NORTH-WESTERN JOURNAL OF ZOOLOGY 7 (1): pp.47-54 ©NwjZ, Oradea, Romania, 2011
Article No.: 111103 www.herp-or.uv.ro/nwjz

Head-body length variation in the mole-shrew (*Anourosorex squamipes*) in relation to annual temperature and elevation

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Received: 09. April 2010 / Accepted: 17. January 2011 / Available online: 25. January 2011

Abstract. Temperature change may affect physiology, distribution, morphology and adaptation of animals. Using individuals captured in Nanchong and Laojunshan Nature Reserve of Sichuan province in western China, we tested head-body length variation in the mole-shrew (*Anourosorex squamipes*) in relation to annual temperature and elevation. Our results indicate that head-body length of both males and females decreases with decreasing ambient temperature along an altitudinal gradient. Likewise, there is significant monthly variation in head-body length, with individuals in warmer months being larger than that colder ones. The seasonal variation in head-body length suggests the higher predator risk and lower food availability in cold months.

Keywords: Body size, *Anourosorex squamipes*, temperature, elevation.

Introduction

Body size has long been considered one of the most important traits of an organism because it influences nearly every aspect of its life history (Peter 1983). Temporal and spatial variation in body size of animals is related to many ecological factors, including ambient temperature, food availability, character displacement, precipitation, predator risk, seasonality and evapotranspiration (Yom-Tov 2001, Ochocinska & Taylor 2003, Yom-Tov & Yom-Tov 2005, Medina et al. 2007, Kryštufek & Quadracci 2008, Entling et al. 2010, Opell 2010, Liao & Lu 2010a,b, Liao et al. 2010). Of these factors, ambient temperature plays an important role in determining optimum body size in animals (Ashton 2002, Ashton & Feldman 2003). In this respect, Bergmann's rule is currently defined as a within-species tendency for in-creasing body size with increasing latitude or decreasing environmental temperature for endothermic vertebrates (Bergmann 1847, Atkinson & Sibly 1997, Ashton et al. 2000, Angilletta & Dunham 2003).

Geographical and seasonal variation in body size of small mammals across different ambient temperatures has been widely reported in recent years (Baumgardener & Kennedy 1993, Smith et al. 1995, Motokawa et al. 1996, Storz et al. 2001, Meiri & Dayan 2003, Ochocinska & Taylor 2003, Yom-Tov & Yom-Tov 2004, Liao et al. 2006, Medina et al. 2007, Lin et al. 2008). Shrews are among the smallest extant small mammals, and there are conflicting reports on correlation between body size and temperature among populations. For example, body size of the short-tailed shrew (*Blarina brevicauda*) negatively correlates with ambient temperature (Jones & Findlay 1954, Huggins & Kennedy 1989). On the other hand, body size of the masked shrew (*Sorex cinereus*) in Alaska decreases with decreasing temperature (Yom-Tov & Yom-Tov 2005). The opposite trends in body size could be probably related to food availability associated with ambient temperature in different seasons, as docu-mented for several shrew species (Ochocinska & Taylor 2003, Yom-Tov & Yom-Tov 2005). Moreover, predatory risk may also affect body size distribution since larger individuals may be at a disadvantage to access burrows or cover (Brown 1995).

The mole-shrew (*Anourosorex squamipes*) is a soricine species living in forest and burrowing in leaf litter and topsoil. It is distributed in Bhutan, Assam, northern Myanmar, northern Thailand, northern Vietnam, Taiwan and western and central China from the provinces of Sichuan and Yunan to Hubei (Motokawa & Lin 2002). It is active all year round and its longevity is about 1.0 yr (Zong 1998). It occurs in a broad range of ambient temperature, including winter, when absolute minimum temperature reaches -3.4°C and summer when absolute maximum temperature is 41.2°C (Liao et al. 2005a). To date, despite the detailed studies on geographic variation and ecology of the species (Yu & Liao 2000, Liao et al. 2004, Motokawa et al. 2004, Liao et al. 2005a,b), information on temporal and spatial variation in body size is unavailable. For this study we used data on head-body length as an indicator of body size for 675 sexually mature individuals of *A. squamipes*. Body mass is highly correlated with metabolism in mammals and may make actually much more sense in the context of temperature/size correlation (McNab 1971). However, variations in pregnancy of females and testicles of males influence body mass, thus do not reflect a true body size (Liao et al. 2005b). In this study we hypothesized that monthly and elevational variation in head-body length of the mole-shrews changes with ambient temperature. We expected increased head-body length with decreasing ambient temperature. As a corollary, we examined if food availability was associated with ambient temperature and body size.

