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# Energetics and thermoregulation during digging in the rodent tuco-tuco (*Ctenomys talarum*)<sup>☆</sup>

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## Abstract

For subterranean rodents, searching for food by extension of the tunnel system and maintenance of body temperature are two of the most important factors affecting their life underground. In this study we assess the effect of ambient temperature on energetics and thermoregulation during digging in *Ctenomys talarum*. We measured  $\dot{V}O_2$  during digging and resting at ambient temperature ( $T_a$ ) below, within, and above thermoneutrality. Digging metabolic rate was lowest at  $T_a$  within the thermoneutral zone and increased at both lower and higher temperatures, but body temperature ( $T_b$ ) remained constant at all  $T_a$ s. Below thermoneutrality, the cost of digging and thermoregulation are additive. Heat production for thermoregulation would be compensated by heat produced as a by-product of muscular activity during digging. Above thermoneutrality, conduction would be an important mechanism to maintain a constant  $T_b$  during digging.

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**Keywords:** Cost of burrowing; *Ctenomys*; Digging energetics; Thermal stress; Thermoregulation

## 1. Introduction

Subterranean habitats are assumed to be microclimatically stable and predictable (Nevo, 1999), but some conditions may nevertheless be stressful for individuals that inhabit burrow systems (Nevo, 1999; Buffenstein, 2000). Environmental characteristics of the underground environment such as darkness, limited air circulation, high humidity, hypercapnia and hypoxia may drive morphological and physiological adaptations in species that live permanently in sealed burrow systems (McNab, 1966; Busch, 1989; Reig et al., 1990; Nevo, 1999).

Production of metabolic heat while digging in a sealed burrow system with low convection, could cause subterranean ro-

dents to overheat (McNab, 1966; McNab, 1979), especially at ambient temperatures above the thermoneutral zone. Thus, it has been proposed that the low rate of metabolism observed among several species of phylogenetically unrelated subterranean rodents, compared to surface-dwelling species of the same body mass (calculated from Kleiber, 1961), could be an adaptation to avoid overheating during digging (thermal stress hypothesis; McNab, 1966; McNab, 1979). Notwithstanding, below thermoneutrality, a low rate of metabolism could preclude to drive an adequate quantity of energy for thermoregulation when animals are exposed to low  $T_a$ s. In accordance, in laboratory conditions, subterranean rodents show low thermoregulatory capacity below thermoneutrality (Buffenstein, 2000; Busch, 1989). However, in natural conditions, a poor thermoregulatory capacity might be solved through different mechanism. A stable environment should help animals to maintain body temperature, or production of metabolic heat during digging could contribute to the maintenance of a constant body temperature.

Descriptions of the effect of  $T_a$  on thermogenesis due to exercise and cold have been made for various species (Hart, 1971). These studies show that, for some warm-acclimated mammals, thermogenesis and exercise are additive in the range

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Table 1a  
Predictions of substitution and addition hypotheses in the relationship between digging and thermoregulatory costs in *Ctenomys talarum*

		Below thermoneutrality	>	Within thermoneutrality
		$\dot{V}_{O_2}$ resting		$\dot{V}_{O_2}$ resting
Substitution	If	$\dot{V}_{O_2}$ digging	=	$\dot{V}_{O_2}$ digging
	Then	$\dot{V}_{O_2}$ digging– $\dot{V}_{O_2}$ resting	<	$\dot{V}_{O_2}$ digging– $\dot{V}_{O_2}$ resting
Addition	If	$\dot{V}_{O_2}$ digging	>	$\dot{V}_{O_2}$ digging
	Then	$\dot{V}_{O_2}$ digging– $\dot{V}_{O_2}$ resting	≥	$\dot{V}_{O_2}$ digging– $\dot{V}_{O_2}$ resting

in which body temperature increases during exercise (from 10 °C of  $T_a$  to thermoneutrality; Hart, 1952; Hart and Jansky, 1963). However, in other species, addition only occurs in cold-acclimated individuals because muscular work is not used for thermogenesis (Hart and Jansky, 1963). Substitution of heat production from exercise for cold thermogenesis occurs in warm-acclimated mammals and birds because exercise eliminates shivering (Mäkinen et al., 1996; Webster and Weathers, 1990).

