

Metabolic rate in *Pitymys subterraneus* of two coat-colour morphs

Marek GĘBCZYŃSKI and Elwira SZUMA*

Gębczyński M. and Szuma E. 1993. Metabolic rate in *Pitymys subterraneus* of two coat-colour morphs. Acta theriol. 38: 291 – 296.

Basal metabolic rate (BMR) of the European pine vole *Pitymys subterraneus* (de Selys-Longchamps, 1836) with typical black agouti coat colour (*A*) is $2.78 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$, and in voles with diluted fur pigment (*D*) it is $3.06 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$. The difference of BMR between the voles of *A* and *D* morphs is not significant. Resting metabolic rate (RMR) was determined at ambient temperatures (T_a) -10°C , 0°C , 6°C , 12°C , 24°C and 30°C . *P. subterraneus* of *A* morph decrease RMR from 9.21 (at -10°C) to $4.80 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$ (at 30°C) while the voles of *D* morph decrease RMR from 10.43 to $4.01 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$, respectively. The differences of RMR between the voles of *A* and *D* morphs are significant at T_a -10°C , 0°C , 6°C , 24°C , and 30°C . Maximum oxygen consumption rate (VO_2NA), after injection of noradrenaline, is not dependent on T_a (measurements were made at -10°C , 0°C , 12°C and 24°C). It equals $8.96 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$ in *P. subterraneus* of *A* morph and $10.18 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$ in the voles of *D* morph, and the difference is significant.

Institute of Biology, Warsaw University, Branch in Białystok, Świerkowa 20B, 15-950 Białystok, Poland

Key words: *Pitymys subterraneus*, coat colour morphs, basal and resting metabolic rates, thermoregulation, maximum oxygen consumption

Introduction

Mammalian coat colour depends on a large number of genetic and environmental factors (Searle 1968). Particular colour morphs, depending on environmental conditions, may play different roles in populations. E.g. melanistic *Sciurus carolinensis* survive better under harsh climatic conditions; they have lower heat losses and lower basal metabolic rate in comparison to grey ones (Innes and Lavigne 1979). Differences in body weight and mortality as well as in oxygen consumption rate have been observed among three coat-colour morphs of *Microtus oeconomus* (Krivosheev *et al.* 1983). Diluting mutants connected with *P* and/or *D* series of alleles (Searle 1968, Silvers 1979) may show deep morphological and/or physiological pleiotropic effects. For instance, the dilution of coat pigment in the mouse affects the inner ear, tending to remove the otoliths from the utriculus. Other diluting genes cause sterility in hamsters. Extreme dilution may be combi-

*Present address: Mammal Research Institute, Polish Academy of Sciences, 17-230 Białowieża, Poland

ned with blood anomalies in mink and cattle as well as in humans (cited after Searle 1968).

We are unaware of data dealing with the relationship between dilution of coat colour and metabolic rate. This study therefore addressed the comparison of metabolism and thermoregulation in European pine voles *Pitymys subterraneus* (de Selys-Longchamps, 1836) of normal coat colour and the morph with diluted pelage pigment.

Material and methods

The wild type of *P. subterraneus* in eastern Poland is black agouti. A breeding colony of this species (here called A morph) has been kept for over 25 years in the laboratory of the Mammal Research Institute PAS at Białowieża (Buchalczyk 1961). During that period a line of differently pigmented *P. subterraneus* appeared in the colony. Individuals of the line show coat color dilution, and have pink eyes (*D* morph). The dilution alleles affect the intensity of coat and eye colour. It is claimed (Searle 1968) that they do it through a clumping of pigment granules so that a black fur appears grey. Detailed genetic background of the colour dilution in *P. subterraneus* has not been determined yet.

Body weight of *D* voles is 8.5 g at 22nd day after birth, while in *A* voles it is 10.1 g at the same age. Some events in morphological development (e.g. separation of digits, opening of acoustic meatus and eyes) take place about one day earlier in agouti voles than in the *D* morph individuals (Misiejuk 1984). We also observed, when handling the voles, that individuals of *D* morph are more aggressive than the others.

The European pine voles of both morphs originated from laboratory stock and both were kept under the same laboratory conditions, i.e. T_a 18 – 22°C and natural light regime. Oxygen consumption was measured in adult voles in a closed-type respirometer (Górecki 1975). Basal metabolic rate (BMR) was measured at $T_a = 30^\circ\text{C}$; before the run voles were deprived of food for two hours. Measurements lasted for 30 minutes, with the registration at every two minutes. The lowest values, not fewer than three, were taken into calculations. Resting metabolic rates (RMR) were determined during one-hour periods, and the animals were not fasted before the measurements. RMRs were run at ambient temperatures of -10°C , 0°C , 6°C , 12°C , 18°C , 24°C , and 30°C . Maximum oxygen consumption (VO_2NA) was determined in non-fasted voles, after injection of noradrenaline, at ambient temperatures -10°C , 0°C , 12°C , and 24°C . Noradrenaline (NA) was injected intramuscularly in a dose according to Heldmaier's (1971) formula:

$$\ln \text{NA (mg/kg)} = 1.82 - 0.458 \ln W,$$

where W is body mass in g. The measurements lasted for 45 minutes with registration of oxygen consumption every 90 sec.

