

CHAPTER 6

**Do non-reproductive female giant mole-rats,
Fukomys mechowii removed from the confines of
their natal colony exhibit induced or spontaneous
ovulation?**

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Abstract

The giant mole-rat is a social subterranean rodent that exhibits aseasonal breeding. Non-reproductive females do not show physiological suppression of reproduction while in the confines of the natal colony. The study aimed to investigate if non-reproductive female giant mole-rats exhibit induced or spontaneous ovulation. Six non-reproductive females were removed from their natal colonies and housed individually without a male for a period of 18 weeks of which the first 6 weeks were for acclimatisation and the subsequent 12 weeks for a control experiment 1. They were then later housed for a further 7.3 weeks for a chemical contact experiment 2, which had females on their own for the first 3.6 weeks before being allowed to non-physical contact with a mature adult male for a further 3.7 weeks. The non-reproductive females were given a further period of isolation for two and half months prior to being paired physically with vasectomized males for 6 weeks, as experiment 3 which inclusively covered 12 weeks together with the earlier 6 weeks of control period. Urine was collected every second day for all three experiments and urine profiles generated. Progesterone values were markedly higher during the second part of Experiment 1 (4.43 ± 2.9 ng /mg Cr, $n=6$) compared to the first part (1.43 ± 0.5 ng Progesterone/mg creatinine, $n=6$). Similarly, the progesterone values measured during the first part of Experiment 2 and 3 tended to be higher ($Z= -2.201$, $p=0.028$ for both comparisons) than those measured during the first 83 days of Experiment 1. However, this was not significant after Bonferroni correction. The progesterone values were also elevated during the second phase of Experiments 2 and 3 but not significantly so (Experiment 1 vs. 2: $Z=-1.782$, $p=0.075$, Experiment 1 vs. 3: $Z=-2.201$, $p=0.028$). Pairwise comparisons of progesterone concentrations between the control and experimental manipulation for 3 experiments, using

Statistica version 8.0, revealed non significant results between Experiments 1 ($P = 0.8707$, $F = 0.348736$) and 2 ($P = 0.361606$, $F = 1.25244$) but experiment 3 was significant at $P = 0.00238$, $F = 9.2374$ (Fig 6.4). However, the varying differences noticed in the progesterone concentration of experiments 1-3, for the control period were not significant i.e. experiment 1, $P = 0.939319$, with $F = 0.205787$, experiment 2, $P = 0.588757$, with $F = 0.86161$ and experiment 3, $P = 0.393088$, with $F = 1.4964$ (Fig 6.4). Thus, chemical or physical stimulation by a male does not appear to be necessary for ovulation in female giant mole-rats but concentrations are enhanced when a non-breeding female is in chemical or physical contact with a vasectomised male. In captivity, the first rise in the urinary progesterone concentrations of the non-breeding females was only observed after 79 days. These results imply that although the giant mole-rat *F. mechowii* is a spontaneous ovulator and that recrudescence of ovulation requires a period of time to occur.

Introduction

In female eutherian mammals ovulation may take place by one of two means, namely spontaneous or induced ovulation Milligan, (1972). Females exhibiting induced ovulation have spontaneous development of the primordial follicles to the Graafian follicle stage, but without copulation and the subsequent vaginal and cervical stimulation that is required the female fails to ovulate. In contrast, females, especially of aseasonal breeders undergoing spontaneously ovulation characteristically possess continuous cycling of the reproductive hormones and subsequent ovulation and the production of corpora lutea of ovulation without the physical and or chemical contact of the male. In induced ovulation the cycling of reproductive hormones and subsequent ovulation only occurs in the physical presence of a

male (vaginal stimulation, tactile or olfactory). In induced ovulation the physical presence always involves coitus.

In subterranean animals the cost of finding a mating partner is largely increased during the dry periods compared to those for species that live above ground due to high energetic costs of digging Vleck, (1979). Rainfall can affect soil hardness and thus the costs of mate searching may be reduced during high rainfall periods as members of the colony are more relaxed due to easier foraging. This may select for induced ovulation in subterranean species and indeed in a number of solitary subterranean rodents exhibiting seasonal reproduction (Bennett & Jarvis 1988a; Herbst *et al.*, 2004; Hart *et al.*, 2006) and the optimal strategy in choice is induced ovulation (van Sandwyk & Bennett 2005). In solitary members of the subterranean Southern African mole-rats (Bathyergidae-*Bathyergus suillus* and *Georchus capensis*) which are seasonal breeders and induced ovulators, elaborate penile structures further facilitate this mode of ovulation Parag *et al.*, (2006).

Apart from some solitary species, the family Bathyergidae contains some representatives that occur socially with a distinct reproductive skew, with reproduction partitioned to a single female and a number of putative breeding males (Bennett & Jarvis, 1988b; Bennett, 1989; Burda, 1989; Bennett *et al.*, 1994; Bennett & Aguilar, 1995). Some of these social bathyergids occur in mesic habitats with high seasonal rainfall that is likely to facilitate frequent dispersal of non-breeders. Females of these species remain anovulatory whilst in the confines of the natal colony but after dispersal and subsequently pairing with unrelated males they exhibit induced ovulation (Malherbe *et al.*, 2004a; Jackson & Bennett, 2005)

facilitating colony foundation by dispersing individuals. This typical induced ovulation is complemented by males with numerous epidermal spines on the penis Parag *et al.*, (2006).

