

Adaptive latitudinal shifts in the thermal physiology of a terrestrial isopod

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ABSTRACT

Temperature is the most important abiotic factor affecting physiology, ecology and evolution in ectotherms. Do organisms broadly distributed along a latitudinal gradient adapt to local differences in temperature? In ectotherms this question has played a central role in evolutionary physiology. By means of an extensive field study and laboratory experiments, we tested the existence of local adaptations in thermal traits along 10° of latitude in northern-central Chile. We studied behavioural and thermal traits that have been directly connected with the thermal physiology of ectotherms, along the latitudinal gradient. Using three populations of the common woodlouse (terrestrial isopod), *Porcellio laevis*, we examined changes in thermal physiology traits (i.e. thermal tolerance, thermal performance) and behavioural traits (i.e. thermoregulation in the field and the laboratory). Performance (i.e. righting response speed) reached a maximum value at intermediate temperatures, and was highest for the northern population. Chill-coma temperature showed an increase towards high latitude, while heat-coma temperature did not vary between populations. On the other hand, optimum temperature was negatively correlated with latitude. Thus, southern woodlice appear to have evolved towards becoming low-temperature specialists, whereas woodlice from warm regions demonstrate low cold tolerance as well as a higher optimum temperature. Our results demonstrate that *P. laevis* woodlice from different parts of the distribution range show patterns in thermal physiology that covary with the thermal environment (i.e. latitude); thus, *P. laevis* adapts to local environments to increase its performance.

Keywords: ectotherm, thermal sensitivity, thermal tolerance, latitudinal gradient, local adaptation, temperature.

INTRODUCTION

Temperature is perhaps the most important abiotic factor affecting physiology, ecology and evolution in endotherms and ectotherms (Cossins and Bowler, 1987; Huey and Kingsolver, 1993). Because environmental temperature varies in time and space at different scales, organisms are continually challenged to maintain homeostasis (Johnston and Bennett,

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1996). Thus, thermal physiology may be a significant factor underlying the ecological and evolutionary success of organisms (Gilchrist, 1995; Pörtner, 2002). This is particularly true for ectotherms, because nearly all physiological and behavioural traits are sensitive to ambient temperature. Populations are expected to evolve adaptations to local conditions in heterogeneous thermal environments (Huey, 1982). Responses to environmental variation have been characterized in a quite consistent relationship in ectotherms, which relates performance and environmental temperature (Carrière and Boivin, 1997, 2001; Wilson and Franklin, 2000; Huey and Berrigan, 2001; van der Have, 2002). This relationship is a right-shifted function, where performance gradually increases with temperature, until reaching an optimum value, and then decreases abruptly towards higher temperatures (Huey and Stevenson, 1979; Huey and Berrigan, 1996). This function is the consequence of biochemical, physiological, morphological and/or behavioural traits and strategies that allow ectotherms to maintain thermal homeostasis within a wide range of temperatures (Angilletta *et al.*, 2002; Neargarder *et al.*, 2003).

Environmental gradients are common-place in nature and are recognized to exert a major effect on patterns of intraspecific variation (Mizera and Meszéna, 2003). This fact, coupled with natural selection in the long term, produces conspicuous differences between populations, which are finally translated into local adaptations (Olsson and Uller, 2003). In this sense, adaptive shifts in the performance function can result from biological processes that occur over long time-scales (Angilletta *et al.*, 2002).

An important approximation for the study of adaptive shifts is to carry out comparisons at a geographical level, especially along latitudinal gradients, since mean annual environmental temperature decreases towards high latitudes. Many studies have used latitude as a natural source of variation, to study the physiological traits of many different organisms (Stillman and Somero, 2000; Gibert and Huey, 2001), and the strategies and mechanism that allow thermal tolerance (Addo-Bediako *et al.*, 2000; Rasmussen and Holmstrup, 2002). However, latitudinal variation in traits closely related to fitness, such as sprint speed (van Berkum, 1988), growth rate (Lonsdale and Levinton, 1985) and mortality rate (Wilson, 1991), have received scant attention.

Studies in evolutionary ecological physiology usually emphasize the analysis of species as a unit. Despite its importance, they pay little attention to physiological variability within a population or between populations of the same species inhabiting different habitats within a region or within individuals through time. In addition, most studies have not controlled for the phylogenetic relationships among species, which may lead to misleading conclusions about adaptation (Garland and Adolph, 1994).