*Ctenomys* is the most speciose genus among subterranean rodents (Reig et al., 1990), and includes species from ~100 g (*C. pumdi*) to more than 1000 g (*C. conoveri*), and is distributed in several environments, from sand dunes to mountain hillslopes. They are endemic to the southern cone of South America (Woods, 1984), and typically inhabit sealed burrow systems dug parallel to the soil surface. *Ctenomys talarum* (Thomas, 1898) occurs in the coastal grasslands of Argentina (Comparatore et al., 1991; Antinuchi and Busch, 1992). In this species, physiological parameters such as low resting metabolic rate and high thermal conductance ( $TC$ ) match those found in other unrelated subterranean rodents (Busch, 1989). Therefore, understanding the effect of ambient temperature on digging energetics in a well knowing species, such as *C. talarum*, clues how energetic interactions determine the physiological features observed among subterranean rodents. The present study aims to evaluate the energy interactions between digging and thermoregulation in *C. talarum*. We defined cost of burrowing, estimated by  $\dot{V}_{O_2}$  during digging, as the net cost of burrowing plus the cost of maintenance plus the cost of thermoregulation (see Wunder, 1975).

Assuming that net costs of burrowing are the same among varying temperature trials because the same soil type was used, and if a substitution of heat production from exercise for cold occur, we predict that costs of burrowing within and below thermoneutrality will be the same because below thermoneutrality energy used to dig will remove the cost of thermoregulation. However, if thermogenesis due to cold and exercise is additive, we predict that cost of burrowing below thermoneutrality will be higher than in thermoneutrality because cost of maintaining constant  $T_b$  is not eliminated. Predictions of this hypothesis are summarized in Table 1a.

On the other hand, the cost of burrowing at  $T_a$  above thermoneutrality will be increased because heat produced while digging needs to be dissipated, perhaps by some high-cost mechanism. Assuming that overheating occur during digging above thermoneutrality, predictions of the relationship  $\dot{V}_{O_2}$  at 35 °C/ $\dot{V}_{O_2}$  at 25 °C are summarized in Table 1b.

## 2. Materials and methods

### 2.1. Study animals and soil extraction

Tuco-tucos (*Ctenomys talarum*, Rodentia, Ctenomyidae) of both sexes (a total of 17 males and 17 females) were live-trapped in Mar de Cobo (37°45'S, 57°56'W, Buenos Aires Province, Argentina). Captured animals were carried to the laboratory and housed individually in cages (0.30×0.40×0.25 m). Wood shavings were provided as nesting material. To standardize laboratory conditions, the animal room was kept at 24±1 °C, and photoperiod was LD 12:12 (lights on at 7:00 a.m.). Animals were fed mixed grasses, carrots, lettuce, corn, alfalfa and sunflower seeds ad lib. Water was not provided since *C. talarum* does not drink free water in the wild (Zenuto et al., 2001). Each individual was tested only once at each temperature, to estimate both  $\dot{V}_{O_2}$  during digging and resting.

Soil samples were collected in Mar de Cobo using a steel tube (1 m length and 0.10 m diameter). We dug down to the mean burrow depth (0.20 m below ground), and then ran the steel tube horizontally to extract soil. Each cylinder of soil was transposed to a PVC tube of the same diameter, and carried to the laboratory. This procedure allowed us to maintain the same properties of the intact soil verified in the natural habitat (Antinuchi and Busch, 1992; Luna et al., 2002).

### 2.2. Oxygen consumption

Oxygen consumption was measured using a computerized positive pressure open-flow respirometry system (Sable System, Henderson, NV, USA). The digging chamber system consisted of a cube-shaped acrylic soil collector attached to an acrylic tube. A perforated tube containing the soil extracted in the field, was placed inside the acrylic tube, leaving an air space between the perforated tube and the outer tube (0.01 m) to permit airflow through the system (total volume 19.17 l). A metallic perforated door separated the cube-shaped soil collector and the acrylic tube, allowing free air flow between both. The door had a magnetic lock that allowed it to be opened from outside of the soil collector, allowing to the animal get into the acrylic tube containing the soil, without opening the digging chamber system. A cylindrical resting chamber (volume 1.84 l) was used to estimate  $\dot{V}_{O_2}$  during resting of *C. talarum*. Body mass of tuco-tucos was measured using an electronic balance (AND, model FX-3000, nearest 0.01 g) at the beginning of each burrowing trial.