Materials and methods

Study area

Fieldwork was conducted from September 2003 to August 2004 in Nanchong (30º48′N, 106º06′E, and 310m a.s.l.) and from April to June 2005 in Laojunshan Nature Reserve of Sichuan province in western China. The climate of Nanchong is characteristic of eastern Asia. The study area has annual average temperature of 17.5°C (sub-zero mean monthly temperatures occur in January–February) and annual total precipitation of 1000mm. The vegetation covering the study site is characterized by Camphor tree (*Cinnamowum camphora*), Black locust (*Robinia pseudocacia*), Willow (*Salix babylonica*), Cedar (*Cedeus deodara*), Fortunes windmill palm (*Trachycarpus fortunei*) and Sikui (*Washingtonia robusta*).

Laojunshan Nature Reserve ranges from 1100 to 2008m a.s.l., has an annual average temperature of 12.5°C, and annual average precipitation of 1500mm (Liao et al. 2008). Vegetation is dominated by Hornbeam (*Carpinus fargesii)*, Tea oil (*Camellia oleifera*), Lianxiang tree (*Cercidiphyllum japonicum*), Dove tree (*Davida involucrate*), Common eurya (*Eurya loquiana*), Azalea (*Rhododendron hunnewellianum*) and Bamboo (*Cnimonobambusa quadrahgu*). Samples were collected in Eryanping Protection Station (28°40′N, 103°51′E, and 1500m a.s.l.) and Laojunshan Protection Station (28°42′N, 103°54′E, and 2000m a.s.l.).

We identified adults and juveniles based on developmental degree of ovary and testicles (Liao et al. 2005b). For all individuals trapped, we measured body weight to the nearest 0.1g using an electronic balance, and head-body length, tail length, hind foot length and ear length of each individual to the nearest 0.02mm using vernier calipers. Climate data (mean monthly temperatures in °C for each 1 degree square) were extracted from Nanchong Weather Station for the period 1971–2000. For Eryanping Protection Station and Laojunshan Protection Station, we obtained variation in ambient temperatures from Weather Station of Pingshan County for the period 1971–2000.

Stomach contents can indicate food availability in *A. squamipes* (Hu & Wang 1984). In Nanchong site, each month stomach contents were collected from 20 specimens using stomach-flushing method (Zong 1998) and stored in separate airtight test tubes in 4% formalin. Prey items were subsequently identified in the laboratory under a binocular microscope. Food composition was evaluated by percentage abundance (%).

Statistical analysis

Sex ratio and percentage abundance (%) of stomachs contents were tested using the Chi-square test. Differences in head-body lengths between males and females were tested using the Student's *t*-test. Correlation between head-body length and temperature was estimated using linear regression. Variations in head-body lengths were examined using a general linear model (West et al. 1997). In the full model, we included temperature as a fixed factor. Latitude, elevation and sex were included as covariates. We used partial correlation analysis rather than simple bivariate correlation analysis to indicate the relationships between temperature and head-body length when the effects of elevation and season were removed. Despite the fact that there was a lack of consistency in methods used to collect data on trapping mode and effort across three sites, this would not introduce a bias in the analysis due to the fact that individuals resulting from random samples were analyzed. Statistical analyses were performed with SPSS 13.0 for Windows packages with P = 0.05 used as a threshold for significance testing. All values given are shown as mean ± SE.