It is broadly accepted that to evaluate evolutionary changes, one must compare populations within species (intraspecific, population level) as well as individuals within breeding populations. In summary, little attention has been paid to interpopulation variation despite its potential use as an approach to study changes in performance between local environments (Angilletta *et al.*, 2002).

Since arthropods inhabit vastly different environments and have a wide range of distribution, they are good models for studying the effect of temperature on physiological performance (Willott, 1997; Sinclair *et al.*, 2003). An especially interesting group are the terrestrial isopods, because they are one of the taxa least adapted to warm conditions (Warburg, 1987, 1993; Carefoot, 1993). We hypothesize that terrestrial isopods should present physiological and behavioural variation in their thermal physiology following exposure to low or high ambient temperatures.

We predict that these organisms will exhibit thermal physiological responses that are closely matched to their habitat's conditions – that is, local adaptation to thermal climate. Hence, in low-latitude populations thermal tolerances should be shifted to better correspond with their warmer environment (i.e. the curve should be shifted to the right) compared with high-latitude populations, where the curve should be shifted to the left.

We used the terrestrial isopod *Porcellio laevis* (Crustacea: Oniscidea) as a study model. This species is widely distributed in Chile, as well as elsewhere around the world (Leistikow and Wägele, 1999). The thermal physiology of *P. laevis* has been poorly described and there are few existing studies about lethal temperatures and genetic polymorphisms for this species (Edney, 1964; McCluskey *et al.*, 1993). The aim of this study was to determine the thermal physiology of this species to provide insight into adaptations to local thermal conditions.

MATERIALS AND METHODS

Animals, study site and maintenance

We collected *P. laevis* individuals from three populations in north and central Chile, spanning a latitudinal range of nearly 10°. The localities were: Antofagasta (23°38'S), La Serena (29°55'S) and Santiago (33°23'S) (Table 1). All specimens collected were placed in plastic containers and transferred to the laboratory in the Department of Ecology of the Pontificia Universidad Católica de Chile.

To remove possible confounding effects of sex we used only males. Males from all three populations were maintained under the same conditions – that is, acclimated at $21 \pm 1^\circ\text{C}$;

Table 1. Summary description of climatological variables for the three localities studied

	Antofagasta	La Serena	Santiago	Source
Latitude	23°38'S	29°55'S	33°23'S	
Longitude	70°26'W	71°15'W	70°42'W	
Altitude (m)	Sea level	Sea level	420	FAO (1985)
Mean annual temperature (°C)	17.0	14.9	13.9	FAO (1985); di Castri and Hajek (1976)
Mean minimum annual temperature (°C)	13.3	11.2	7.7	FAO (1985); di Castri and Hajek (1976)
Mean maximum annual temperature (°C)	20.1	18.9	22.1	FAO (1985); di Castri and Hajek (1976)
Total rainfall (mm)	2.2	127.4	356.2	FAO (1985); di Castri and Hajek (1976)
Total radiation (cal · cm ⁻² · day ⁻¹)	423	340	310	FAO (1985); di Castri and Hajek (1976)
Relative humidity (%)	72	80	75	FAO (1985); di Castri and Hajek (1976)
Climate	Desert	Semi-arid	Mediterranean	di Castri and Hajek (1976)

photoperiod 14 : 10 light : dark; and fed *ad libitum* with dry spinach. They were kept in plastic cages with a wet layer of plaster of Paris covering the bottom of each cage. Plaster of Paris is an inert material that preserves humidity and also provides a source of calcium carbonate, which is needed for exoskeleton synthesis.

Field body temperature measurements

To test for local adaptation in body temperature (T_b), we measured individual T_b and ambient temperature (T_a) for each locality during the austral summer of 2003. We manipulated the woodlice using latex gloves for a period of 5 s or less; during this time we inserted the thermocouple into the body of each individual. Ambient temperature was simultaneously measured with a thermocouple placed 1 cm over the site occupied by the isopod. Both body temperature and ambient temperature were measured using thin Cu-Constantan thermocouples and Cole-Parmer digital thermometers.

We calculated the differential temperature between T_b and T_a ($dT = T_b - T_a$). This index indicates the increase of T_b over T_a , as a measurement of thermoregulatory capacity.