Digging and resting chamber systems received dry and CO<sub>2</sub> free air at 3 l min<sup>-1</sup> from a flowmeter (Cole-Parmer Instrument Company, Vernon Hills, IL, USA), or at 1.5 l min<sup>-1</sup> from a mass

Table 1b  
Prediction of overheating hypothesis in *Ctenomys talarum*

Within thermoneutrality		Above thermoneutrality
$\dot{V}_{O_2}$ resting	<	$\dot{V}_{O_2}$ resting
$\dot{V}_{O_2}$ digging	<<	$\dot{V}_{O_2}$ digging
During resting		During digging
$\dot{V}_{O_2}$ at 35 °C/ $\dot{V}_{O_2}$ at 25 °C	<	$\dot{V}_{O_2}$ at 35 °C/ $\dot{V}_{O_2}$ at 25 °C

Table 2

$\dot{V}O_2$  during digging (DMR) and during resting (RMR) of *Ctenomys talarum* at different ambient temperatures ( $T_a$ )

$T_a$	Mass (g)	DMR (ml O <sub>2</sub> h <sup>-1</sup> )	RMR (ml O <sub>2</sub> h <sup>-1</sup> )
15 °C	131.61±9.98 (10)	426.78±39.84 <sup>a1</sup>	179.12±13.22 <sup>d2</sup>
25 °C	126.48±5.81 (14)	260.24±18.53 <sup>b1</sup>	116.79±5.00 <sup>e2</sup>
35 °C	142.46±11.32 (10)	577.32±51.92 <sup>c1</sup>	193.58±19.63 <sup>d2</sup>

Values are  $\bar{X} \pm \text{S.E.M.}$   $n$  in parenthesis. Small letters indicate statistical differences between-subject factor ( $T_a$ ), small numbers indicate differences within-treatment factors (DMR or RMR, for whole-animal metabolism, Repeated Measures ANCOVA,  $F_{2,31}=14.82$ ,  $P<0.01$ ).

flow controller (Sierra Instruments, Monterey, CA, USA), respectively. Air passed through a CO<sub>2</sub>-absorbent (IQB®) and water scrubber (Silica Gel) before and after passing through the chamber. Excurrent air from digging and resting chamber systems was subsampled at  $180 \pm 10$  ml min<sup>-1</sup> and oxygen consumption was obtained from an Oxygen Analyzer FC-1B every 0.5 s by a Datacan V—PC program (Sable System, Henderson, NV, USA).

We introduced the animal in the cube-shaped soil collector, closed the metallic perforated door, and connected it to the tube containing the soil, closing the digging chamber system. After that we connected the air pump to the system, to allow the complete turnover of the chamber air. The digging chamber was left for 35 min to achieve equilibration (Lasiewski et al., 1966; Withers, 1977). During this period animals were quiet. After equilibration period, we opened the metallic perforated door, and the tuco-tucos began to burrow within 10 min and continued until it reached the opposite end of the chamber. Data were discarded if animals stopped digging, and remained inactive in the digging chamber system. Oxygen consumption values were calculated using Eq. 4a of Withers (1977). Mean digging metabolic rate was estimated for each individual as the average during the plateau in oxygen consumption in the digging trial.

$\dot{V}O_2$  during resting (RMR) was measured as the 5 min lowest steady-state values of the last 30 min of a 90 min trial. *C. talarum* shows an arrhythmic pattern of activity (Luna et al., 2000), thus digging and resting trials were performed between 9:00 and 17:00 h. Additionally, total time trial was chosen to obtain the metabolism of tuco-tucos (Busch, 1989; Zenuto et al., 2002) at

any day-time. For comparative purposes the data were expressed as mass-independent rate of oxygen consumption (ml O<sub>2</sub> g<sup>-0.67</sup> h<sup>-1</sup>), according to the allometric equation proposed for subterranean rodents (McNab, 1983). No statistical analysis was performed with these data. An equivalent of 19.94 J ml<sup>-1</sup> O<sub>2</sub> was used to convert oxygen consumption to energy values (Vleck, 1979). Oxygen consumption of soil microorganisms was negligible, so the baseline of the digging chamber system was set at 20.95% O<sub>2</sub> at the beginning of each experiment. For measuring the effect of  $T_a$  on  $\dot{V}O_2$  during digging and resting, the whole respirometry system was kept in a temperature controlled room at  $T_a$  of  $15 \pm 1$  °C,  $25 \pm 1$  °C (thermoneutrality, see Busch, 1989), and  $35 \pm 1$  °C. For each temperature condition, a subset of individuals was chosen (10 individuals for 15 °C and 35 °C, and 14 for 25 °C, all with the same proportion of males and females).