In contrast, the two eusocial species of mole-rat, the Damaraland mole-rat, *Fukomys damarensis* and the naked mole-rat, *Heterocephalus glaber* experience strong ecological constraints in their arid habitats that provide non-breeders with little or no opportunity to disperse Jarvis *et al.*, (1994). In the habitats occupied by these mole-rats, rainfall is unpredictable and sporadic, and years may pass before soil properties are ideal for dispersal. The non-reproductive females of the naked and Damaraland mole-rats are physiologically suppressed whilst in the confines of the colony. In the presence of the reproductive female, the concentrations of luteinizing hormone are low and the rest of females remain anovulatory (Bennett *et al.*, 1993; Molteno & Bennett, 2002). Removal of the queen from the colony removes the suppression of reproduction in the Damaraland mole-rat yet the non-reproductive females will only reproduce if unrelated males reside in the colony (Rickard & Bennett, 1997). In contrast, queen succession from within can occur in the naked mole-rat, however, as in the Damaraland mole-rat non-breeding females are physiologically suppressed in the presence of a female breeder Faulkes *et al.*, (1990). When removed from the influence of the breeding female non-reproductive females of both species are relieved of suppression and subsequently pair with genetically unrelated males to form new colonies. However, in captivity the appearance of first elevation of progesterone for the non-breeding females is dependent on the species with others producing elevations within 7 days Faulkes *et al.*, (1990). Males of both species lack the elaborate ornamentation of the penis found in the other Bathyergidae Parag *et al.*, (2006) and female Damaraland mole-rats exhibit

spontaneous ovulation Snyman *et al.*, (2006). This mode of ovulation may have arisen or persisted in these two species because colony turnover is less frequent compared to the species in mesic habitats. As a consequence of extreme natal philopatry and the more prolonged mating opportunities, there may be no selective advantage to induced ovulation.

Recent molecular phylogenies place another member of the Bathyergidae, the giant mole-rat (*Fukomys mechowii*) in close relationship with the Damaraland mole-rat (Faulkes *et al.*, 2004; Ingram *et al.*, 2004). Like the Damaraland mole-rat they breed throughout the year Scharff *et al.*, (2001), however, they occur in areas with higher rainfall and thus colony dispersal opportunities may arise frequently (Bennett & Faulkes 2000). Interestingly, in the giant mole-rat, non-reproductive female members of the colony exhibit similar circulating basal concentrations of LH as the reproductive female as well as a similar response to an exogenous GnRH challenge of LH release from the pituitary to the reproductive female Bennett *et al.*, (2000). Thus in the giant mole-rat reproductive inhibition within colonies appears to be due to incest taboos. Phylogeny would suggest that the eusocial giant mole-rat *F. mechowii* and *F. damarensis* are closely related species and are both spontaneous ovulators. This relationship between the two species further elaborates that although there was geographical structuring of haplotypes in both species, there was no apparent clinal pattern to their distribution, possibly as a result of weak bootstrap support nodes within the species clade and low levels of sequence divergence Faulkes *et al.*, (2004). In such a case the penis of the male should also lack elaborate ornamentation such as small spines or protrusions. In contrast, rainfall patterns in their habitat would point towards a mode of induced ovulation and penile ornamentation. We evaluate these possibilities in the present

study that phylogeny rather than habitat characteristics alone would determine the mode of ovulation in females and penile morphology in males.

Materials and methods

Six non-reproductive females of giant mole-rat were captured from 6 different colonies in Kakalo and Mushishima Farm blocks in Chingola, Copperbelt Province, Zambia. Twelve males were also captured in the same Farm blocks but from 6 colonies located 1000m distant from where the females were collected to ensure that they were not closely related. Animals were shipped to Pretoria and experimental details are chronologically explained in Chapter 2.

Results

Experiment 1 showed the increased urinary progesterone concentrations which commenced after 37 days in captivity and during the last 10 days of an experiment. The highest progesterone concentration (97.7nmolsP/mmols creatinine) was recorded in animal No. 4 on the 49th day of the experiment while animal No. 6 had the lowest concentration (7.2 nmolsP/mmols creatinine) amongst all non-breeding females on the same 49th day of experimental period (Fig. 6.1). Experiment 2 steadily continued with progesterone concentrations in both control and chemical periods with 32.6 and 36.1nmolsP/mmols creatinine recorded highest on 15th and 35th days for control and chemical periods for animal Nos 6 and 2, respectively. Unlike in the first experiment, animal No. 4 was recorded with the lowest progesterone concentration on the 49th day (11.3 nmolsP/mmols creatinine)(Fig. 6.2). In experiment 3, the control period was recorded with alternating progesterone

concentrations per animal which ranged from 15 to 30.3 nmolsP/mmol creatinine. In the second part of experiment 3, females were physically brought into contact with a vasectomised male and mating took place within the first couple of days. In this part of experiment, the range of progesterone was observed highest from 29.9 to 152.7 nmolsP/mmol creatinine suggesting that ovulation appears to be enhanced in non-reproductive females by the presence of a non-related breeding male (Fig. 6.3).

Baseline period

Progesterone values were markedly higher during the second part of Experiment 1 (4.43 ± 2.9 ng /mg Cr, n=6) compared to the first part (1.43 ± 0.5 ng Progesterone/mg creatinine, n=6) (Fig. 6.1). Similarly, the progesterone values measured during the first part of Experiment 2 and 3 tended to be higher ($Z = -2.201$, $p = 0.028$ for both comparisons) than those measured during the first 83 days of Experiment 1. However, this was not significant after Bonferroni correction. The progesterone values were also elevated during the second phase of Experiments 2 and 3 but not significantly so (Experiment 1 vs. 2: $Z = -1.782$, $p = 0.075$, Experiment 1 vs. 3: $Z = -2.201$, $p = 0.028$). Thus, chemical or physical stimulation by a male does not appear to be necessary for ovulation in female giant mole-rats (Fig. 6.1).

