

# Energetics reveals physiologically distinct castes in a eusocial mammal

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**Eusociality, which occurs among mammals only in two species of African mole-rat, is characterized by division of labour between morphologically distinct ‘castes’<sup>1</sup>. In Damaraland mole-rats (*Cryptomys damarensis*), colony labour is divided between ‘infrequent worker’ and ‘frequent worker’ castes<sup>2</sup>. Frequent workers are active year-round and together perform more than 95% of the total work of the colony, whereas infrequent workers typically perform less than 5% of the total work<sup>3</sup>. Anecdotal evidence suggests that infrequent workers may act as dispersers, with dispersal being limited to comparatively rare periods when the soil is softened by moisture<sup>4,5</sup>. Here we show that infrequent workers and queens increase their daily energy expenditure after rainfall whereas frequent workers do not. Infrequent workers are also fatter than frequent workers. We suggest that infrequent workers constitute a physiologically distinct dispersing caste, the members of which, instead of contributing to the work of the colony and helping the queen to reproduce, build up their own body reserves in preparation for dispersal and reproduction when environmental conditions are suitable.**

In communally breeding species, certain individuals forego reproduction and instead assist others to reproduce<sup>6,7</sup>. This phenomenon is seen at its most extreme in eusocial organisms, where reproductive ‘queens’ are responsible for producing all the offspring of a colony. Forsaking reproduction in favour of assisting the reproduction of others is the most extreme form of altruism known among non-human animals and, as such, poses a fundamental problem for the theory of evolution<sup>8,9</sup>. In most communally breeding species, the level of reproductive skew is thought to be determined by the balance between the inclusive fitness benefits arising from remaining within the natal group and the costs associated with ecological constraints on dispersal<sup>8,10</sup>. This cost–benefit scenario might lead to the evolution of two alternative strategies among helpers<sup>11,12</sup>. The first would involve remaining within the natal colony and expending resources to maximize indirect fitness gains; the second would involve minimizing the contribution to indirect fitness gains in the current colony, but maximizing direct fitness gains by dispersing and reproducing elsewhere whenever opportunities to disperse arise.

African mole-rats (Rodentia, Bathyergidae) include the only two known species of eusocial mammal: the naked mole-rat *Heterocephalus glaber* and the Damaraland mole-rat *Cryptomys damarensis*<sup>3</sup>. Because both species are entirely subterranean, dispersal can only be achieved by extending the burrow system, which is only possible when the soil has been softened by recent rainfall<sup>3</sup>. In Damaraland mole-rats, labour is divided between two castes: infrequent workers and frequent workers<sup>3</sup>, with the infrequent workers amounting to 25–40% of the individuals within a colony<sup>3</sup>. Anecdotal evidence suggests that immediately after a period of heavy rainfall, infrequent workers make prospecting forays away from the colony in an attempt

to mate with members of other colonies<sup>4</sup> or found new colonies<sup>5</sup>. This suggests that the primary function of infrequent workers may be to disperse when environmental conditions permit.