Results

From September 2003 to August 2004 we collected 812 *A. squamipes* individuals, including 624 adults (297 males and 327 females) and 188 juveniles at Nanchong site. The sex ratio did not differ significantly from 1:1 (Chi-square test: $\chi^2 = 1.44$, $df = 1$, $P > 0.05$). Average head-body length did not differ significantly between females and males (females, 97.76 ± 0.39 mm; males, 96.75 ± 0.36 mm; Student's *t*-test: $t = 1.87$, $P = 0.07$). Within each month, the difference between male and female head-body length was not significant (Table 1). From April to June 2005 we collected 51 adults (13 males and 9 females at the 1500m site and 17 males and 12 females at the 2000m site). A male biased sex ratio was not found in Eryanping Protection Station (sex ratio: 1.5:1.0, $\chi^2 = 1.50$, $df = 1$, $P > 0.05$) and Laojunshan Protection Station (sex ratio: 1.4: 1.0, $\chi^2 = 0.94$, $df = 1$, $P > 0.05$). Head-body length of females was significantly larger than males at both sites (Laojunshan: females, 94.00 ± 1.24 mm; males, 90.61 ± 0.63 mm; $Z = 2.60$, $P < 0.01$; Eryanping: fe-

Table 1. The difference of body size between male and female *Anourosorex squamipes* during the twelve months from September 2003 to August 2004 in Nanchong, western China.

Month	Sex	Samples	Body length (mm) Mean ± SE	*U* -test	*P* -value
January	♀	14	92.97 ± 1.02	$Z = 1.52$	$P = 0.13$
	♂	19	91.06 ± 1.05		
February	♀	19	92.79 ± 1.16	$Z = 0.19$	$P = 0.85$
	♂	10	92.50 ± 1.49		
March	♀	24	100.69 ± 0.98	$Z = 1.16$	$P = 0.21$
	♂	32	98.87 ± 0.76		
April	♀	25	102.72 ± 0.83	$Z = 1.01$	$P = 0.11$
	♂	30	101.47 ± 0.64		
May	♀	24	102.58 ± 1.08	$Z = 1.59$	$P = 0.08$
	♂	17	101.71 ± 0.80		
June	♀	27	102.33 ± 0.86	$Z = 0.92$	$P = 0.19$
	♂	30	101.77 ± 0.49		
July	♀	21	103.81 ± 1.09	$Z = 1.71$	$P = 0.09$
	♂	15	102.60 ± 0.86		
August	♀	27	103.11 ± 0.71	$Z = 1.08$	$P = 0.28$
	♂	21	102.14 ± 0.79		
September	♀	15	98.80 ±1.20	$Z = 1.77$	$P = 0.10$
	♂	11	96.18 ± 0.72		
October	♀	37	96.12 ±1.05	$Z = 1.51$	$P = 0.13$
	♂	30	94.02 ± 1.01		
November	♀	53	91.48 ± 0.75	$Z = 0.86$	$P = 0.39$
	♂	52	92.49 ± 0.76		
December	♀	41	93.29 ± 0.73	$Z = 2.14$	$P = 0.21$
	♂	30	90.77 ± 0.95		

males, 95.28 ± 0.98 mm; males, 92.38 ± 1.07 mm; $Z = 1.98$, $P < 0.05$).

Monthly variation in ambient temperature in twelve months was marked (Fig. 1; Kruskal-Wallis *H*-test: $H = 209.00$, $df = 11$, $P < 0.001$). During the twelve months, there was a significant monthly variation of head-body lengths in both sexes (Fig. 2; one-way ANOVA: females, $F = 33.74$, $df = 11$, $P < 0.001$; males, $F = 42.26$, $df = 11$, $P < 0.001$). Head-body length in males was positively correlated to mean monthly temperature (temperature = -118.01 + 1.38 body size, $F = 5.42$, $r^2 = 0.35$, $P = 0.04$). Similarly, head-body length in females was positively correlated with mean monthly temperature (temperature = -115.05 + 1.33 head - body length, $F = 6.69$, $r^2 = 0.40$, $P = 0.03$).

