, which could be used for evaluation of similar experiments which focus on working activity.

I observed several types of behaviour, from digging chains during which individuals performed activities such as tail pulling, leg pulling, back biting, passing side by side/over/under, head scratching, chasing, reverse ramming and explorative behaviour like “sniffing the barrier”, “slow testing bites”, “ quick reverse ramming” and more.

Jacobs et al. (1991), besides describing castes based on work created relatively complex ethogram with 17 behavioural interactions for standard situations in the system (Allogrooming, Gaping, Biting, Sparring, Co-operative feeding, Passing side by side, Head scratching, Chasing, Thrust gaping, Purposive pass under, Competitive feeding, Reverse ramming, Head lowering, Dragging, Resisting, Shunting, Passing over, Ramming, Head pressing, Retreating, Vocalized hopping). In my opinion barrier served at least partly as enrichment, mainly at the beginning, therefore all other behaviour performed, except biting, can be evaluated and serve as a possible experiment for describing dominance hierarchy of families.

6 BIBLIOGRAPHY

- Aoki S. 1982. Soldiers and altruistic dispersal in aphids. UK: Westview Press.
- Anderson C, Franks N. 2001. Teams in animal societies. *Behav Ecol.* 12(5). 534–540.
- Batra SWT. 1966. Nest and social behaviour of halictine bees of India. *Indian J Entomol.* 28:375-393.
- Bennett NC. 1989. The social structure and reproductive biology of the common mole-rat, *Cryptomys h. hottentotus* and remarks on the trends in reproduction and sociality in the family Bathyergidae. *J Zool.* 219: 45-59.
- Bennett NC. 1990. Behaviour and social organization in a colony of the Damaraland mole-rat *Cryptomys damarensis*. *J Zool.* 220: 225-248.
- Bennett NC. 1992. Aspects of the social behaviour in a captive colony of common mole-rat *Cryptomys hottentotus* from South Africa. *Z Saugetierkd.* 57: 294-309.
- Bennett NC, Jarvis JUM. 1988. The social structure and reproductive biology of colonies of the mole-rat, *Cryptomys damarensis* (Rodentia, Bathyergidae). *J Mammal.* 69(2): 293-302.
- Bennett NC, Jarvis JUM, Wallace DB. 1990. The relative age structure and body masses of complete wild-captured colonies of two social mole-rats, the common mole-rat, *Cryptomys hottentotus hottentotus* and Damaraland mole-rat, *Cryptomys damarensis*. *J Zool.* 220: 469-485.
- Bennett NC, Faulkes CHG. 2000. African mole-rats: ecology and eusociality. UK: Cambridge University Press.

- Boesch C. (2002). Cooperative hunting roles among Tai chimpanzees. *Hum Nature*, 13(1): 27–46.
- Bowman BM, Miller SC. 1999. Skeletal mass, chemistry and growth during and after multiple reproductive cycles in the rat. *Bone*. 25: 553-559.
- Braude SH. 1991. Which naked mole-rats volcano? In: *The biology of the naked mole-rat*. Sherman PW, Jarvis JUM, Alexander RD. USA: Princeton University Press: 185-194.
- Braude SH. 2000. Dispersal and new colony formation in wild naked mole-rats: evidence against inbreeding as the system of mating. *Behav Ecol*. 11: 7-11.
- Brett RA. 1991. The populations structure of naked mole-rat colonies. In: *The biology of the naked mole-rat*. Sherman PW, Jarvis JUM, Alexander RD. USA: Princeton University Press: 97-136.
- Burda H. 1990. Constrains of pregnancy and evolution of sociality in mole-rats with a special reference to reproductive and social patterns in *Cryptomys hottentotus* (Bathyergidae, Rodentia). *Z zool Syst Evolut Forsch*. 28: 26-39.
- Burda H. 1999. Syndrome of eusociality in african subterranean mole-rats (Bathyergidae, Rodentia), its diagnosis and aetiology. In: *Evolutionary Theory and Processes: Modern Perspectives*. Wasser S. NLD: Springer Netherlands: 385-418.
- Cant MA, Field J. (2001). Helping effort and future fitness in cooperative animal societies. *P Roy Soc Lond B Bio* 268(1479): 1959–1964.
- Clutton-Brock TH. (2006). Cooperative breeding in mammals. In: Kappeler PM, van Schaik CP. *Cooperation in Primates and Humans*. Springer, Berlin, Heidelberg

173–190.