We tested this suggestion by measuring the daily energy expenditure of the two castes of Damaraland mole-rat worker and the queens before and after rainfall using the doubly labelled water (DLW) technique<sup>13</sup>. We also measured body fat content by isotope dilution<sup>14</sup>, resting metabolic rate by indirect calorimetry and sustained metabolic scope (the ratio of daily energy expenditure to resting metabolic rate<sup>15</sup>) in all individuals. We predicted that daily energy expenditure should be relatively constant for frequent workers but should increase after rainfall for infrequent workers. We also predicted that if infrequent workers were dispersers, they should have lower energy expenditure while dispersal is constrained and carry more energy reserves to fuel eventual dispersal<sup>4,12</sup>. Different castes had significantly different body masses ( $F_{2,71} = 23.49$ ,  $P < 0.001$ ) but body mass did not differ within castes between seasons ( $F_{1,71} = 0.08$ ,  $P = 0.781$ ). There was no change in body mass during the DLW experimental period for any category of animal either in dry conditions (paired  $t_5 = 0.96$ ,  $P = 0.38$ ,  $t_{13} = 1.64$ ,  $P = 0.12$  and  $t_{20} = 0.94$ ,  $P = 0.36$  for queens, infrequent workers and frequent workers, respectively; where subscript numbers indicate degrees of freedom) or after rainfall ( $t_5 = 1.35$ ,  $P = 0.26$ ,  $t_{12} = 1.55$ ,  $P = 0.15$  and  $t_{17} = 0.61$ ,  $P = 0.55$  for queens, infrequent workers and frequent workers, respectively) (Table 1). Overall, queens weighed  $119 \pm 23.4$  g ( $n = 12$ ), infrequent workers weighed  $136 \pm 33.2$  g ( $n = 27$ ) and frequent workers weighed  $85 \pm 28.5$  g ( $n = 39$ ) (Table 1). There was no difference in body mass between male and female frequent workers ( $76.5 \pm 21.8$  g and  $92.8 \pm 30.8$  g for males and females respectively,  $F_{1,30} = 3.07$ ,  $P = 0.09$ ) but male infrequent workers were significantly heavier than female infrequent workers ( $141.3 \pm 31.9$  g and  $111.3 \pm 26.9$  g respectively,  $F_{1,23} = 4.31$ ,  $P = 0.049$ ). There was a significant effect of caste, but not of season or sex, on the fatness of an individual ( $F_{2,58} = 3.42$ ,  $P = 0.04$ ,  $F_{1,58} = 1.88$ ,  $P = 0.175$  and  $F_{1,58} = 1.39$ ,  $P = 0.244$  for caste, season and sex, respectively). Post-hoc tests (Tukey Least Significant Difference (LSD)) revealed that this was because infrequent workers were significantly fatter than frequent workers; infrequent workers carried, on average,  $14.5 \pm 5.3$  g of fat whereas frequent workers carried only  $7.5 \pm 4.8$  g. The difference in the amount of body fat between castes did not completely account for the difference in total body mass, suggesting that infrequent workers also had greater lean mass. Greater muscle mass may be advantageous when it comes to digging burrows, but the costs and benefits in this circumstance are complex because a larger-bodied individual may be more capable at digging, but require a larger tunnel to fit its body through.

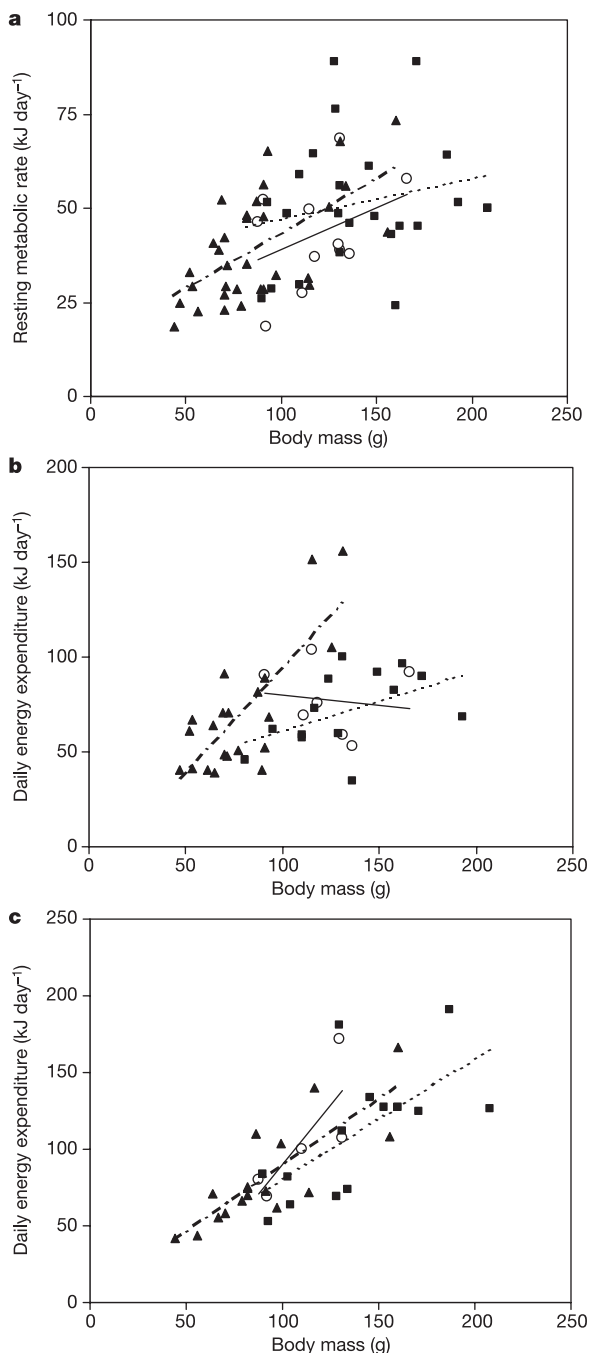
There was a significant positive relationship between resting metabolic rate and body mass ( $F_{1,65} = 20.14$ ,  $P < 0.001$ ; least-squares