Regressions of oxygen consumption *versus* ambient temperature were computed by the method of least squares. Regression coefficients were compared by *t*-test (Zar 1984). The statistical significance of differences between group means were tested by ANCOVA – with two factors (RMR and VO_2NA) or one (BMR) – and log/log body mass as the covariate.

Results and discussion

Basal metabolic rate

Oxygen consumption rate measured under basal conditions in *P. subterraneus* of *A* morph did not statistically differ from those in the voles of *D* morph. When

Table 1. Basal metabolic rate (BMR) in *P. subterraneus* of two coat colour morphs determined at ambient temperature 30°C. A – voles with black coat colour (agouti), D – voles with diluted colour of pelage and pink eyes, n – number of voles examined.

| | A | D |
|---|-------------|-------------|
| n | 8 | 7 |
| Body mass g ± SD | 17.8 ± 1.0 | 15.2 ± 1.2 |
| cc O ₂ g ⁻¹ hr ⁻¹ ± SD | 2.78 ± 0.36 | 3.06 ± 0.28 |
| kJ animal ⁻¹ day ⁻¹ | 23.87 | 22.43 |

oxygen consumption values were calculated in kJ (assuming that 1 cc O₂ equals 20.10 J) per animal per day, and it appeared slightly higher in A voles than in D voles (Table 1), but the differences are not significant.

The basal metabolic rate in wild *P. subterraneus* has previously been determined (23.58 kJ/day, Gębczyński 1964), and it is close to BMR values as measured in *P. subterraneus* of A and D morphs. In the North American pine vole *P. pine-torum*, BMR equals 23.88 kJ/day (after Koteja and Weiner 1993). A number of other small rodent species present similar BMR levels (see Koteja and Weiner 1993). In melanistic *Sciurus carolinensis* lower BMR than in the grey morph was observed in winter, but no such differences were found between the colour morphs in summer pelage (Innes and Lavigne 1979).

Resting metabolic rate

Both morphs of *P. subterraneus* demonstrated different thermoregulatory reactions (Fig. 1). The differences between the morphs depends on the temperature level. Below 18°C the metabolic rate changed sharply. The decrease of oxygen consumption between T_a 18°C and -10°C in the voles of A morph is 0.24 cc O₂ g⁻¹ hr⁻¹ × deg., while it is 0.32 cc O₂ g⁻¹ hr⁻¹ × deg. in D morph. There are no changes in RMR among T_a 18°C and 30°C in A morph, and in D morph it is 0.06 cc O₂ g⁻¹ hr⁻¹ × deg., only. The differences in oxygen consumption between the two morphs at T_a -10°C, 0°C, 6°C, 24°C, and 30°C, are significant, while those at T_a 12° and 18°C are not.

Body temperature (T_b) in the European pine vole of the both coat-colour morphs were measured independently of RMRs runs. The T_b shows that at ambient temperature 12°C the voles are normothermic and do not differ from the animals measured immediately after removing from their nest. At T_a 2°C there is a significant ($p < 0.01$) decrease of T_b in agouti, and even deeper ($p < 0.001$) in diluted morphs (Table 2).

In *M. oeconomus* oxygen consumption at T_a 20°C is significantly higher in dark colour morph voles in comparison to light and intermediate ones, while at T_a 2°C dark and intermediate morphs show significantly higher metabolic rates than do