Before and after each burrowing trial,  $T_b$  was measured using an electronic digital thermometer (MediLife, accuracy:  $\pm 0.1$  °C), inserted 2 cm into the rectum. Thermal conductance of burrowing individuals was estimated according to Seymour et al. (1998), as  $TC = MR / (T_b - T_a)$ , where MR is the metabolic rate, and  $T_b$  is the final body temperature. After each burrowing trial, the volume of soil removed was estimated. Soil removed samples were dried at 60 °C to constant mass, and moisture percentage was estimated. Average burrowing speed was calculated as total length burrowed divided by the time to complete the digging. In each burrowing trial, the net cost of transport (NCOT) was calculated as ( $\dot{V}O_2$  during digging –  $\dot{V}O_2$  during resting)/burrowing speed (Taylor et al., 1970; Seymour et al., 1998).

### 2.3. Statistical analysis

All values are expressed as mean  $\pm$  S.E.M. Since no differences in DMR or RMR between sexes was found (Luna et al., 2002), no intersexes analysis was performed. On the other hand, since the same individuals were used to measure  $\dot{V}O_2$  in both digging and resting conditions, a Repeated Measures ANCOVA was used to test the null hypothesis that there were no differences in oxygen consumption between digging metabolic rate and resting metabolic rate among ambient temperatures. Between-subject factors were ambient temperature, and within-treatment factors were metabolic rates (digging and resting). Body mass was included as a covariate throughout.

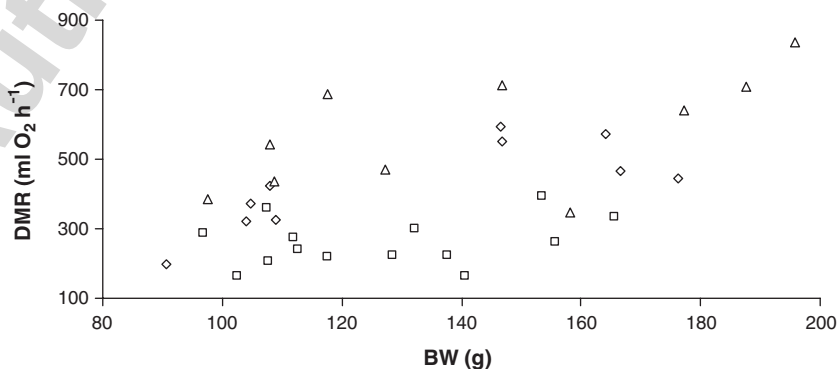


Fig. 1. Effect of ambient temperature on the relationship between  $\dot{V}O_2$  during digging (DMR) and body mass (BW) of *Ctenomys talarum*. Rhombus:  $T_a$  of 15 °C. Squares:  $T_a$  of 25 °C. Triangles:  $T_a$  of 35 °C.

Table 3  
Effect of ambient temperature ( $T_a$ ) on mass-independent  $\dot{V}_{O_2}$  during digging (DMR), mass-independent  $\dot{V}_{O_2}$  during resting (RMR), and DMR–RMR in *Ctenomys talarum*

$T_a$	DMR (ml O <sub>2</sub> g <sup>-0.67</sup> h <sup>-1</sup> )	RMR (ml O <sub>2</sub> g <sup>-0.67</sup> h <sup>-1</sup> )	DMR–RMR (ml O <sub>2</sub> g <sup>-0.67</sup> h <sup>-1</sup> )
15 °C	16.12±1.07 (10)	6.87±0.35 (10)	9.25 (10)
25 °C	10.24±0.71 (14)	4.61±0.16 (14)	5.63 (14)
35 °C	20.95±1.48 (10)	6.89±0.43 (10)	

Values are  $\bar{X}$ ±S.E.M.  $n$  in parenthesis.