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**Table 1 | Energetic measurements**

	Queen during dry conditions (mean $\pm$ s.d.)	Queen during wet conditions (mean $\pm$ s.d.)	Infrequent worker during dry conditions (mean $\pm$ s.d.)	Infrequent worker during wet conditions (mean $\pm$ s.d.)	Frequent worker during dry conditions (mean $\pm$ s.d.)	Frequent worker during wet conditions (mean $\pm$ s.d.)
<i>n</i>	6	6	14	13	21	18
Body mass ( <i>x</i> , <i>y</i> ) (g)	124, 126 $\pm$ 24, 22	110, 107 $\pm$ 23, 20	133, 132 $\pm$ 31, 31	139, 137 $\pm$ 37, 37	78, 79 $\pm$ 23, 23	93, 93 $\pm$ 32, 33
RMR (kJ day <sup>-1</sup> )	48.3 $\pm$ 12.0	48.5 $\pm$ 23.0	55.5 $\pm$ 15.2	59.1 $\pm$ 23.0	44.3 $\pm$ 15.7	43.3 $\pm$ 16.3
Mass-corrected RMR (kJ day <sup>-1</sup> )	45.4 $\pm$ 11.8	48.7 $\pm$ 20.3	49.3 $\pm$ 16.1	52.2 $\pm$ 22.3	50.8 $\pm$ 14.5	47.7 $\pm$ 12.0
DEE (kJ day <sup>-1</sup> )	77.7 $\pm$ 18.8	107.2 $\pm$ 46.0	71.9 $\pm$ 20.0	113.4 $\pm$ 42.5	70.3 $\pm$ 33.4	81.5 $\pm$ 32.5
Mass-corrected DEE (kJ day <sup>-1</sup> )	68.6 $\pm$ 24.4	105.9 $\pm$ 36.5	57.6 $\pm$ 19.1	96.1 $\pm$ 32.4	86.9 $\pm$ 24.7	90.0 $\pm$ 22.3
SusMS	1.64 $\pm$ 0.35	2.53 $\pm$ 1.23	1.39 $\pm$ 0.48	2.20 $\pm$ 1.12	1.87 $\pm$ 0.83	1.88 $\pm$ 0.34
Percentage fat	10.9 $\pm$ 2.0	8.5 $\pm$ 1.9	11.1 $\pm$ 3.5	10.3 $\pm$ 2.9	8.9 $\pm$ 3.5	7.7 $\pm$ 4.0

Mean and standard deviations (s.d.) of body mass, resting metabolic rate (RMR), mass-corrected resting metabolic rate, daily energy expenditure (DEE), mass-corrected daily energy expenditure, sustained metabolic scope (SusMS) and percentage fat for queens, infrequent workers and frequent workers. The two values of body mass (*x*,*y*) indicate measurements taken at the beginning and the end, respectively, of the DLW study. *n* denotes sample sizes. Resting metabolic rate was converted to kJ day<sup>-1</sup> using an oxygen-equivalent of 20.51 kJ l<sup>-1</sup> O<sub>2</sub>; a respiratory quotient of 0.8 was assumed<sup>19,20</sup>.



regression: resting metabolic rate (kJ day<sup>-1</sup>) = 24.1 + 0.227 × body mass (g),  $r^2 = 0.24$ ; Fig. 1a). However, once the effects of mass had been accounted for, there was no additional relationship between season and resting metabolic rate, or caste and resting metabolic rate ( $F_{1,62} = 0.01$ ,  $P = 0.979$  and  $F_{2,62} = 0.26$ ,  $P = 0.771$ , respectively; Table 1).