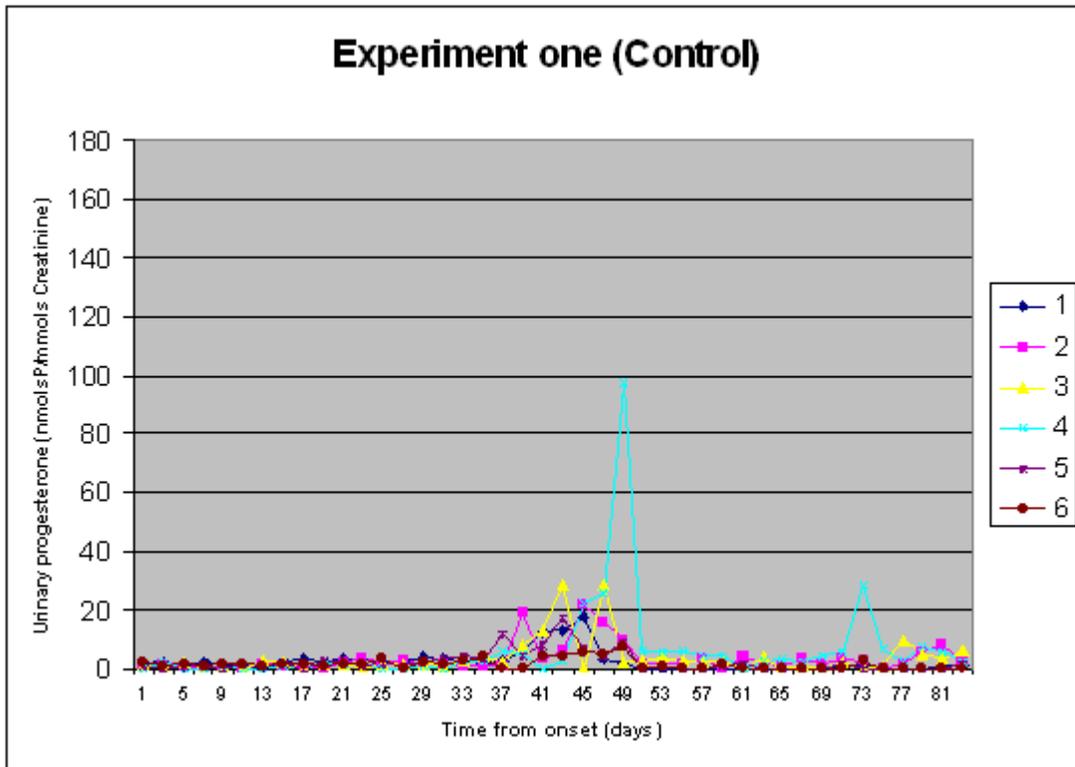


Figure 6.1. Mean urinary progesterone profile (nmolsP/mmols creatinine) before experimental manipulation of female (*F. mechowii*) housed singly without any contact with a male.

Progesterone values tended to be higher during the second part of Experiment 1 (4.43 ± 2.9 ng /mg Cr, n=6) compared to the first part (1.43 ± 0.5 ng Progesterone/mg creatinine, n=6), however, this was not significant (Wilcoxon, $Z=-2.201$, $p=0.075$). Similarly, results of progesterone values measured in the first part for Experiment 2 were not significant ($Z= -0.943$, $p>0.05$; 9.57 ± 2.6 ng Progesterone /mg Creatinine with n=6) compared to the second part of Experiment: (8.45 ± 2.2 ng Progesterone/ mg creatinine) (Fig. 6.2).

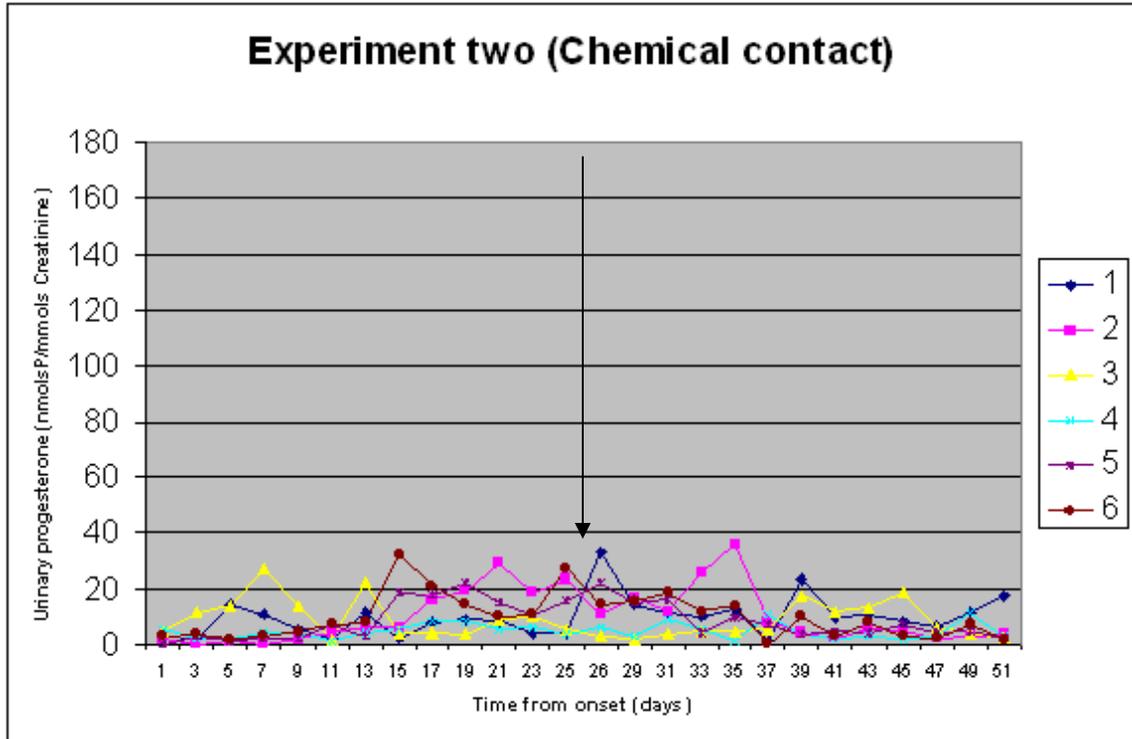


Figure 6.2. Mean urinary progesterone profile (nmolsP/mmols creatinine) after experimental manipulation of female (*F. mechowii*) housed in a chemical contact with a male. The middle arrow indicates the first part of experiment to the left and second part of experiment to the right.