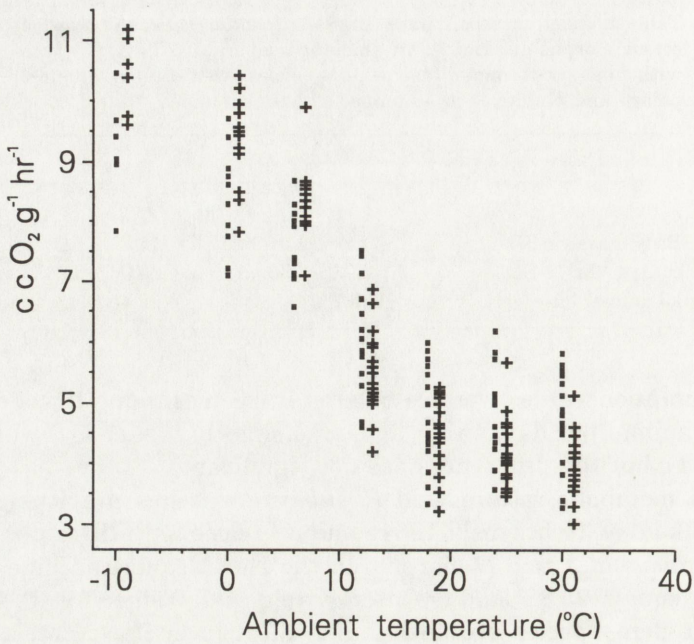


Fig. 1. Resting metabolic rate in *Pitymys subterraneus* at different ambient temperatures. Regressions of oxygen consumption versus ambient temperature were computed.

Dots: *P. subterraneus* with agouti coat colour (A morph): $y = 8.17 - 0.15x$; $r = -0.78$ (at $T_a -10^\circ\text{C}$ to 12°C). Crosses: *P. subterraneus* with diluted coat colour and pink eyes (D morph): $y = 8.92 - 0.24x$; $r = -0.88$ (at $T_a -10^\circ\text{C}$ to 12°C). Data for voles of D morph have been shifted to the right, for clarity.

Table 2. Body temperature (T_b) in *Pitymys subterraneus* of two coat-colour morphs. Body temperature was measured rectally with an electric thermometer, with F6 probe accurate to 0.1°C . Voles were removed from their nests (nest), or after one hour at $T_a 12^\circ\text{C}$ (12°), or after one hour at $T_a 2^\circ\text{C}$ (2°). n - number of voles examined. Description of the morphs in Table 1.

| Coat-colour morph | Nest | 12° | 2° |
|-------------------|-----------------------------|-----------------------------|-----------------------------|
| | $\bar{x} \pm \text{SD}$ (n) | $\bar{x} \pm \text{SD}$ (n) | $\bar{x} \pm \text{SD}$ (n) |
| A | 37.8 ± 0.6 (70) | 37.5 ± 0.6 (66) | 36.4 ± 0.7 (70) |
| D | 37.6 ± 0.6 (34) | 36.8 ± 1.1 (31) | 34.2 ± 1.8 (34) |

the voles of light coat colour. In *M. oeconomus* the increase of oxygen consumption between $T_a 18^\circ\text{C}$ and 2°C was 0.20 , 0.18 and $0.15 \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1} \times \text{deg.}$, in intermediate, dark and in the light morph voles, respectively (Krivosheev *et al.* 1983).

In the both morphs of *P. subterraneus* fairly broad thermoneutral zones were found (Fig. 1). However, the metabolism level within the zones is lower in D morph than in the ones of A morph voles. Such a broad thermoneutral zone has been determined in some other small mammals, e.g. *Microtus arvalis* and *Lagurus*

lagurus (Bashenina 1960), as well as in *Alticola argentatus* and *Ochotona daurica* (Weiner and Górecki 1981).

The two *P. subterraneus* colour morphs significantly differ (*t*-test, $p < 0.001$) with regard to the slope of their metabolic response calculated for T_a range -10°C to 12°C . The elevations of the regression lines are significant ($p < 0.002$) as well.

Maximum metabolic rate

There was no effect of ambient temperature upon the thermogenic response after NA injection in both colour morphs of the European pine voles. Maximum oxygen consumptions averaged $8.96 (\pm 0.87) \text{ cc O}_2 \text{ g}^{-1} \text{ hr}^{-1}$ in A morph, and $10.18 (\pm 0.97)$ in D morph, respectively. The difference is significant. It suggests that noradrenaline-induced heat production, being an equivalent to physiologically induced non-shivering thermogenesis (Böckler *et al.* 1982), is higher in *P. subterraneus* with diluted pelage than in voles having dark coat colour.

Table 3. Maximal oxygen consumption ($\text{VO}_{2\text{NA}}$) in *P. subterraneus* of two coat colour morphs, after injection of noradrenaline, at different ambient temperatures. Descriptions of the morphs in Table 1.

| | Ambient temperature ($^\circ\text{C}$) | | | |
|---|--|------------------|------------------|-----------------|
| | -10 | 0 | 12 | 24 |
| Agouti | | | | |
| <i>n</i> | 6 | 8 | 8 | 10 |
| Body mass, g \pm SD | 16.3 \pm 1.1 | 15.1 \pm 0.8 | 16.0 \pm 1.7 | 16.3 \pm 2.3 |
| cc $\text{O}_2 \text{ g}^{-1}\text{hr}^{-1} \pm$ SD | 9.20 \pm 1.00 | 9.33 \pm 0.73 | 8.68 \pm 0.85 | 8.71 \pm 0.88 |
| Diluted | | | | |
| <i>n</i> | 6 | 6 | 8 | 12 |
| Body mass, g \pm SD | 15.5 \pm 1.4 | 16.0 \pm 3.2 | 15.0 \pm 1.0 | 16.7 \pm 2.3 |
| cc $\text{O}_2 \text{ g}^{-1}\text{hr}^{-1} \pm$ SD | 10.45 \pm 0.77 | 10.31 \pm 1.03 | 10.26 \pm 0.51 | 9.92 \pm 1.27 |