ANOVA was used to test the null hypothesis that there were no differences in  $T_b$  after each burrowing trial, mass of soil removed, burrowing speed and net cost of transport among temperature conditions. Afterwards, an ANOVA was also used to test the null hypothesis that there were no differences in thermal conductance among ambient temperatures. Student  $t$  test was used to examine the null hypothesis that there were no differences in DMR–RMR between  $T_a$  of 15 and 25 °C. Because different individuals were used to estimate RMR and DMR at 25 and 35 °C, we selected values randomly from each condition to estimate the ratios: RMR at 35 °C to RMR at 25 °C, and DMR at 35 °C to DMR at 25 °C. We repeat the last procedure 1000 times using Monte Carlo iteration method. After that we calculated the 95% confidence intervals and compared them to determine the differences between both ratios.

### 3. Results

Given that soil samples were extracted in the same place, soil characteristics did not vary among temperature conditions. Average soil humidity (3.34±0.18%) and density (1.61±0.03 g cm<sup>-3</sup>) were similar to those measured by Luna et al. (2002) for gravelly sand. Individuals of *C. talarum* exhibited sexual dimorphism in body mass (BW<sub>male</sub>=155.8±5.4 g, BW<sub>female</sub>=109.6±2.7 g,  $t=-7.96$ ,  $P<0.001$ ).

$\dot{V}_{O_2}$  during digging was different from  $\dot{V}_{O_2}$  during resting in each measured temperature (Scheffé,  $P<0.05$ , Table 2).  $\dot{V}_{O_2}$  during digging of individuals exposed to  $T_a$  of 15 °C was higher than those individuals at  $T_a$  of 25 °C (Scheffé,  $P=0.02$ ). Moreover,  $\dot{V}_{O_2}$  during digging at 35 °C was higher than both at 15 and 25 °C ( $P<0.05$ , Fig. 1, Table 3).  $\dot{V}_{O_2}$  during resting of individuals exposed to 15 and 35 °C were higher than those at

Table 4  
Effect of ambient temperature ( $T_a$ ) on body temperature ( $T_b$ ) after and before digging, delta of body temperature ( $\Delta T$ ), and thermal conductance (TC), after each burrowing trial in *Ctenomys talarum*

$T_a$	$T_b$ before (°C)	$T_b$ after (°C)	$\Delta T$ (°C)	TC (mL O <sub>2</sub> g <sup>-0.67</sup> h <sup>-1</sup> °C <sup>-1</sup> )
15 °C	35.7±0.2	36.1±0.2	0.4±0.2 <sup>a</sup> (10)	0.47±0.06 <sup>a</sup> (10)
25 °C	35.6±0.1	36.7±0.2	1.0±0.1 <sup>a</sup> (14)	0.50±0.09 <sup>a</sup> (14)
35 °C	36.0±0.1	36.5±0.2	0.6±0.1 <sup>a</sup> (10)	9.88±1.44 <sup>b</sup> (10)

Values are  $\bar{X}$ ±S.E.M.  $n$  in parenthesis. Small letters indicate statistical differences within each variable (ANOVA,  $\Delta T$ ,  $F_{2,31}=2.77$ ,  $P=0.08$ ; TC,  $F_{2,31}=88.67$ ,  $P<0.01$ ).

Table 5  
Net cost of transport (NCOT) and burrowing speed (BS) in *Ctenomys talarum* at different ambient temperatures ( $T_a$ )

$T_a$	NCOT (J m <sup>-1</sup> )	BS (m h <sup>-1</sup> )	Expected NCOT (J m <sup>-1</sup> )
15 °C	1162.87±118.27 <sup>a</sup> (10)	4.33±0.31 <sup>a</sup> (10)	
25 °C	647.47±86.13 <sup>b</sup> (14)	4.68±0.56 <sup>a</sup> (14)	3.16* [205] 4.85** [134]
35 °C	1532.56±200.55 <sup>a</sup> (10)	5.16±0.46 <sup>a</sup> (10)	

Values are  $\bar{X}$ ±S.E.M.  $n$  in parenthesis. Small letters indicate differences in each variable among  $T_a$ s by ANOVA test (NCOT,  $F_{2,31}=11.75$ ,  $P<0.01$ ; BS,  $F_{2,31}=0.72$ ,  $P=0.50$ ). \* Expected NCOT for a mammal of 130g (Taylor et al., 1970). \*\* Expected NCOT for a running *C. talarum* at maximum locomotor speed (2.7 km h<sup>-1</sup>, Luna and Antinuchi, 2003). In brackets, observed value in thermo-neutrality to expected value ratio.