UNIANOVA with elevation, latitude and season as fixed factors showed that elevation and season were two main factors affecting difference in ambient temperature (elevation, $F_{2,\ 674} = 4.69$, $P = 0.01$; season, $F_{11,\ 665} = 1105.94$, $P < 0.001$), and latitude among three sites did not strongly affect the ambient temperature ($F_{1,\ 674} = 0.72$, $P = 0.40$). Average ambient temperatures at the 2000-m site (13.1 ± 2.4°C) were significantly lower than those at both the 1500-m site (15.6 ± 2.1°C; Mann–Whitney *U*-test: $Z = 2.29$, $P = 0.022$) and the 310-m site (17.5 ± 3.2°C; $Z = 2.34$, $P = 0.02$) sites. Difference was also found between the later two populations (Mann–Whitney *U*-test: $Z = 2.12$, $P = 0.05$). Moreover, elevation had a significant effect on temperature variability (as an indicator of climatic harshness or predictability), which was higher at the 2000-m site than at both lower elevations ($F_{2,\ 17} = 3.61$, $P < 0.05$).

We used a general linear model to gain insight on spatial variation in adult head - body length across ambient temperatures at three different elevations. This is because both the effects of elevation and seasonal variation are addressed simultaneously. An initial analysis of adult head-body length indicated that none of the possible two-way interactions between sex, latitude and temperature were significant ($F_{1,\ 674} \geq 1.513$, $P \geq 0.219$). Therefore, the analysis was limited to an evaluation of the main effects only. While the effect of latitude

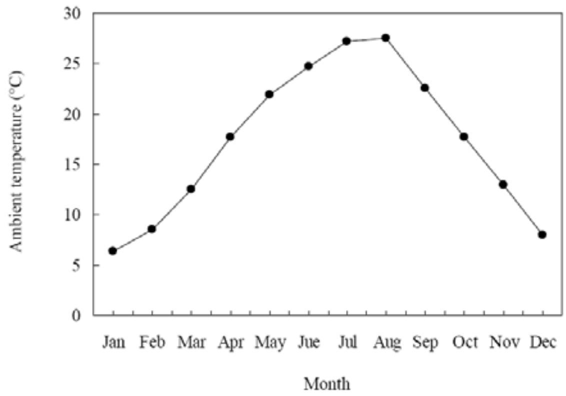

Figure 1. The variation of ambient temperature during the twelve months in Nanchong, western China (Data resulting from Weather Station of Nanchong).

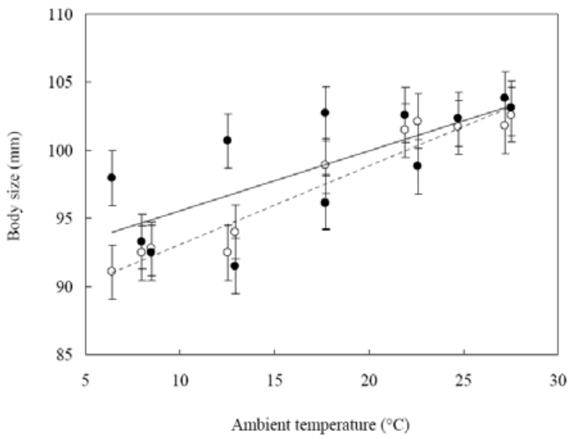

Figure 2. Monthly varation in body size of *Anourosorex squamipes* males (empty circle) and females (solid circle) from September 2003 to August 2004 at Nanchong in western China.

was not significant ($F_{1,\ 674}$ = 0.42, P = 0.67), both the elevation and season temperature terms explained significant amounts of variation in the mean head-body length of adults (season, $F_{1,\ 674}$ = 49.22, $P < 0.001$, elevation, $F_{1,\ 674}$ = 12.67, $P < 0.001$). Head-body length increased with increasing ambient temperature when the effects of elevation and season were removed (Fig. 3; female, F= 14.54, $P < 0.001$; male, F = 13.47, $P < 0.001$).