There was a positive relationship between daily energy expenditure and body mass ( $F_{1,75} = 41.32$ ,  $P < 0.001$ ; least-squares regression: daily energy expenditure (kJ day<sup>-1</sup>) = 22.5 + 0.561 × body mass (g),  $r^2 = 0.36$ ). In addition to this overall mass effect there were complex effects of both caste and season on the levels of daily energy expenditure, which were reflected in a significant three-way interaction between season, caste and body mass on daily energy expenditure ( $F_{2,65} = 3.83$ ,  $P = 0.028$ ). This suggested that there was an effect of caste on the levels of daily energy expenditure, but that this effect differed with season. To explore these effects further we analysed the data for dry and wet seasons separately. In the dry season there was a positive relationship between daily energy expenditure and body mass ( $F_{1,36} = 8.67$ ,  $P = 0.006$ ) and a significant interaction between body mass and caste ( $F_{2,36} = 6.51$ ,  $P = 0.004$ ), indicating that the gradients of the changes in daily energy expenditure with mass were significantly different between castes (Fig. 1b). In absolute terms the energy demands of the different castes were 70.3 kJ day<sup>-1</sup> for the frequent workers, 71.9 kJ day<sup>-1</sup> for the infrequent workers and 77.7 kJ day<sup>-1</sup> for the queens. Once the mass effects had been removed, the mass-corrected daily energy expenditure averaged 87 kJ day<sup>-1</sup> for frequent workers, but only 58 kJ day<sup>-1</sup> and 69 kJ day<sup>-1</sup> for infrequent workers and queens, respectively, which were significantly lower than the costs for the frequent workers ( $F_{2,39} = 7.09$ ,  $P = 0.002$ ; Tukey LSD) (Table 1). In the wet season there was also a significant effect of mass on daily energy expenditure ( $F_{1,29} = 17.58$ ,  $P < 0.001$ ) but this time no significant interaction between body mass and caste ( $F_{2,29} = 0.65$ ,  $P = 0.529$ ); in this season the effect of caste disappeared ( $F_{2,32} = 0.62$ ,  $P = 0.546$ ; Tukey LSD) (Fig. 1c). The mass-corrected daily energy expenditure of the infrequent workers and queens increased during the wet season, to an average 96 kJ day<sup>-1</sup> and 106 kJ day<sup>-1</sup>, respectively, so that their daily energy expenditures did not differ from that of the frequent workers, which was unchanged at an average of 90 kJ day<sup>-1</sup> (Table 1). Absolute energy demands of the three castes in the wet season were 81.5, 113.4 and 107.2 kJ day<sup>-1</sup> for frequent workers, infrequent workers and queens, respectively.

**Figure 1 | Resting metabolic rate and daily energy expenditure for *C. damarensis*.** **a**, Resting metabolic rate against body mass of *C. damarensis* for wet and dry periods combined. **b**, Daily energy expenditure against body mass during a dry period. **c**, Daily energy expenditure against body mass during a wet period. Open circles, solid squares and solid triangles (together with solid, dotted and dotted-dashed least-squares regression lines) denote queens, infrequent workers and frequent workers, respectively.

Because the mass-corrected resting metabolic rate was independent of caste and season, the trends in sustained metabolic scope mirrored the changes in daily energy expenditure. There was a significant interaction between season and caste for sustained metabolic scope ( $F_{2,61} = 3.97$ ,  $P = 0.025$ ). Post-hoc tests revealed that infrequent workers had higher sustained metabolic scope values after rainfall but frequent workers did not (Table 1).

Our results show that infrequent workers are larger than frequent workers and have a higher percentage of body fat. One possibility is that the lower energy demands of the infrequent workers occurred because they were better insulated by their larger fat stores. However, this is unlikely to explain their lower mass-corrected energy demands, because the summer temperatures in the burrow systems are relatively high<sup>16</sup> (averaging  $31.9 \pm 1.0^\circ\text{C}$ ). Thus, these animals routinely live above their lower critical temperatures and do not have significant thermoregulatory demands that would be alleviated by improved insulation. After a period of rainfall, infrequent workers increased their daily energy expenditure and sustained metabolic scope more than did frequent workers, suggesting that rainfall triggers energetically expensive activities such as digging<sup>5,17</sup>. Taken together with previous evidence that dispersing individuals are generally larger<sup>4,12</sup>, and that moist conditions stimulate reproductive activity<sup>18</sup>, these results suggest that infrequent workers constitute a dispersing caste within Damaraland mole-rat colonies. That is, during dry periods infrequent workers, instead of contributing to the work of the colony, build up stores of body fat in preparation for meeting the costs of dispersal and reproduction<sup>3</sup>. Because these individuals are larger and thus have greater energy demands, they place a double burden on the colony—doing virtually no work, but requiring more food.

## METHODS

The study was carried out near the town of Hotazel, South Africa. We trapped mole-rats in March 2003 when conditions were dry (no rainfall for at least a month) and in March 2004 after approximately 40 mm of rain. We injected captured individuals with DLW, released them at the site of capture and then recaptured them after approximately 2–5 days. Of the 55 animals initially captured and injected during the dry period, we re-captured 41 from 8 different colonies; of the 60 individuals initially captured and injected during the wet period, we re-captured 37 from 7 colonies. On completion of experiments, animals were returned to their original capture sites. For full methodological details of measurement of daily energy expenditure and resting metabolic rate, and for details of statistical analysis, see Supplementary Information.