Presumably non-shivering thermogenesis enhances survival in cold-acclimated mammals (Jansky 1973), and is negatively correlated with body size (Heldmaier 1971, Weiner 1989). However, both morphs of *P. subterraneus* were kept under the same conditions and their body mass is too small to explain the observed differences in maximal oxygen consumption (Table 3). Therefore the diluted colour mutations in the European pine vole affects some other physiological mechanism. It might be that the same mechanism differentiates RMR of the both morphs, within thermoneutral zone as well as at low ambient temperatures, but does not affect their basal metabolism.

Acknowledgements: The studied European pine voles originated from Mammal Research Institute, Polish Academy of Sciences at Białowieża; where some RMR measurements were obtained. Dr J. Weiner has kindly suggested the methods of statistical analysis and critically commented on an earlier draft of the manuscript. Three anonymous referees are thanked for their remarks.

References

- Böckler H., Steinlecher S. and Heldmaier G. 1982. Complete cold substitution of noradrenaline-induced thermogenesis in the Djungarian hamster, *Phodopus sungorus*. *Experientia* 38: 261 – 262.
- Bashenina N. V. 1960. Materials on the ontogenesis of thermoregulation in the common field mouse (*Microtus arvalis* Pall.) and *Lagurus lagurus* Pall. [In: Fauna and ecology of the rodents. A. N. Formosov, ed]. Publ. House of Moscow Univ., Moscow 6: 111 – 154. [In Russian with English summary]
- Buchalczyk A. 1961. *Pitymys subterraneus* (de Séllys-Longchamps, 1835) under laboratory conditions. *Acta theriol.* 4: 282 – 284.
- Gębczyński M. 1964. Effect of light and temperature on the 24-hour rhythm in *Pitymys subterraneus* (de Sél.-Long.). *Acta theriol.* 9: 127 – 137.
- Górecki A. 1975. Kalabukhov-Skvortzov respirometer and resting metabolic rate measurements. [In: Methods of ecological bioenergetics. W. Grodziński, R. Z. Klekowski and A. Duncan, eds]. Blackwell, Oxford, London, Edinburgh, Melbourne: 310 – 314.
- Heldmaier G. 1971. Zitterfreie Wärmebildung und Körpergrösse bei Säugetieren. *Z. vergl. Physiol.* 73: 222 – 248.
- Innes S. and Lavigne D. M. 1979. Comparative energetics of coat colour polymorphs in the great eastern grey squirrel, *Sciurus carolinensis*. *Can. J. Zool.* 57: 585 – 592.
- Jansky L. 1973. Non-shivering thermogenesis and its thermoregulatory significance. *Biol. Rev.* 48: 85 – 132.
- Koteja P. and Weiner J. 1993. Mice, voles and hamsters: metabolic rates and adaptive strategies in muroid rodents. *Oikos* 66: 505 – 514.
- Krivosheev V. G., Tzvetkova A. A. and Umantzeva N. D. 1983. Polymorphism by fur colour and morpho-physiological differences in colour morphs in a tundra population of *Microtus oeconomus*. *Zool. Z.* 62: 1087 – 1096. [In Russian with English summary]
- Misiejuk M. 1984. Rozwój postnatalny u darniówek (*Pitymys subterraneus*) pochodzących z dwóch hodowlanych linii genetycznych. M. Sc. thesis, manuscript. Białystok. [In Polish]
- Searle A. G. 1968. Comparative genetics of coat colour in mammals. Logos Press, London: 1 – 308.
- Silvers W. K. 1979. The coat colors of mice. A model for mammalian gene action and interaction. Springer Verlag, New York, Heidelberg, Berlin: 1 – 379.
- Weiner J. 1989. Metabolic constraints to mammalian energy budgets. *Acta theriol.* 34: 3 – 35.
- Weiner J. and Górecki A. 1981. Standard metabolic rate and thermoregulation in five species of Mongolian small mammals. *J. comp. Physiol.* 145: 127 – 132.
- Zar J. H. 1984. Biostatistical analysis. Prentice Hall, Englewood Cliffs, New York: 1 – 718.

Received 23 April 1993, accepted 5 October 1993.