Progesterone values measured in the first part for Experiment 3 were also not significant ($Z = -1.572$, $p > 0.05$; 6.61 ± 1.2 ng Progesterone /mg creatinine with $n = 6$ compared to the second part of Experiment: 14.59 ± 8.4 ng Progesterone /mg creatinine) (Fig. 6.3).

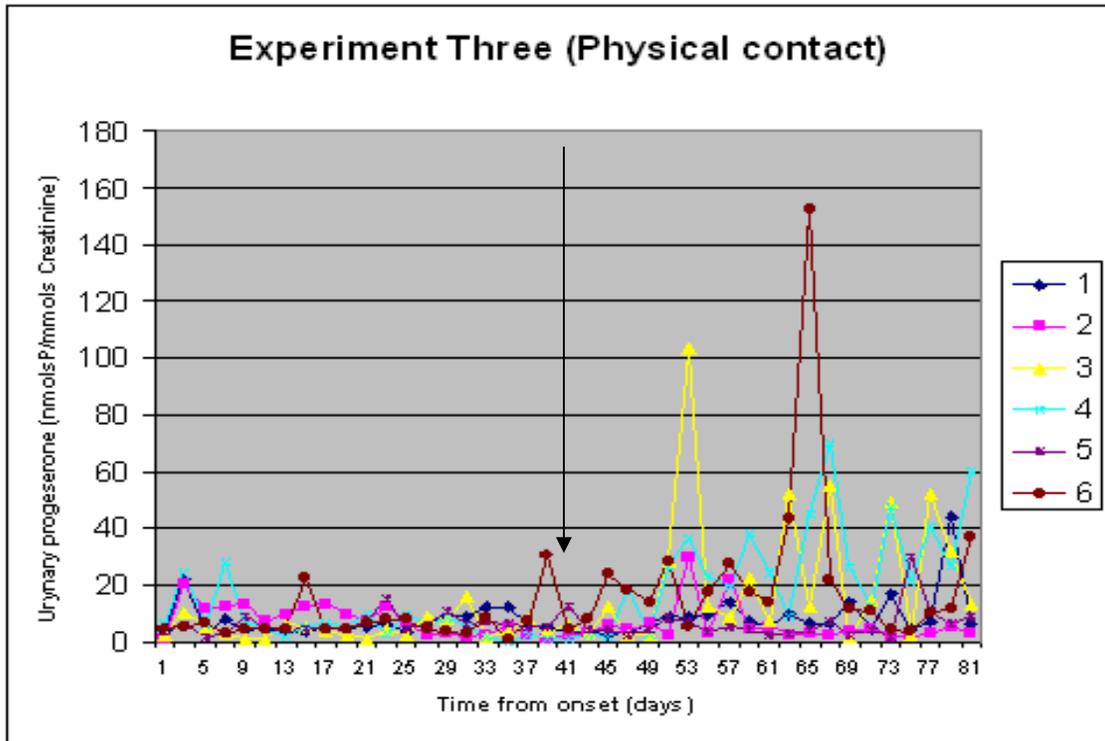


Figure 6.3. Mean urinary progesterone profile (nmolsP/mmols creatinine) before and after experimental manipulation for female (*F. mechowii*) housed singly, in physical contact with a vasectomised male. The middle arrow indicates the first part of experiment to the left and second part of experiment to the right.

Experimental manipulations

Pairwise comparisons of progesterone concentrations between the control and experimental manipulation for 3 experiments revealed non significant results between Experiments 1 ($P = 0.8707$, $F = 0.348736$) and 2 ($P = 0.361606$, $F = 1.25244$) but experiment 3 was significant at $P = 0.00238$, $F = 9.2374$ (Fig 6.4). However, the varying differences noticed in the progesterone concentration of experiments 1-3, for the control period were not significant

i.e. experiment 1, $P = 0.939319$, with $F = 0.205787$, experiment 2, $P = 0.588757$, with $F = 0.86161$ and experiment 3, $P = 0.393088$, with $F = 1.4964$ (Fig 6.4).

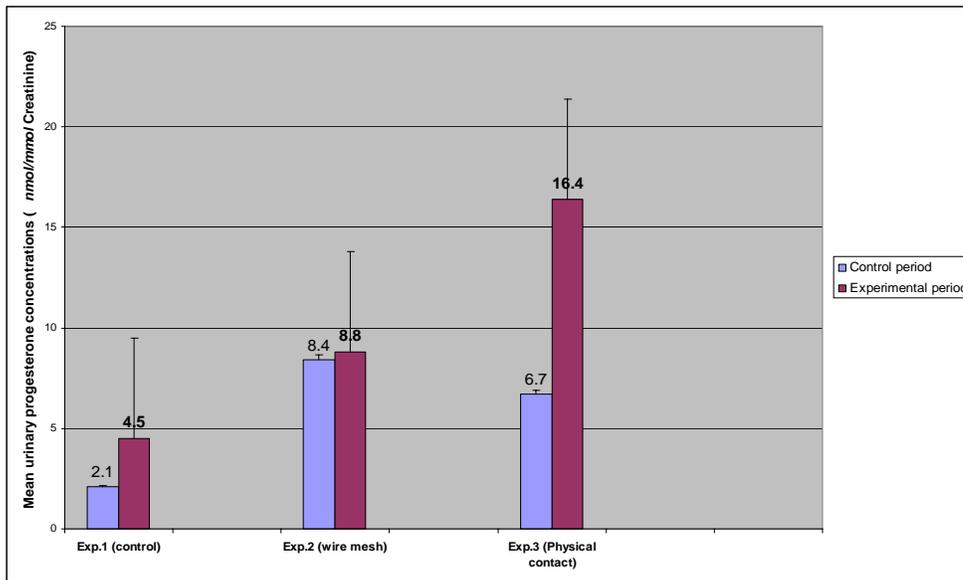


Figure 6.4. Mean urinary progesterone concentrations (nmolsP/mmol/creatinine) before and after experimental manipulation for female (*F. mechowii*) housed singly, in a chemical contact with a male and physical contact with a vasectomised male.