25 °C (Scheffé,  $P<0.01$ ). No differences were observed in  $\dot{V}_{O_2}$  during resting between 15 and 35 °C ( $P=0.74$ ).

When DMR–RMR was evaluated, values at  $T_a$  of 15 °C were higher than those observed at 25 °C ( $t=2.94$ ,  $P=0.01$ ). Ratio of RMR at 35 °C to RMR at 25 °C (ratio of means 1.49, 95% confidence interval, 1.35–1.71) and ratio of DMR at 35 °C to DMR at 25 °C (ratio 2.05, 95% confidence interval, 1.81–2.57, see Material and methods for details) were different.

There were no differences in  $T_b$  after each burrowing trial among different  $T_a$  conditions ( $P=0.08$ , Table 4). Thermal conductance was higher at  $T_a$  of 35 °C compared with 15 and 25 °C ( $P<0.01$ , Tukey, 15–25 °C,  $P=0.99$ , 15–35 °C,  $P<0.01$ , 25–35 °C,  $P<0.01$ ).

No differences were found across temperatures in the mass of soil removed per minute ( $\bar{X}=373.5\pm32.7$  g min<sup>-1</sup>,  $F_{2,31}=0.22$ ,  $P=0.80$ ), or mean burrowing speed ( $\bar{X}=4.72\pm0.28$  m h<sup>-1</sup>,  $P=0.84$ , Table 5). The net cost of transport of *C. talarum* was related to ambient temperature ( $P<0.01$ , Table 5), but not to body mass ( $r^2=0.05$ ,  $P=0.29$ ). NCOT at 15 °C, and 35 °C were higher than those at 25 °C (Tukey, 15–25 °C,  $P=0.04$ , 35–25 °C,  $P<0.01$ ). No differences were observed in NCOT between 15 and 35 °C ( $P=0.17$ ).

### 4. Discussion

Subterranean habitats provide their occupants with shelter, protection against predators, access to food resources, and a thermally stable environment (Nevo, 1999). However, there may also be difficulties involved with this lifestyle. For example, limiting food quality and/or quantity may be an important component of sustained survival of subterranean rodents (Buffenstein, 2000), which may limit energy availability and, hence digging metabolic rates (Vleck, 1979). Additionally, a moist and stagnant microenvironment has been hypothesized to limit heat loss of individuals, affecting maximal digging metabolic rate that they can reach (McNab, 1966, 1979).

The DMR to RMR ratio within thermo-neutrality in *C. talarum* was reported as one of the lowest found among subterranean rodents (Luna et al., 2002), even compared with the closely related *Spalacopus cyanus* (Bozinovic et al., 2005). The RMR of *C. talarum* is not different from the expected for subterranean rodents (80% of the expected value of Kleiber equation; Busch, 1989). The low ratio observed, compared to other subterranean rodents (Vleck, 1979; Du Toit et al., 1985; Lovegrove, 1989),

reflect low cost of tunnel construction in gravelly sand (Luna et al., 2002).

As a by-product of the increased metabolic rate associated with digging, heat could potentially be available to supplement thermogenesis. Previous studies on the effects of  $T_a$  on energy cost of submaximal activity suggest that thermoregulatory and exercise costs are additive in some small mammals (*Eutamias merriami*, Wunder, 1970). However, in other mammals (*Rattus norvegicus*, Mäkinen et al., 1996) and birds (*Auriparus flaviceps*, Webster and Weathers, 1990), heat production by muscle activity suppresses thermoregulatory heat production in the cold. As was observed for  $\dot{V}O_2$  during resting, ambient temperatures above or below the thermoneutral zone affect  $\dot{V}O_2$  during digging of individuals burrowing in gravelly sand. DMR–RMR at 15 °C, was higher than that observed in thermoneutrality (Table 3). Therefore, metabolic heat production during digging in *C. talarum* below the thermoneutral zone does not seem to contribute effectively to decrease the cost of maintaining a constant  $T_b$ .

The addition of heat production from exercise and cold appears to be associated with non-shivering thermogenesis (Hart and Jansky, 1963). Although shivering thermogenesis has not been evaluated in *C. talarum*, it is absent in the closely related *Spalacopus cyanus* (Nespolo et al., 2001).