Clutton-Brock T. (2009). Structure and function in mammalian societies. *Philos T Roy Soc B*. 364(1533): 3229–3242.

Clutton-Brock TH, Gaynor D, Kansky R, MacColl ADC, McIlrath G, Chadwick P, Skinner JD. (1998). Costs of cooperative behaviour in suricates (*Suricata suricatta*). *P Roy Soc Lond B Bio*, 265(1392): 185–190.

Clutton-Brock TH, Maccoll A, Chadwick P, Gaynor D, Kansky R, Skinner JD. (1999). Reproduction and survival of suricates (*Suricata suricatta*) in the southern Kalahari. *Afr J Ecol*. 37(1): 69–80.

Clutton- Brock TH, Brotherton PNM, O’Riain MJ, Griffin AS, Gaynor G, Sharpe L, Kansky R, Manser MB, McIlrath GM. 2000. Individual contributions to babysitting in a cooperative mongoose, *Suricata suricatta*. *P Roy Soc Lond B Bio*. 267: 301-305.

Clutton-Brock TH, Russell AF, Sharpe LL. (2003). Meerkat helpers do not specialize in particular activities. *Anim Behav*. 66(3): 531–540.

Creel NM, Creel S. 2002. *The African wild dog: behavior, ecology and conservation*. US: Princeton University press.

Crespi BJ. 1992. Eusociality in Australian gall thrips. *Nature*. 359: 724-726.

Crespi BJ, Yanega D. 1994. The definition of eusociality. *Behav Ecol*. 6: 109-115.

Dammann P, Sell DR, Begall S, Strauch CH, Monnier VM. 2011. Advanced glycation end-products as markers of aging and longevity in the long-lived Ansell’s mole-rat (*Fukomys anselli*). *J. Gerontol. A. Biol. Sci. Med. Sci.* glr208.

- Danchin E, Giraldeau LA, Cézilly F. 2008. Behavioural ecology. UK: Oxford University Press.
- Davies NB, Krebs JR, West SA. 2012. An introduction to behavioural ecology. UK: John Wiley & Sons.
- Darwin CH. 1859. The origin of species. UK: Murray.
- Desmet N, Van Daele PAAG, Adrians D. 2012. A methodical analysis of behavioral observation in social African mole-rats (Bathyergidae, Rodentia). *Mammalia* 77.1: 59-69.
- Emmett Duffy J, Macdonald KS. (2010). Kin structure, ecology and the evolution of social organization in shrimp: A comparative analysis. *Proc R Soc London B*, 277(1681): 575–584.
- English S, Browning LE, Raihani NJ. (2015). Developmental plasticity and social specialization in cooperative societies. *Anim Behav*. 106: 37–42.
- Fritzsche P, Gattermann R. (2002). Sozialverhalten, Kommunikation und Arbeitsteilung beim Sambischen Graumull *Cryptomys anselli* In Gruppenmechanismen: 159–172.
- Gabathuler U, Bennett NC, Jarvis JUM. 1996. The social structure and dominance hierarchy of the Mashona mole-rat *Cryptomys darlingi* (Rodentia: Bathyergidae) from Zimbabwe. *J Zool*. 240: 221-231.
- Gadagkar R. 1994. Why the definition of eusociality is not helpful to understand its evolution and what should we do about it. *Oikos* 70: 485-488.
- Gaylard A, Harrison Y, Bennett NC. 1998. Temporal changes in the social structure of a captive colony of the Damaraland mole-rat, *Cryptomys damarensis*: the