Penile structure of the giant mole-rat

Electrone micrograph of the penis of the giant mole-rat, revealed it to possess a number of slightly raised longitudinal ridges with a distinct lack of obvious spines or rounded protrusions as similar to that of *Fukomys damarensis* and the naked mole-rat (*Hetercephalus glaber*) which also exhibit spontaneous ovulators Parag *et al.*, (2006) (Fig. 6.5 a, b).

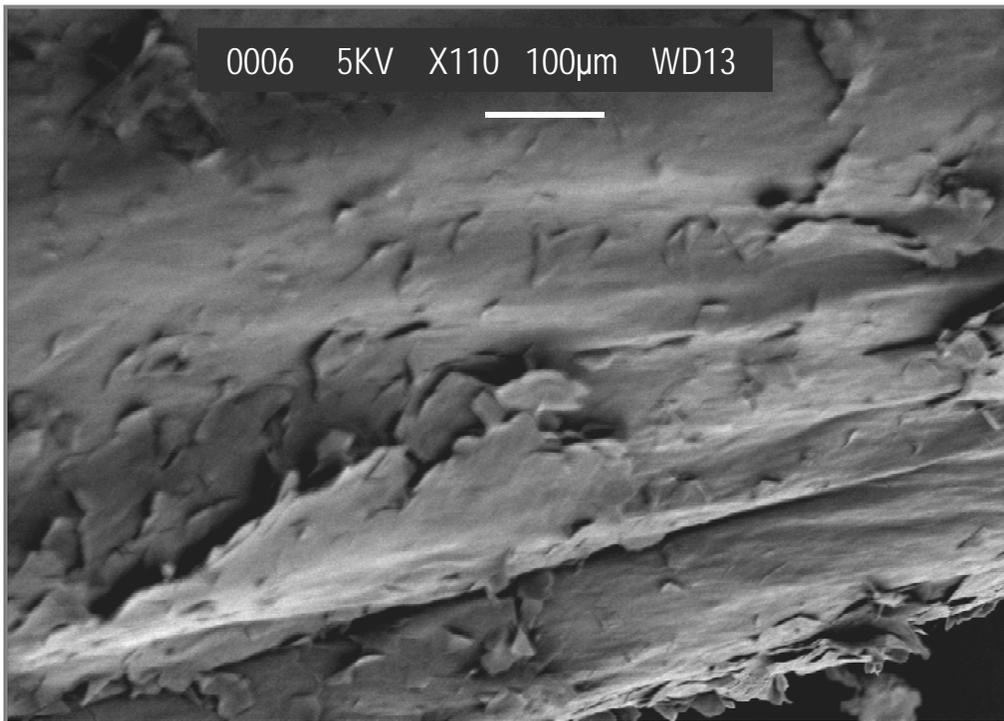


Figure. 6.5a. Electrone micrograph of penis of male *F. mechowii* showing smooth glans and shallow longitudinal ridges on the surface of penis.



Figure. 6.5b. Electron micrograph of penis of male *F. mechowii* showing smooth glans and shaft with shallow longitudinal ridges.

All results in this study have indicated that the giant mole-rat can be considered a spontaneous ovulator since in all control portions of experiment 1, 2 and 3 as well as the experimental section of experiment 1, there are cycles of progesterone observed in non-reproductive females which are housed singly. When non-reproductive females are housed with or placed in the presence of a vasectomised but not allowed physical contact or physically in contact, the intensity of the progesterone concentrations are enhanced showing that males do have some component. The spontaneous ovulation of the giant mole-rat is also confirmed with the lack of penile ornamentation on the males as one of the hypothesis in the social mole-rats.

Discussion

African mole-rats are the only truly subterranean rodent group that exhibits a wide range of sociality and reproductive tactics (Jarvis & Bennett, 1990). Currently, most other families of subterranean rodents have representatives that are entirely solitary and in the majority of cases the females exhibit induced ovulation (Zarrow & Clark, 1968). Solitary subterranean species rely heavily on brief periods of interaction with opposite sexed conspecifics. In the solitary Ctenomyidae, the tuco tuco, *Ctenomys talarum* ovulation is found to be induced Weir, (1974) as is predicted by the hypothesis that solitary species of fossorial rodents are induced ovulators, whereas the solitary Mediterranean mole-rat Spalacidae, *Spalax ehrenbergi* exhibits spontaneous ovulation Shanas *et al.*, (1995). Our hypothesis distinctly elaborates that induced ovulation requires the mechanical stimulation of the cervix and vaginal walls to initiate follicular development and subsequent ovulation Milligan, (1972). Within the family Bathyergidae, the solitary Cape mole-rat, *Georychus capensis* is an

induced ovulator (van Sandwyk & Bennett, 2005). Examination of the penis of the highveld mole-rat, *Cryptomys hottentotus pretoriae* has shown that the glans and the shaft of the penis to be covered with small spinose protrusions. It is thus surprising that within the two species of social African mole-rat, *Cryptomys hottentotus natalensis* and the high veld mole-rat *Cryptomys hottentotus pretoriae*, exhibit seasonal breeding yet their ovulation pattern is induced (Malherbe *et al.*, 2004b; Jackson & Bennett, 2005). Studies on the highveld mole-rat, *Cryptomys h. pretoriae* and the Natal mole-rat, *Cryptomys natalensis* have shown that the males of these species have small protrusions emanating from the glans and shaft of the penis Parag *et al.*, (2006).