We only analyzed stomachs contents from 144 specimens at Nanchong site. Besides invertebrate prey, plant materials, and shed skin were identified (Table 2). Food availability (percent of empty stomachs) showed major changes during the study period (Chi-square test: χ^2 = 78.33, df = 11, $P < 0.001$). The occurrence of empty stomachs was correlated negatively with ambient temperature (Fig. 4; F = 58.49, r^2 = 0.85, $P < 0.001$). Significant variations were recorded in the frequency of stomachs containing plant material and also invertebrate prey (Ophisthopora and Hymenoptera, but not Coleoptera) (Table 2).

Discussion

Our study demonstrated an altitudinal and seasonal decrease in head-body length with decreasing ambient temperature in *A. squamipes*. This is similar to observations made previously on small mammals in general (Mezhzherin 1964, Freckleton et al. 2003, Yom-Tov 2003, Millien 2004, Yom-Tov & Yom-Tov 2005). However, the findings are the inverse of the increase in body size with decreasing ambient temperature in other *Sorex* species (Kirkland & Van Deusen 1979, Huggins & Kennedy 1989).

The heat conservation prediction indicates that temperature variation should be the best environmental explanatory variable for average body size. It also indicates that covariation of body size with temperature should be stronger for small-bodied than large-bodied species because their greater surface area to volume ratios mean that small bodied species face more of a challenge in keeping warm than do large-bodied species (James 1970). However，there is no definitive evidence that correlations between temperature and body size should be stronger in smaller mammals (Ashton et al. 2000, Meiri & Dayan 2003, Meiri et al. 2007). Our results showed that head-body length of the mole-shrew declined with decreasing ambient temperature, thus behaving opposite to the heat conservation hypothesis, similar to other *Sorex* species in the northern Palearctic region (Mezhzherin 1964, Ochocinska & Taylor 2003, Yom-Tov & Yom-Tov 2005).

Body size is one of the key phenotypic traits of

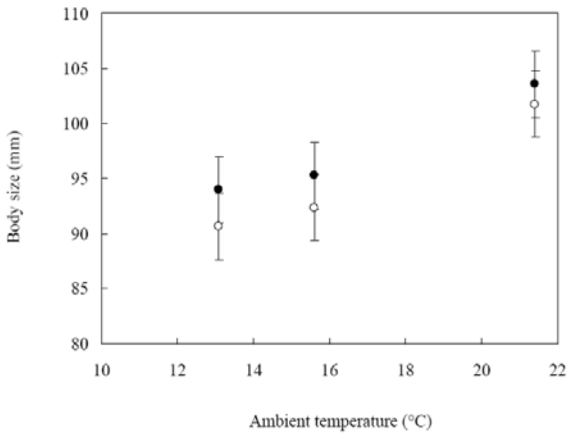

Figure 3. The relationship between ambient temperature and body size of *Anourosorex squamipes* males (empty circle) and females (solid circle) in three sites at different elevations in western China.

Table 2. The percentage abundance (%) of stomachs contents with plant debris, shed-skin and invertebrate content from September 2003 to August 2004 in Nanchong, western China.

Month	Vegetation	Sichuan shrew	Coleoptera -imago	Ophisthopora	Hymenoptera	Others
January	48.1	32.4	0	0	0	19.5
February	44.6	38.2	0	0	0	17.2
March	22.6	22.8	10.2	12.1	27.3	5.0
April	24.5	3.7	9.8	32.1	25.4	6.5
May	26.6	4.7	11.6	24.2	28.6	4.3
June	15.8	2.1	13.4	32.2	30.8	5.7
July	16.2	1.5	16.7	36.6	27.9	1.1
August	14.9	2.6	13.6	27.6	30.7	0.4
September	18.7	2.4	10.8	30.5	28.7	9.9
October	22.9	8.4	7.8	29.6	19.8	12.7
November	33.6	29.4	5.2	4.6	10.4	15.8
December	46.2	37.7	0	0	0	16.1
χ^2	58.64	162.90	8.72	34.21	14.25	37.76
P	0.000	0.000	0.37	0.000	0.008	0.000