Thermal sensitivity and thermal tolerances

We used righting response speed or rollover speed (i.e. the speed for an individual to change from an inverse position to an upright position) as a measure of the thermal sensitivity to different temperatures. Rollover speed was measured between 0° and 41°C. Specifically, we measured this trait every 1°C in the extremes of each population's tolerance range (0° to 10°C and 36° to 41°C), and every 2°C inside the range 12° to 34°C.

Rollover speed was considered as a measure of performance (Lutterschmidt and Hutchison, 1997). We estimated the physiological performance in each population, and then compared several indices of this performance among populations (i.e. optimum temperature, breadth temperature and maximum rollover speed). At extreme temperatures, some individuals had performance equal to zero. Those woodlice that exhibited a complete loss of righting response under extreme temperatures were considered to be in thermal coma (Lutterschmidt and Hutchison, 1997). This measurement is non-lethal and is useful for a large sample size (Huey *et al.*, 1992).

To determine rollover speed, we used the following experimental protocol. At each temperature treatment (0° to 41°C), 25 individuals were placed in a plastic chamber (100 × 100 × 20 mm) with 25 subdivisions (20 × 20 × 20 mm) to separate individuals. The plastic chamber was then placed inside an incubator and maintained at the experimental temperature (held constant ($\pm 0.5^\circ\text{C}$) using a water bath). After 30 min, we evaluated rollover speed for 10 min (i.e. a measure of righting response). If an individual was unable to right itself after this time, it was considered to be in thermal coma. Tolerance and performance curves were population measures, thus each woodlouse was used for only one experimental temperature. Animals that died during this measurement were excluded from the experiment. Body mass (m_b) was recorded before all measurements using an analytical balance (CHYO JK-180, with precision ± 0.01 mg).

After obtaining rollover speeds, we calculated the means for each experimental temperature. We observed that the means of each population followed a normal distribution along a wide range of temperatures (see Results). To calculate the optimum temperature for

performance of each population, we fitted this mean to a Gaussian function. From this fit, we obtained the populational optimum temperature ($T_{\text{opt}} \pm 1$ standard error). Using the mean values of rollover speed for each temperature, we calculated breadth of temperature (T_{br}) for each population (Gilchrist, 1996):

$$T_{\text{br}} = \sqrt{\sum \left[\frac{u_i(T_i - T_{\text{opt}})^2}{u_{\text{max}}} \right]}$$

where u_i is mean rollover speed at temperature T_i , and u_{max} is the maximum rollover speed for each population.

Thermal preference in the laboratory

We built a thermal gradient in the laboratory using a $500 \times 300 \times 3$ mm metal plate, covered with a 4 mm layer of plaster of Paris. The plate was divided into three separate runways (each 480 mm long and 90 mm wide) using opaque walls 80 mm high. The corners of the runways were rounded to avoid any corner effect (Warburg, 1993). To establish the thermal gradient, we heated one end of the metal plate with three infrared bulbs (250 W) and cooled the other end with a chilled water bath (-5°C) directly below the floor. The surface temperature in the gradient ranged from 5° to 35°C and did not change during the course of each trial. We placed a cold light source at the cold end to avoid the formation of a luminous gradient.

The experiment was performed at night, since these organisms are nocturnal. The thermal gradient was established for 30 min before the trials. Woodlice were removed from their cages, weighed in an analytical balance (CHYO JK-180, with precision ± 0.01 mg) and placed individually in separate runways at a random position within the thermal gradient. Then, individuals were allowed to acclimate for 30 min before we began testing. At the onset of the experiment we recorded T_b (initial T_{pref}) using an infrared thermometer with precision of $\pm 0.5^\circ\text{C}$ (TempTest IR, Oakton®). Then, we recorded T_b every 30 min for both sets of experiments (i.e. middle T_{pref} and final T_{pref} , respectively). At the end of the experiments woodlice were returned to their cages.