In contrast, the two eusocial species of mole-rat, the naked mole-rat, *Heterocephalus glaber* and the Damaraland mole-rat, *Fukomys damarensis* are both reportedly spontaneous ovulators (Faulkes *et al.*, 1990; Snyman *et al.*, 2006). These two species are found in arid habitats where rainfall is sporadic and unpredictable, furthermore rains may fail in some years and hence dispersal is impossible due to unfavourable soil conditions for burrowing. Spontaneous ovulation may have arisen or persisted in these two species because colony turnover is less frequent. As a consequence of extreme natal philopatry and more prolonged mating opportunities, there may be no selective advantage to induced ovulation and hence elaborate ornamentation to the penis in these two species Parag *et al.*, (2006). We predicted that based on the sociality and phylogenetic relationship between the *F. mechowii* and *F. damarensis* where both species were found with geographical structuring of haplotypes and low levels of sequence divergence within the species clade Faulkes *et al.*, (2004), the giant mole-rat should also be a spontaneous ovulator. This is borne out in the endocrine profiles

where in the control phase of the study there are raised concentrations of progesterone indicative of an ovulation event in non-reproductive females removed from the colony and isolated on their own. The longitudinally ribbed characteristic of the penis and shaft further reflects the lack of mechanical stimulation required in a species where follicular development and subsequent ovulation is spontaneous and does not require the mechanical stimulation of the cervix and vagina. In solitary and social species exhibiting induced ovulation, the male is characterized by possessing spines or blunt spinose ornamentation along the glans and shaft of the penis Parag *et al.*, (2006). However, as in the Damaraland mole-rat and naked mole-rats (Faulkes *et al.*, 1990; Snyman *et al.*, 2006), the progesterone concentrations of the giant mole-rat acquires elevated progesterone concentrations only when females are in direct physical contact with males. Ovulation also appears to be enhanced in presence of non-related male (Fig. 6.3.)

The release from the inhibition of ovarian cyclicity in the non-breeding females of giant mole-rat is longer than in other social mole-rat species. The urinary progesterone concentrations of the naked mole-rats in captivity showed ovarian cyclicity within 7 days Faulkes *et al.*, (1990) while the first elevation of progesterone for the giant mole-rats were only observed on the 79th day in captivity following removal from the colony. Nevertheless, the giant mole-rat is a spontaneous ovulator. Both the hormone profiles (Fig. 6.2 and 6.3) and the findings from the structure of the penis of the male support the general reasoning that ovulation is a spontaneous affair in the giant mole-rat.

In species that are obligate outbreeders such as the giant mole-rat there is incest avoidance between father and daughters, as well as between brothers and sisters, but the incest avoidance between mother and sons seems to be weaker. Experiments and observations to date (Burda 1995; H. Burda unpublished) suggest that behavioural control (mate guarding) by the dominant male (the father) may also play an important role in inhibition of male reproduction. This fact may have represented a selective pressure leading to significant increase in the body mass (and sexual dimorphism) of breeding males in this species.

Bennett *et al.*, (2000) demonstrated that non-reproductive female giant mole-rats are not physiologically suppressed at the level of the pituitary and furthermore that the response of the pituitary to an exogenous overdose of GnRH is similar to that exhibited in reproductive female giant mole-rats. All of this evidence points to the fact that non-reproductive female giant mole-rats are capable of reproduction but do not because of strict incest taboos.

CHAPTER 7

Synthesis

The giant mole-rat

Considerable efforts have been made in the studies of reproductive biology, burrow system and colony size of the giant Zambian mole-rat (Bennett & Aguilar, 1995; Scharff *et al.*, 2001; Burda & Kawalika 1993). Other separate studies include the pituitary sensitivity to exogenous GnRH Bennett *et al.*, (2000). Until recently however, the existing published information on the reproduction of the giant mole-rat was derived from laboratory studies (Bennett & Aguilar, 1995; Scharff *et al.*, 1999) with a small field study Scharff *et al.*, (2001) in which aseasonal reproduction was suggested to take place in the giant mole-rat. Other anecdotal reports were from Ansell (1978) who captured young animals throughout the year. Mostly, the speculation on the larger group sizes of captured animals which varied from 40+ animals Scharff *et al.*, (2001) to 60+ animals (Burda & Kawalika, 1993), either involved two or more colonies or was gleaned from local hunters. With the low sample sizes, colonies involved and the period of sample collection in these studies, reliable and conclusive results could not be attained.

This study is the most extensive to date and is mainly focused on the description of the general burrow architecture of the giant mole-rat from 32 burrow systems, excavated in their entirety, throughout the year. It was imperative to employ the burrow fractal dimensions analysis on the burrows of the giant mole-rat, in order to compare its effects on the burrow structure, length and its relationship to foraging success Le Comber *et al.*, (2006) in both rainy and dry seasons (Fig. 3.1). Furthermore, the fractal dimension provides a good indication of the extent to which a burrow explores the surrounding area and also offers a more useful burrow metric than burrow length Le Comber *et al.*, (2006). It was also

predicted that formidable indicators emanating from the burrow fractal dimensions can explain and relate the importance of the Aridity Food Distribution Hypothesis (AFDH) (Jarvis *et al.*, 1994; 1998), in the giant mole-rat.

The fractal analysis component was then employed on the mapped burrow systems to answer three general questions relating specifically to the assumptions of the AFDH: (1) Is burrow fractal dimension higher in the rainy season, as might be predicted if either or both the energetic costs of digging, or differences in patterns of food distribution, vary between seasons? (2) Do larger colonies have burrows with higher fractal dimensions? This might be the case if, as the AFDH suggests, foraging is more efficient in larger, cooperatively foraging colonies. (3) Is higher fractal dimension associated with a greater mass of food in the burrow? (Le Comber *et al.*, 2006) showed, using computer simulations, that burrows with high fractal dimension located more food; here, this study tests whether this is reflected in larger food stores in a natural situation.