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1. Jarvis, J. U. M., O'Riain, M. J., Bennett, N. C. & Sherman, P. W. Mammalian eusociality: a family affair. *Trends Ecol. Evol.* **9**, 46–51 (1994).
2. Bennett, N. C. & Jarvis, J. U. M. The social structure and reproductive biology of colonies of the mole-rat *Cryptomys damarensis* (Rodentia: Bathyergidae). *J. Mamm.* **69**, 293–302 (1988).
3. Bennett, N. C. & Faulkes, C. G. *African Mole-rats Ecology and Eusociality* 273 (Cambridge Univ. Press, Cambridge, 2000).
4. Hazell, R. W. A., Bennett, N. C., Jarvis, J. U. M. & Griffin, M. Adult dispersal in

the cooperatively breeding Damaraland mole-rat, *Cryptomys damarensis*: a case study from the Waterberg region of Namibia. *J. Zool.* **252**, 19–26 (2000).

5. Jarvis, J. U. M. & Bennett, N. C. Eusociality has evolved independently in two genera of bathyergid mole-rats—but occurs in no other subterranean mammal. *Behav. Ecol. Sociobiol.* **33**, 353–360 (1993).
6. Stacey, P. B. & Koenig, W. D. *Cooperative Breeding in Birds* (Univ. Cambridge press, Cambridge, 1990).
7. Solomon, N. G. & French, J. F. *Cooperative Breeding in Mammals* (Cambridge Univ. Press, Cambridge, 1997).
8. Emlen, S. T. in *Behavioural Ecology an Evolutionary Approach* 4th edn (eds Krebs, J. R. & Davies, N. B.) 228–253 (Blackwell, Oxford, 1997).
9. Emlen, S. T. in *Behavioural Ecology an Evolutionary Approach* 2nd edn (eds Krebs, J. R. & Davies, N. B.) 301–337 (Blackwell, Oxford, 1991).
10. Keller, L. & Reeve, H. K. Partitioning of reproduction in animal societies. *Trends Ecol. Evol.* **9**, 98–102 (1994).
11. Waterman, J. M. Mating tactics of male Cape ground squirrels, *Xerus inauris*: consequences of year-round breeding. *Anim. Behav.* **56**, 459–466 (1998).
12. O'Riain, M. J., Jarvis, J. U. M. & Faulkes, C. G. A dispersive morph in the naked mole-rat. *Nature* **380**, 619–621 (1996).
13. Speakman, J. R. *Doubly Labelled Water, Theory and Practice* (Chapman and Hall, London, 1997).
14. Scantlebury, M., Oosthuizen, M. K., Speakman, J. R., Jackson, C. R. & Bennett, N. C. Seasonal field energetics of the Hottentot golden mole (*Ambysomus hottentotus longiceps*) at 1500m altitude. *Physiol. Behav.* **84**, 739–745 (2005).
15. Hammond, K. A. & Diamond, J. Maximal sustained energy budgets in humans and animals. *Nature* **386**, 457–462 (1997).
16. Bennett, N. C., Jarvis, J. U. M. & Davies, K. C. Daily and seasonal temperatures in the burrows of African rodent moles. *S. Afr. J. Zool.* **23**, 189–195 (1988).
17. Burland, T. M., Bennett, N. C., Jarvis, J. U. M. & Faulkes, C. G. Eusociality in African mole-rats: new insights from patterns of genetic relatedness in the Damaraland mole-rat. *Proc. R. Soc. Lond. B* **269**, 1025–1030 (2002).
18. Molteno, A. J. & Bennett, N. C. Relaxation of socially-induced reproductive inhibition in colonies of the eusocial Damaraland mole-rat: the effect of aridity as an ecological constraint promoting philopatry. *J. Zool.* **256**, 445–448 (2002).
19. Jacobs, D. S., Bennett, N. C., Jarvis, J. U. M. & Crowe, T. M. The colony structure and dominance hierarchy of the Damaraland mole-rat, *Cryptomys damarensis* (Rodentia: Bathyergidae) from Namibia. *J. Zool.* **224**, 553–576 (1991).
20. Hardy, R. N. *Temperature and Animal Life. The Institute of Biology. Studies in Biology no. 35* (Arnold, London, 1972).

Supplementary Information is linked to the online version of the paper at [www.nature.com/nature](http://www.nature.com/nature).

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