Busch (1989) estimated that during resting at 15 °C,  $T_b$  falls about 4 °C. Thus, because shivering thermogenesis has been shown to be absent in *S. cyanus*, it is unlikely to be present in *C. talarum*, and a fall in  $T_b$  of around 4 °C might therefore be expected during digging, unless heat production is compensated by heat produced as a by-product of muscular activity. Therefore, for *C. talarum* maintenance of  $T_b$  during digging in a cold environment would be the result of non-shivering thermogenesis and the compensation of exercise heat production.

Above the thermoneutral zone, the relationship DMR at 35 °C/DMR at 25 °C and RMR at 35 °C/RMR at 25 °C, appears to fit our predictions. The difference between both ratios could indicate that an additional cost is incurred to dissipate the heat produced as a by-product of digging to maintain a constant  $T_b$ . Thus, dissipation of heat by conduction to the soil is insufficient, thereby necessitating an increase in thermal conductance compared to 15 and 25 °C.

Although evaporation from the body surface is generally an effective way of cooling in surface-dwelling animals (Schmidt-Nielsen, 1990), it is not observed in subterranean rodents that live in stagnant and moist environments. Evaporative and convective heat exchanges are restricted by the lack of air movement and the high humidity conditions commonly found in tunnel systems. Thus, high rates of heat loss between the animal and the soil provide the primary source of dissipating metabolic heat (Buffenstein, 2000; McNab, 1966, 1979).

In the digging chamber, individuals of *C. talarum* effectively address the ventral surface of their body against the soil when this was removed. Moreover, heat loss through the belly could be related to the low density and depth of ventral fur compared to dorsal fur found in *C. talarum* (Cutrera and Antinuchi, 2004), further suggesting that conduction is an important mechanism through which heat is dissipated during digging bouts.

It is not possible to determine directly if conduction or evaporation acts as the main mechanism to prevent overheating in experimental conditions, because dry air was flowing through the digging chamber (see Lasiewski et al., 1966). However, in natural conditions evaporative heat exchanges are restricted and individuals can suffer overheating at 35 °C. Notwithstanding,  $T_a$  in burrows rarely reaches this value (Cutrera and Antinuchi, 2004). A component of the daily energy expenditure is correlated with the distance that an individual travels in a day and its associated cost. Taylor et al. (1970) determined the net cost of transport using the slope of the relationship between metabolic rate and speed for terrestrial locomotion by mammals, and defined the cost of posture as the extrapolation of metabolic cost to zero running speed. Cost of posture (2.08 times RMR; Taylor et al., 1970; Schmidt-Nielsen, 1972) was higher than the range of other species of rodents, because subterranean rodents use more energy to move underground than running. Digging requires scraping the soil from the tunnel work-face, pushing it back along the tunnel, and lifting it to the surface (Vleck, 1979, 1981). Hence, the net cost of transport may depend on soil density, adhesion, moisture content or by thermal balance when an animal digs above thermoneutrality. For a ~130 g-*C. talarum* digging in gravelly sand, NCOT is 205 times the prediction for a running mammal of the same body mass (Table 5), but similar to those observed in other subterranean rodents and insectivores (Lovegrove, 1989; Du Toit et al., 1985; Vleck, 1979; Seymour et al., 1998; Vleck and Kenagy 1987 in Lovegrove, 1989), and 134 times the cost predicted for running tuco-tucos at its maximal locomotor speed (Table 5, Luna and Antinuchi, 2003). NCOT values were different at different  $T_a$ s, despite the high variance in the burrowing speed observed (Table 5). Thus, differences in DMR affect directly estimations of NCOT. Below thermoneutrality, high NCOT can be seen as the addition of both digging and non-shivering thermogenesis costs, since no variation in burrowing speed was found. Above thermoneutrality, high NCOT suggest that an additional cost is incurred to dissipate the heat produced during digging.

In conclusion, our data show that digging is an expensive behaviour in *C. talarum*; digging costs are affected by the thermal environment, but  $T_b$  remains constant. Below thermoneutrality, digging and thermoregulation costs are additive and digging could compensate to maintain constant  $T_b$ . Above thermoneutrality, our experimental conditions do not exclude evaporation and convection as mechanisms for heat dissipation in *C. talarum*. Nevertheless, digging behaviour suggests that conduction is an important avenue through which heat is dissipated to avoid overheating.

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