Statistical analysis

For all cases we tested for a correlation between body mass (m_b) and the thermal traits. Whenever this correlation was significant, we incorporated m_b as a covariate in an analysis of covariance (ANCOVA). To test for differences in field T_b , we performed a linear correlation between T_b and T_a , and a linear regression between T_b and T_a and between the range of field body temperature (T_f) and maximum T_f . Also, to determine differences in dT among populations, we performed a mixed-model two-way analysis of variance (ANOVA). Locality was designated as a random factor (since we selected three populations from an infinite number of localities along a latitudinal gradient), and time of day was designated as a fixed factor (only two possible levels: day and night). Interaction between the two factors was considered random. Performance and thermal tolerance were analysed using a variety of methods. First, we used an ANOVA to test for differences in u_{max} and heat coma among populations (random factor). Furthermore, we performed a correlation between T_{opt} and

latitude and between T_{opt} and mean annual temperature for each locality. To compare T_{opt} among populations, we computed t -tests, with populational means and standard errors obtained from the fits to the Gaussian function, for all three populations (see above). To correct for type I errors, we performed sequential Bonferroni corrections (Rice, 1989). To calculate the populational T_{br} , we utilized equation (1); then, we examined the correlation between this index and latitude, mean annual temperature and u_{max} separately. Finally, to evaluate differences in chill-coma temperature, we used a separate-slopes ANCOVA model with locality as the random factor and m_b as a covariate (Statistica 6.0). To evaluate differences in T_{pref} among localities, we performed a repeated-measures ANOVA, where the repeated measure was the T_{pref} of an individual during the three experiments. To compare the field body temperature and laboratory body temperature to each population (T_f and T_{pref} final, respectively), we used a t -test with sequential Bonferroni corrections. Before all analyses, we tested the data for normality and homoscedasticity (Kolmogorov-Smirnov and Cochran C tests, respectively). When necessary to meet assumptions, the data were \log_{10} -transformed. When differences in the means were significant at the $P < 0.05$ level, they were also tested with an *a posteriori* Tukey test (HSD). All statistical analyses were conducted using Statistica 6.0™ software (Statsoft Inc., Tulsa, OK).

RESULTS

Field body temperature

Body temperature was strongly and positively correlated with T_a ($\beta = 0.901 \pm 0.02$, $t_{500} = 46.33$, $P < 0.0001$), and T_a explained 81% of the variation in T_b ($r^2 = 0.81$, $F_{1,500} = 2146.3$, $P < 0.0001$). In general, T_b was higher than T_a in this species (Fig. 1). We observed that T_f increased with latitude in *P. laevis* (Table 2). The T_f range (i.e. maximum minus minimum T_f recorded in the field) also increased towards high latitudes. This increment in the range was positively correlated with maximum T_f ($r = 0.997$, $P < 0.05$).

We found that dT was highest in Antofagasta and lowest in La Serena (Table 3). At night, dT was higher than during the day (Fig. 2). However, both day and night dT varied along

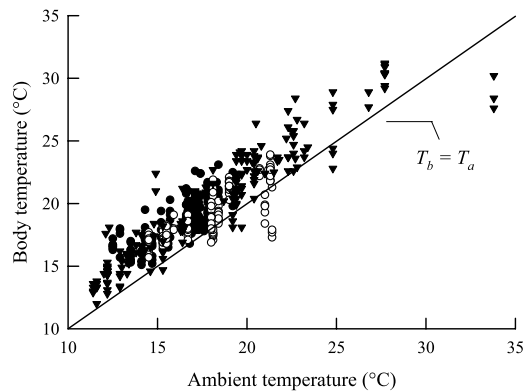


Fig. 1. Relationship between body temperature (°C) and ambient temperature (°C) for each population of *P. laevis*. The solid line indicates a slope equal to 1 ($T_b = T_a$). ●, Antofagasta; ○, La Serena; ▼, Santiago.

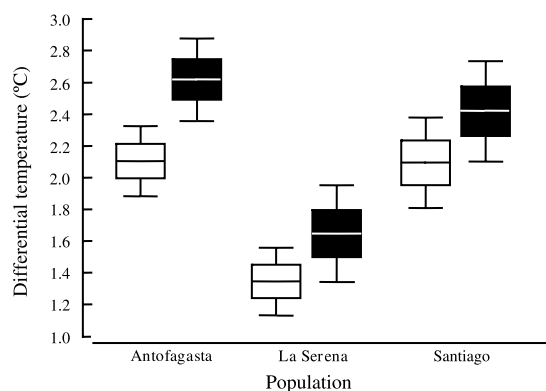
Table 2. Distribution of field body temperature (T_f) of woodlice populations