Furthermore, there was need in this study to address the uncertainties involved in colony composition of the colonies of giant mole-rats captured. The cranial morphometric analyses and the structure of age classes, based on tooth-wear and eruption patterns were carried out to ascertain whether (i) the species is sexually dimorphic and (ii) if sexual dimorphism is evident does it arise early in the life history or following maturation and the attainment of adulthood. There was also need in this study to investigate whether the giant mole-rat is an aseasonal or seasonal breeder. The final component of the study investigated if ovulation in

non-reproductive females removed from the confines of the colony is of a spontaneous or induced nature.

Burrow Systems

The general burrow architecture of 32 burrow systems was described, including the burrow structure, length and diameter of tunnels, location and depths of nests, food stores and toilets. Foods found in food stores, including stored tuber crops and geophytes were identified and weighed for each colony.

The length of burrow system did not differ between seasons but increased with the number of animals in the colony. The range of burrow lengths of the giant mole-rat fell within the category of solitary species *Barthyergus suillus* (Davies & Jarvis, 1986), *Bathyergus janetta* (Herbst & Bennett, 2006) and *H. argenteocinereus* (Šumbera *et al.*, 2003c). The food store had a higher mass during the rainy season than in dry season (Fig. 3.2).

Burrow fractal dimensions

A number of studies on the Bathyergidae have examined burrow architecture although not necessarily fractal dimensions (Hickman, 1977; Davies and Jarvis, 1986; Zuri & Terkel, 1996; Rosi *et al.*, 2000; Spinks *et al.*, 2000a; Šumbera *et al.* 2003c and Herbst & Bennett, 2006) but the majority of these have concentrated on solitary species that only have plural occupancy during the breeding season or when mother has young. There have been relatively few studies reporting the burrow fractal dimensions and architecture of social dwelling mole-rats (Spinks *et al.*, 2000a) but usually, the sample sizes were very small.

Fractal dimensions increased with both the length of the burrow and the number of animals, but especially adults in the colony and was higher during the rainy season than the dry. Similarly, the mass of food in the burrow also tended to increase with fractal dimensions and was higher during the rainy season than in the dry season (Fig. 3.2). An interesting point to note was that while fractal dimensions were greater in rainy season, there was no overall difference in total burrow length. This might be due to the point that burrows in rainy season had more of the short side tunnels than those in dry season and resulted into substantial increase in fractal dimensions in the rainy season without necessarily increasing the total burrow length (Fig. 3.1). During periods when soil was softened by rainfall, geophytes began to proliferate and mole-rats took this advantage to forage and store sufficient food to last them through the dry season. Thus, the limiting factor for burrow excavation in mole-rats mainly depends on (i) amount and periodicity of rainfall and (ii) number of animals in the burrow system available for digging. These results provide evidence in support of each of these factors.

During the rainy season, foraging was found to be more efficient and colony size increased, prompting higher fractal dimensions and more food mass in burrows. These results concur with foraging models of (Lovegrove & Wissel, 1988) and (Spinks & Planganyi (1999) which suggested that colony size is important in foraging risk, especially for mole-rat species occurring in arid environments.

These results are in line with previous data in which Jarvis *et al.*, (1998) indicated that larger colonies of social mole-rats have greater survival and make for the first time the link between colony size, burrow architecture and foraging success (Table 3.1). To summarize, these results are important because they link for the first time rainfall, colony size, burrow architecture and foraging success in a single social mole-rat species, and thus support the critical assumptions that underlies the aridity food distribution hypothesis.

Aseasonality and Colony size

Colony size and reproductive biology are important factors to African mole-rats because colony size particularly is a crucial comparative parameter and component which plays an important role in sociality and is an indirect measure of the degree of dispersal/philopatry. Until recently, previous studies have shown uncertainties on the colony size in free living colonies of giant mole-rat. The first study by Burda & Kawalika, (1993) revealed that the social group size of giant mole-rat to be over 60 animals per colony. Unfortunately, this information was sourced from local hunters without any distinct colony surveying knowledge. Another small study done by Scharff *et al.*, (2001) only compared six complete colonies, five of which ranged from 3-12 in group size, but the sixth possibly totalling 40 animals or more. However, it is speculated that the animals from the latter colony may have been caught from neighbouring burrows, as the area in question was difficult to survey. Both studies were based on small sample sizes with very little time by workers devoted to extensive field work. Information pertaining as to whether the giant mole-rat is aseasonally or seasonally breeding has also been wanting. Anecdotal reports by Ansell (1978) were merely based on capturing young animals throughout the year. A lack of information on

maximum colony size as well as data on whether free living giant mole-rats are aseasonal or seasonal breeders remained uncertain. Much of the information pertaining to the reproductive biology of giant mole-rats was from laboratory studies, which may be mostly limited to litter size and gestation periods of the species.

From the 32 carefully surveyed and excavated free living colonies, the mean colony size of the giant mole-rat is now clarified to be around 10, being closer but below that of *F. damarensis*, an extensively studied species which has the colony size of 12 (Bennett & Faulkes, 2000). Throughout the entire period of 10 months which included both rainy and dry seasons, one or occasionally two pregnant females and juveniles were recorded in the burrows of the giant mole-rat (Tables 4.1). Sex ratio in the wild is skewed towards females, with males being larger than females. This concurs with earlier reports by Scharff *et al.* (2001), from the 5 colonies where it was reported that colonies range from 3-12 animals. Results of this an extensive field data collection complements the findings of earlier laboratory studies that the giant mole-rat is without doubt an aseasonal breeder.

Relative age classes

The use of tooth-wear on its own has been shown to be a poor ageing method in mammals (Hall *et al.* 1957; Keiss, 1969; Morris, 1972 and Gilbert & Stolt, 1990). This study suggests that in the absence of data on the absolute ageing, the estimation of age based on molar eruption and tooth-wear may be an appropriate method for relative ageing in the giant mole-rat. In studies on single geographically related species where the diet is fairly uniform,

application of tooth-wear on the cusps of the molariform teeth may be used as a relative rather than absolute variable for ageing a population.