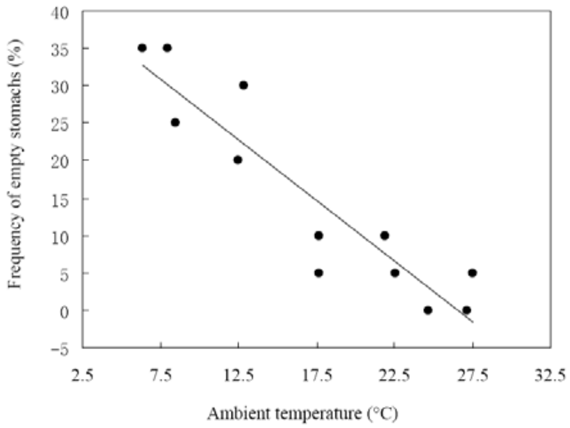

Figure 4. The correlation between the percentage abundance of empty stomachs of *Anourosorex squamipes* and ambient temperature during the twelve months from September 2003 to August 2004 in Nanchong in western China.

animals. It has a profound influence on their morphology, physiology and ecology (Yom-Tov 2001). Previous studies have indicated that body size of animals is positively related to effects of predators via survival (Sullivan & Best 1997; Yom-Tov & Yom-Tov 2005). It is well-known that head-body length in *A. squamipes* is positively correlated with age (Liao et al. 2005a). In this study, only adult individuals were used to analyze the relationship between head-body length and temperature. Therefore, the influence of age on head-body length was negligible. Small size could be an adaptation to reduced predation pressure (Heaney 1978, Brown 1995). Moreover, temporal variation in body size was significant due to differential predation in different seasons. Small mammals in winter are the main prey of Short-eared owls (*Asio flammeus*), in which 97.2% of the pellets' content is *A. squamipes*. In summer, owls or other predators have other prey from which to choose besides the shrews (Li et al. 2007). This pattern suggested greater predator risk in cold months, thus resulting in smaller body size in winter. Since shrews live about one year, their life cycle follows closely the annual variation in ambient temperature. Shrew births occur preferentially in summer, increase in body size and more older individuals in warmer months could not be due to linear growth (Liao et al. 2005b). Likewise, higher mortality of the older and larger individuals in winter results in smaller body size in both sexes due to higher energy consumption during the reproductive process in the autumn (Liao et al. 2005a).

The results of this study indicate that there were monthly differences in *A. squamipes* foraging success. The high number of empty stomachs in winter showed food resource scarcity. Food availability might also be an important driver of temporal variation in body size for small-bodied mammals (Kirkland & Van Deusen 1979). Mean ambient temperatures in summer were higher by 21.1°C than in winter. It seems conceivable to assume that these changes resulted in higher food availability in summer, enabling *A. squamipes* to have large body size accordingly. Monthly variation in body size across ambient temperature indicated that food availability was the main cause of body size increase in warmer months (adding to the effect of selective predation), a pattern which shows a cyclic variation in body size over one year.

Along the elevation gradient sampled, we found that shrews from the high-elevation population had an average smaller body size than those from the low-elevation population. Food availability is related to environmental condition and climate (Covaciu-Marcov et al. 2010, Lima et al. 2010), which might be the main factor behind the apparent temperature-body size correlation. A similar increase in body size for the Japanese field mouse (*Apodemus speciosus*) was attributed to elevated ambient temperatures and increased food availability (Yom-Tov & Yom-Tov 2004). Moreover, large temperature variability from high elevations resulted in a smaller body size in *A. squamipes*, and indeed, this seems to be a general pattern among small mammals (Lindstedt et al. 1985, Millar & Hickling 1990).

Acknowledgements. We thank Pan Wen Wu, Ya Bing Liu, and Hong Wu for assistance with field work. Financial support is provided by Sichuan Keystone Subject Foundation (SZD 0420), the Scientific Research Foundation of Sichuan Provincial Education Department (09ZC010) and the Scientific Research Foundation of China West Normal University (09B001, 10A004).

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