Population	Latitude	Mean (°C)	SE (°C)	N	Minimum T_f (°C)	Maximum T_f (°C)	Range T_f (°C)
Antofagasta	23°38'S	18.7	0.13	180	15.1	23.1	8
La Serena	29°55'S	19.1	0.16	146	15.7	23.9	8.2
Santiago	33°23'S	20.4	0.36	176	12	31.2	19.2

Table 3. Results for morphological, physiological and behavioural traits for *Porcellio laevis* populations from Antofagasta, La Serena and Santiago (mean \pm standard error)

Traits	Antofagasta 23°38'S	La Serena 29°55'S	Santiago 33°23'S
Body mass (mg)	79.50 \pm 1.51	102.10 \pm 1.52	87.20 \pm 1.53
T_{opt} (°C)	24.69 \pm 0.54	20.94 \pm 0.22	21.12 \pm 0.65
T_{br} (°C)	8.75	11.73	12.27
u_{max} (s ⁻¹)	2.22 \pm 0.28	1.18 \pm 0.17	2.04 \pm 0.30
Initial T_{pref} (°C)	11.07 \pm 1.09	12.58 \pm 1.09	9.40 \pm 1.11
Middle T_{pref} (°C)	12.72 \pm 1.31	14.27 \pm 1.21	13.45 \pm 1.34
Final T_{pref} (°C)	14.68 \pm 1.12	16.72 \pm 1.12	12.02 \pm 1.14
T_f (°C)	18.70 \pm 0.13	19.10 \pm 0.16	20.40 \pm 0.36
dT (°C)	2.34 \pm 0.11	1.29 \pm 0.12	2.11 \pm 0.11

Note: Traits are abbreviated as follows: optimum temperature (T_{opt}), temperature breadth (T_{br}), maximum rollover speed (u_{max}), preferred temperature (T_{pref}), field body temperature (T_f), temperature differential (dT = body temperature minus ambient temperature).

**Fig. 2.** Temperature differential ($dT = T_b - T_a$, °C) in the field for three sites arranged along a latitudinal gradient. Open boxes represent the daytime differential and the solid boxes indicate the night-time differential. Middle points represent the mean value, the box indicates ± 1 standard error, and the whiskers represent the mean $\pm 95\%$ confidence interval.

the latitudinal gradient, and was marginally significant among populations (Table 4). Antofagasta was the only population with statistically different dT between day and night, when compared with the other populations (*a posteriori* Tukey's HSD test).

Thermal sensitivity and thermal tolerances

Rollover speed tended to increase with temperature in all populations, with maximum performance being reached at intermediate temperatures and then decreasing at higher temperatures (Fig. 3). Maximum rollover speed (u_{\max}) was highest in Antofagasta and lowest in La Serena (Table 3) with significant differences (one-way ANOVA: $F_{2,70} = 4.98$, $P < 0.01$). In each population, u_{\max} was attained at different optimum temperatures (T_{opt}); again, the highest values were observed in Antofagasta and the lowest in La Serena (Table 3). We found significant differences between Antofagasta and La Serena (*a posteriori* Tukey's HSD test). Differences in T_{opt} between populations were statistically significant for comparisons of Antofagasta versus La Serena ($t_{56} = 8.37$, $P < 0.0001$), and Antofagasta versus Santiago ($t_{56} = -16.07$, $P < 0.0001$). The comparison between La Serena and Santiago was not significantly different ($t_{56} = 0.42$, $P = 0.68$). Populational T_{br} increased towards high latitudes (Table 3). This index is positively correlated with latitude and negatively correlated with mean annual temperature, but both tendencies were not significant ($r = 0.97$, $P = 0.15$ and $r = -0.98$, $P = 0.11$, respectively).

Table 4. Results of the mixed-model two-way ANOVA testing for the effects of locality (random factor) and time of day (fixed factor) on thermal differential, dT ($^{\circ}\text{C}$)

Factor	SS	d.f.	MS	<i>F</i>	<i>P</i> -value
Locality	57.14	2	28.57	19.98	<0.001**
Time of day	13.01	1	13.01	13.99	0.06
Locality \times time of day	1.85	2	0.93	0.65	0.52
Error	693.03	486	1.43		