The time of tooth-wear and eruption is less variable and can serve as a valuable marker of relative age. This method can in conjunction with skull morphometrics be used to investigate potential sexual dimorphism with age in populations of mammals. Indeed, Bennett *et al.*, (1990) successfully used this method to investigate sexual dimorphism in two species of social mole-rat, they found that within colonies, it was absent in the common mole-rat, *C. h. hottentotus* but distinctly marked in the Damaraland mole-rat, *F. damarensis*. Similarly, sexual dimorphism and relative age variation in nine age classes estimated from the degree of molar tooth-row eruption and wear in the giant mole-rat were assessed using traditional cranial morphometric data and a range of univariate and multivariate analyses.

In addition, the use of body mass to estimate absolute age in mammals has also been considered to be inappropriate. However, as has been found in the solitary Cape dune mole-rat (Hart *et al.*, 2007), body mass which has previously been used to assess the extent of sexual dimorphism and age variation in other social species such as the highveld mole-rat, (*Cryptomys hottentotus pretoriae*) Janse van Rensburg *et al.*, (2004) was also used in the current study for comparative purposes.

All the analyses conducted on the giant mole-rat revealed craniometric differences between individuals of age classes 1–3 and those of age classes 5–9, with those of age class 4 being intermediate between these two age class groupings, suggesting that age class 4 is at a point

on a hypothetical growth curve where it begins to stabilize. In contrast, the analyses revealed the absence of sexual dimorphism in the younger individuals of the giant mole-rat of age classes 1–4 and its presence in older age classes 5–9 (Figs. 5.3 & 5.4), and these results were supported by the analysis of the data on body mass, which was non-significant in younger age classes 1-5 and significant in older age classes 6-9 (Fig. 5.5).

Ovulation

African mole-rats are the only truly subterranean rodent group that exhibits a wide range of sociality and reproductive tactics (Jarvis & Bennett, 1990). Currently, most other families of subterranean rodents have representatives that are entirely solitary and in the majority of cases, females undergo induced ovulation (Zarrow & Clark, 1968). To date, numerous studies have classified these species of mole-rats into (i) Solitary and seasonal species where animals live singly in burrows, except at the time of mating or when the mother has pups Hart *et al.*, (2007) and (ii) Social, seasonal or aseasonal species where the natal colony restricts reproduction to a single reproductive female and one or potential two reproductive males allow control of the recruitment of pups into the colony, with most members exhibiting socially induced-sterility and being monogamous (Bennett *et al.* 1993; Bennett, 1994; Bennett *et al.*, 1994; Bennett *et al.*, 1996; Jarvis & Bennett 1991).

The patterns of ovulation for the African mole-rats may be induced where the male counterpart shows elaborate penile ornamentation or spontaneous where a decrease of penis ornamentation results Parag *et al.*, (2006). The common underlying hypothesis distinctly elaborates that induced ovulation requires the mechanical stimulation of the cervix and

vaginal walls to initiate follicular development and subsequent ovulation Milligan, (1972) while apart from the two social and seasonal species, *Cryptomys h. natalensis* and *Cryptomys h. pretoriae* which exhibit induced ovulation (Jackson & Bennet, 2005; Malherbe *et al.*, 2004a), most studied eusocial and aseasonal species *F. damarensis* and *H. glaber* exhibit spontaneous ovulation (Faulkes *et al.*, 1990; Snyman *et al.*, 2006).

In solitary and social species exhibiting induced ovulation, the male is characterized by possessing spines or blunt spinose ornamentation along the glans and shaft of the penis Parag *et al.*, (2006). This condition is different in the eusocial species exhibiting spontaneous ovulation like the *F. damarensis* and *H. glaber* which have ridges on the shaft of penis. In this study, it was predicted that based on the sociality and phylogenetic relationship between the *F. mechowii* and *F. damarensis* where both species were found with geographical structuring of halotypes and low levels of sequence divergence within the species clade Faulkes *et al.*, (2004), the giant mole-rat should also be a spontaneous ovulator.

The males of the eusocial and aseasonal giant mole-rat had longitudinally ridged characteristic of the penis and shaft like those of *H. glaber* and *F. damarensis*, which was the first indication of the species being a spontaneous ovulator (Figs 6.5 a, b). Cyclicity in the non-breeding females of the giant mole-rat in captivity was longer than other social mole-rat species like the naked mole-rat, with the first elevation of progesterone only observed on the 79th day in captivity (Fig. 6.1). Nevertheless, the giant mole-rat is a spontaneous ovulator with both the hormone profiles (Fig. 6.2 and 6.3) and the findings

from the penis structure of the male supporting the general reasoning that ovulation is a spontaneous affair. As in the Damaraland mole-rat and naked mole-rats (Faulkes *et al.*, 1990; Snyman *et al.*, 2006), the progesterone concentrations of the giant mole-rat are elevated only when females are in direct physical contact with males. Ovulation also appears to be enhanced in the presence of a non-related male (Fig. 6.3).

Summary

In this thesis, I have demonstrated that the mean colony size of the giant mole-rat occurring in the mesic Copperbelt of Zambia is lower than that of its more arid adapted counterpart the Damaraland mole-rat. Furthermore, a detailed analysis of the burrow structures excavated throughout the year revealed that with an increase in precipitation, there is a concomitant complexity of the burrow system but the burrow system retains a mean maximal length irrespective of the season. As with other members of the genus *Fukomys* so far studied to date, breeding by the dominant female(s) occurs throughout the year, yet unlike the Damaraland mole-rat, in the giant mole-rat, there may be plural breeding in colonies. The pattern of ovulation is spontaneous as is that of the Damaraland mole-rat and may be phylogenetically constrained. Giant mole-rats exhibit sexual dimorphism that appears to become more apparent with the advancing age of the animals. There is a definite age structure in colonies with the breeding animals being the oldest. This study has furthered our understanding of this little studied central African mole-rat species.