- relationship of sex and age to dominance and burrow-maintenance activity. *J Zool.* 244: 313-321.
- Gazda SK, Connor RC, Edgar RK, Cox F. (2005). A division of labour with role specialization in group-hunting bottlenose dolphins (*Tursiops truncatus*) of Cedar Key, Florida. *P Roy Soc Lond B Bio*, 272(1559): 135–140.
- Grzimek B. (2004). *Grzimek's animal life encyclopedia*. MI: Farmington Hills.
- Hamilton WD. 1964. The genetical evolution of social behaviour. II. *J. Theoret Biol.* 7: 17-52.
- Hazell RWA, Bennett NC, Jarvis JUM, Griffin M. 2000. Adult dispersal in the cooperatively breeding Damaraland mole-rat (*Cryptomys damarensis*): a case study from the Waterberg region of Namibia. *J Zool.* 252:19-25.
- Henry EC, Dengler-Crish CHM, Catania KC. 2007. Growing out of a caste – reproduction and the making of the queen mole-rat. *J Exp Biol.* 210:261-268.
- Hölldobler B, Wilson EO. 1990. *The ants*. USA: Harvard University Press.
- Jacobs DS, Bennett NC, Jarvis JUM, Crowe TM. 1991. The colony structure and dominance hierarchy of the Damaraland mole-rat *Cryptomys damarensis* (Rodentia: Bathyergidae), from Namibia. *J Zool.* 224: 553-576.
- Jarvis JUM. 1981. Eusociality in a mammal: cooperative breeding in naked mole-rat colonies. *Science.* 212: 571-573.
- Jarvis JUM, O’Riain MJ, McDaid E. 1991. Growth and factors affecting body size in naked mole rats. In: *The biology of the naked mole-rat*. Sherman PW, Jarvis JUM, Alexander RD. USA: Princeton University Press: 358-383.
- Jarvis JUM, Sherman PW. 2002. *Heterocephalus glaber*. *Mamm Spec.* 706: 1-9.

- Johnson BR. (2008). Within-nest temporal polyethism in the honey bee. *Behav Ecol Sociobiol.* 62(5): 777–784.
- Kent DS, Simpson JA. 1992. Eusociality in the beetle *Austroplatypus incompertus* (Coleoptera: Curculionidae). *Naturwissenschaften.* 79: 86-87.
- Korb J, Hartfelder K. (2008). Life history and development - a framework for understanding developmental plasticity in lower termites. *Biol Rev*, 83(3): 295–313.
- Krebs JR, Davies NB. 1997. *Behavioural ecology: an evolutionary approach.* UK: Blackwell Publishing.
- Kutsukake N, Inada M, Sakamoto SH, Okanoya K. (2019). Behavioural interference among eusocial naked mole rats during work. *J Ethol.* 37(1): 101–109.
- Lacey EA, Sherman PW. 1991. Social organization of naked mole-rat colonies: Evidence for divisions of labor. In: *The biology of the naked mole-rat.* Sherman PW, Jarvis JUM, Alexander RD. USA: Princeton University Press: 275-336.
- Lovegrove BG. 1988. Colony size and structure, activity patterns and foraging behaviour of a colony of the social mole-rat *Cryptomys damarensis* (Bathyergidae). *J Zool.* 216: 319-402.
- Lövy M, Šklíba J, Šumbera R. 2013. Spatial and temporal activity patterns of the free-living giant mole-rat (*Fukomys mechowii*), the largest social Bathyergid. *Plos one.* 8(1): 1-9.
- Lukas D, Clutton-Brock T. (2012). Cooperative breeding and monogamy in mammalian societies. *P Roy Soc Lond B Bio.* 279(1736): 2151–2156.

- Mitchener CD. 1974. Comparative social behavior of bees. *Annu Rev Entomol.* 14:299-342.
- Moehlman P. (1987). Social Organization in Jackals: The complex social system of jackals allows the successful rearing of very dependent young. *Am Sci.* 75(4): 366-375.
- Moolman M, Bennett NC, Schoeman AS. 1998. The social structure and dominance hierarchy of the highveld mole-rat *Cryptomys hottentotus pretoriae* (Rodentia: Bathyergidae). *J Zool.* 246: 193-201.
- Mooney SJ, Filice DCS, Douglas NR, Holmes MM. 2015. Task specialization and task switching in eusocial mammals. *Anim Behav.* 109: 227-233.
- Nunes S, Duniec TR, Schweppe SA, Holekamp KE. (1999). Energetic and endocrine mediation of natal dispersal behavior in Belding's ground squirrels. *Horm Behav.* 35(2), 113–124.
- O'Riain MJ, Jarvis JUM, Faulkes CHG. 1996. A dispersive morph in the naked mole-rat. *Nature.* 380: 619-621.
- O'Riain MJ, Jarvis JUM. 1998. The dynamics of growth in naked mole-rats: the effects of litter order and changes in social structure. *J Zool.* 246: 49-60.
- O'Riain MJ, Jarvis JUM, Alexander R, Buffenstein R, Peeters C. (2000). Morphological castes in a vertebrate. *Pnas*, 97(24): 13194–13197.
- Patzenhauerová H, Šklíba J, Bryja J, Šumbera R. 2013. Parentage analysis of Ansell's mole-rat family groups indicates a high reproductive skew despite relatively relaxed ecological constraints on dispersal. *Mol Ecol.* 22: 4988-5000.
- Pepper JW, Braude SH, Lacey EA, Sherman PW. 1991. Vocalizations of the naked mole-rat. In: *The biology of the naked mole-rat.* Sherman PW, Jarvis JUM, Alexander RD. USA: Princeton University Press: 243-274.