*** $P < 0.001$; * $P < 0.05$.

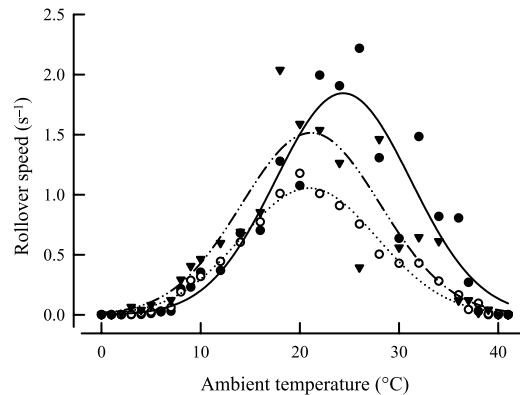


Fig. 3. Thermal-sensitivity functions for rollover speed (s^{-1}) in the three studied populations arranged along a latitudinal gradient. See text for details. ●, Antofagasta; ○, La Serena; ▼, Santiago.

The critical thermal minimum (CT_m), represented by the chill-coma temperature (T_{cc}), was significantly different among populations (Table 5). There was latitudinal variation in T_{cc} ($r = -0.997$, $P = 0.05$). An *a posteriori* Tukey's HSD test revealed significant differences between all populations (Fig. 4). There were no significant differences observed in the critical thermal maximum (CT_M), represented by the heat-coma temperature (T_{hc}), among populations (one-way ANOVA: $F_{2,236} = 1.48$, $P = 0.23$).

Laboratory thermal preferences

T_{pref} increased among experimental replicates; however, Santiago was the only population that exhibited a lower final T_{pref} than middle T_{pref} (Table 3). We found that T_{pref} varied between measurements, and among populations (Table 6). Comparisons among populations showed a mean T_{pref} higher in La Serena than Antofagasta and Santiago.

We also compared T_{pref} between individuals in the laboratory and in the field. We compared final T_{pref} in the laboratory with nocturnal T_b in the field (T_f) within each population. We found significant differences between T_{pref} and T_f for populations from Antofagasta and Santiago ($t_{118} = 4.77$, $P < 0.001$ and $t_{98} = 7.15$, $P < 0.001$, respectively), but not in La Serena where T_f was higher than T_{pref} ($t_{67} = 0.81$, $P = 0.42$; see Table 2).

Table 5. Results of the separate slopes model ANCOVA testing for the effect of locality on chill-coma temperature (°C), with body mass (m_b) as a covariate

Factor	SS	d.f.	MS	F	P-value
Locality	7.20	2	3.6	7.83	<0.001***
Locality $\times m_b$	8.52	3	2.84	6.17	<0.001***
Error	149.41	349	0.46		

*** $P < 0.001$; * $P < 0.05$.

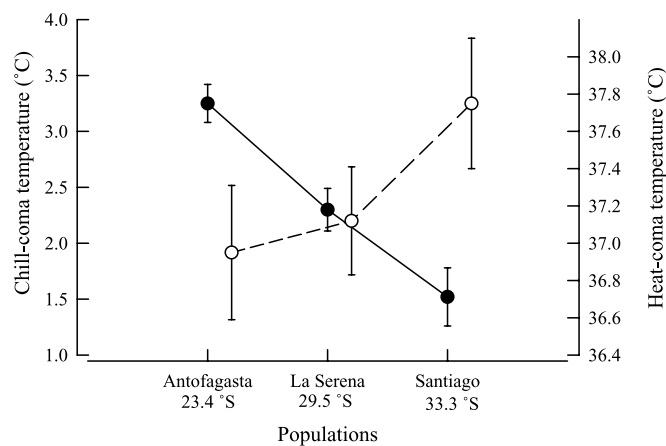


Fig. 4. Chill-coma temperature (●) and heat-coma temperature (○) of the three studied populations arranged along a latitudinal gradient. Bars represent \pm standard error. See text for details.

Table 6. Results of the repeated-measures ANOVA testing for the effect of locality on preferred temperature, T_{pref} , along the three measures over time

Factor	SS	d.f.	MS	<i>F</i>	<i>P</i> -value
Locality	458.59	2	229.3	4.97	<0.01**
Error	3967.44	86	46.13		
T_{pref}	492.74	1	492.74	18.22	<0.001***
Locality \times T_{pref}	17.49	2	8.75	0.32	0.72
Error	2325.86	86	27.04		

*** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$.

DISCUSSION

How population differentiation affects thermal physiological ecology has been widely studied in different groups of ectotherms (see Parsons, 1993; Forsman, 1999; Angilletta *et al.*, 2002; David *et al.*, 2003). In this study, we documented intraspecific differences in organismal traits among populations of the terrestrial isopod, *P. laevis*, separated by 10° of latitude. We found differences in thermal performance between populations both in the field and under laboratory conditions. This may indicate that thermal traits have diverged in response to local selective pressures (Johnston and Bennett, 1996).

Field T_b measurements

Porcellio laevis showed a low capacity for thermoregulation in the field, since T_b was observed to be highly dependent on T_a (Hertz *et al.*, 1993). This was the case for almost all temperatures among the populations studied, with the exception of the highest T_a (see Fig. 1). Probably, behavioural mechanisms permitted isopods to avoid high temperatures; we also observed that T_b increased its mean, variation and range with latitude. These results support the classical latitudinal hypothesis, which predicts that low-latitude ectotherms should be adapted to a narrower range of temperatures, whereas mid-latitude ectotherms should be adapted to a broader range of temperatures (van Berkum, 1988). In addition, this pattern is supported by our climatological data, which show a higher temperature oscillation at high latitudes than low latitudes (see Table 1). When T_b was corrected for T_a , there was no consistent change with latitude (or mean annual temperature). This lack of pattern can be explained by differences in the use of thermal patches in animals' microhabitats (Melville and Schulte, 2001), or by different thermoregulatory strategies that are not associated with thermal factors in the long term. However, we observed differences in dT between populations (when T_b was corrected for by T_a), which may imply the existence of different thermoregulatory mechanisms in each population. Soil-dwelling organisms (i.e. cryptozoic fauna), such as terrestrial isopods, inhabit terrestrial habitats with marked variations in abiotic conditions. Microclimatic conditions are defined as the specific environment experienced by an animal on the immediate spatial and temporal scale. Here we observed a difference of about 2°C in T_b (dT) between day and night. These differences may be due to the nocturnal activity of terrestrial isopods (Warburg *et al.*, 1984), which also may increase their metabolic rate. The foraging behaviour and digestive processes associated with feeding result in a significant increase in metabolism. This process has been studied in

individuals from a variety of taxonomic groups, including vertebrates, and appears to be quite a general phenomenon (McEvoy, 1984; Cortés *et al.*, 1994; Spicer and Gaston, 1999; Kearney and Predavec, 2000; Willmer *et al.*, 2000; Whiteley *et al.*, 2001).

An alternative hypothesis for the observed difference in temperature between day and night is that nocturnal temperatures have been behaviourally selected for in microhabitats where isopods are forced to perform some physiological processes (such as digestion or reproduction) that generally require warm temperatures (Steward, 1981). There is a significant lack of information regarding day–night patterns in the T_b of nocturnal invertebrates, and this paper is the first report on terrestrial isopods.

Thermal sensitivity

The performance function varied significantly between populations. We observed that maximum rollover speeds were reached under different thermal optima. T_{opt} decreased with latitude. Hence, we can attribute the variation in T_{opt} to local thermal conditions, but this factor does not explain all of the variation we observed for this trait. Furthermore, very little is known about variation in T_{opt} on a latitudinal basis. Wilson (2001) reported that cool-temperate populations of the striped marsh frog showed a lower T_{opt} than tropical populations. Other key parameters to characterize the thermal performance are T_{br} and u_{max} . These parameters can be measured at populational or individual levels and these could differ. Population performance could be determined by polymorphism in thermal traits, whereas individual performance could be influenced by genetic factors or phenotypic plasticity. We found that these indices, at the population level, increased and decreased with latitude, respectively. The correlation between these parameters will help us to determine potential trade-offs in population performance curves. This trade-off has been interpreted as an increment in maximum velocities and a reduction in T_{br} , also known as the ‘jack-of-all-trades is a master of none’ hypothesis (Huey and Hertz, 1984; Huey and Kingsolver, 1989). We observed a negative correlation between population T_{br} and population u_{max} , but it was not significant ($r = -0.52$, $P = 0.65$). Thus, our results appear to support this hypothesis but more work is needed to confirm it for this species. A potential drawback of the study is that we included few populations ($n = 3$), which reduced the statistical power of our tests.