- Rood JP. 1990. Group size, survival, reproduction and routes to breeding in dwarf mongooses. *Anim Behav.* 39(3): 566-572.
- Russell AF, Sharpe LL, Brotherton PN M, Clutton-Brock TH. (2003). Cost minimization by helpers in cooperative vertebrates. *P Natl A Sci*, 100(6): 3333–3338.
- Scantlebury M, Speakman JR, Oosthuizen MK, Roper TJ, Bennett NC. 2006. Energetics reveals physiologically distinct castes in eusocial mammal. *Nature.* 440: 795-797.
- Schielke CHKM, Begall S, Burda H. 2012. Reproductive state does not influence activity budgets of eusocial Ansell's mole-rats, *Fukomys anelli* (Rodentia, Bathyergidae): A study of locomotor activity by means of RFID. *Z Saugetierkd.* 77: 1-5.
- Schmidt CM, Jarvis JUM, Bennett NC, Schmidt CM, Jarvis JUM. (2016). The long-lived queen : reproduction and longevity in female eusocial Damaraland mole-rats The long-lived queen : reproduction and longevity. *Afr Zool* 48(1):193-196.
- Sherman PW, Lacey EA, Reeve HK, Keller L. 1995. The eusociality continuum. *Behav Ecol.* 6: 102-108.
- Šklíba J, Lövy M, Burda H, Šumbera R. (2016). Variability of space-use patterns in a free living eusocial rodent, Ansell's mole-rat indicates age-based rather than caste polyethism. *Sci Rep UK*, 6(12): 1–9.
- Stallcup JA, Woolfenden GE. (1978). Family status and contributions to breeding by Florida scrub jays. *Anim Behav.* 26:1144–1156.
- Stander PE. (1992). Cooperative hunting in lions: the role of the individual. *Behav Ecol Sociobiol*, 29: 445-454.

- Thorley J, Mendonça R, Vullioud P, Torrents-Ticó M, Zöttl M, Gaynor D, Clutton-Brock T. (2018). No task specialization among helpers in Damaraland mole-rats. *Anim Behav.* 143: 9–24.
- Tofts CH, Franks NR. 1992. Doing the right thing: Ants, Honeybees and naked mole-rats. *Tree.* 7: 346-349.
- Vajda EG, Bowman BM, Miller SC. (2001) Cancellous and cortical bone mechanical properties and tissue dynamics during pregnancy, lactation and poslactation in the rat. *Biol Reprod.* 65(3): 689-695.
- Wallace ED, Bennett NC. 1998. The colony structure and social organization of the giant Zambian mole-rat, *Cryptomys mehowi*. *J Zool.* 244: 51-61.
- Wilson EO. (1963). The Social Biology of Ants. *Annu Rev Entomolo*, 8(1): 345–368.
- Wilson EO. 1971. *The Insect Societies*. US: Harvard University Press.
- Yeboah S, Dakwa KB. 2002. Colony and social structure of the Ghana mole-rat (*Cryptomys zechi*, Matchie) (Rodentia: Bathyergidae). *J Zool.* 256: 85-91.
- Young AJ, Bennett NC. 2010. Morphological divergence of breeders and helpers in wild Damaraland mole-rat societies. *Evolution.* 64(11): 3190-3197.
- Zöttl M, Vullioud P, Goddard K, Torrents-Ticó M, Gaynor D, Bennett NC, Clutton-Brock T. (2018). Allo-parental care in Damaraland mole-rats is female biased and age dependent, though independent of testosterone levels. *Physiol Behav.* 193(5): 149–153.