Thermal tolerances

Thermal tolerances varied with latitude only in the case of T_{cc} . The chill-coma temperature provides a good index of activity at low temperatures, and our results show that the measure of minimum critical temperature, CT_m , decreases towards high latitudes. Hence, populations that experience low mean annual temperature in their habitats show a high tolerance for low temperatures (Table 1). Moreover, these traits exhibit clinal variation. This result is in line with the study of Gibert and Huey (2001), who found that T_{cc} was inversely related to latitude in *Drosophila subobscura*. Low-latitude populations showed less thermal tolerance than high-latitude populations. This suggests a potential adaptation to cold temperatures towards high latitudes.

The heat-coma temperature did not vary between populations. Heat tolerance in *P. laevis* showed a rigid response along the latitudinal gradient studied. This agrees with Edney (1964), who reported that the upper lethal limit in *P. laevis* of California, North America,

did not differ between populations. A similar trend was reported in the lizard *Eleutherodactylus* sp., which did not exhibit differences along an altitudinal gradient (Christian *et al.*, 1988). On the other hand, in *Littorina* species, researchers have observed extensive variation in T_{hc} between populations and species (Clarke *et al.*, 2000; Sokolova and Pörtner, 2003). A macro-scale study proposed that geographical variation at the upper limits of an organism's temperature tolerance range is less variable than the lower limit (Addo-Bediako *et al.*, 2000). This rigidity has two possible explanations: (1) a biochemical limitation stemming from the fact that proteins lose stability at high temperatures and may be denatured (Tomanek and Somero, 2002); and (2) small terrestrial arthropods such as *P. laevis* live under stones, pieces of wood and soil litter, where ambient temperatures are not higher than T_{hc} (see Table 2) across populations (Sutton and Holdich, 1984). For this reason, woodlice probably do not need differential adaptations to high temperatures between populations.

Thermal preferences in the laboratory

In the laboratory study, we observed significant temporal variation in T_{pref} within each population. T_{pref} increased sequentially, which indicates that woodlice tended to prefer progressively higher temperatures. This demonstrates how it can be misleading to use only one record of T_b to infer T_{pref} . It appears that individuals tend to shift among thermal patches for a period of time before selecting the final T_{pref} (i.e. 'final preferendum'). Final temperature preferendum is defined as the temperature around which all individuals will ultimately congregate regardless of their thermal experience before being placed in the gradient. Thus, it is a process that is not influenced by acclimation factors (Reynolds and Casterlin, 1979). Our results for T_{pref} are different from those reported for other species from the same family, such as *Armadillidium vulgare*, whose T_{pref} is around 18–21°C (Refinetti, 1984). On a geographical scale, we did not observe a particular pattern of T_{pref} along the latitudinal gradient. T_{pref} decreased with latitude, indicating that populations from high latitudes preferred low temperatures in the laboratory. However, we cannot support our hypothesis of a clinal variation in these traits.

Comparisons between T_{pref} and T_f were consistent and highly significant: T_f was always higher than T_{pref} . These differences may be generated by laboratory conditions, where animals are not experiencing the environmental constraints that influence thermoregulation in the field (Hertz *et al.*, 1993). This limitation evokes the popular trade-off of precision versus realism: laboratory settings provide precision and an exact response to a specific controlled effect but may not apply in field conditions. However, there are many questions that are virtually impossible to address without laboratory experiments.

The intraspecific differences that we observed among populations could result from different developmental conditions due to local conditions or from genetic differentiation. These differences in thermal traits could be the end-product of local adaptation to micro-habitats. According to Olsson and Uller (2003), local adaptation in thermal limits can be achieved by selection on survival at low temperatures. Therefore, in terrestrial isopods, the minimum temperature during winter is probably a more important limitation to distribution than the maximum temperature during summer. This idea agrees with studies performed in the woodlouse, *Hemilepistus reaumuri*, of North Africa (see Coenen-Stass, 1984).

In summary, we found that populations of a terrestrial isopod have diverged in their thermal traits in accordance with their micro-environmental conditions. We observed

that physiological and behavioural differences persist following laboratory acclimation. Consequently, we conclude that differences in thermal physiology are the consequence of local adaptation rather than phenotypic plasticity. Thus, southern woodlice (i.e. the Santiago population) appear to have evolved towards low temperature tolerance, whereas northern woodlice (i.e. the Antofagasta population) appear to have a higher optimum temperature.

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