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Bergmann's rule in the ant lion *Myrmeleon immaculatus* DeGeer (Neuroptera: Myrmeleontidae): geographic variation in body size and heterozygosity

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Abstract

Aim Geographic variation in body size and heterozygosity were surveyed for discrete populations of the ant lion, *Myrmeleon immaculatus* DeGeer, collected from the central and northeastern United States.

Location Collection sites were located in the central and eastern United States ranging from western Oklahoma to northern New York.

Methods We collected 872 *M. immaculatus* larvae from thirty-four collecting sites. At each site, we randomly sampled ant lion pits and collected between fifteen and fifty-two larvae in total. Larvae were preserved in 95% ETOH for morphological analysis and frozen in a −80°C freezer for protein electrophoresis. We measured the body size of eighty-five preserved adult *M. immaculatus* obtained from museum collections using head width as an indicator of body size. Five enzymes [GPI (glucose phosphate isomerase), MDH (malate dehydrogenase), PEP (peptidase), DIA (diaphorase) and SOD (superoxide dismutase)] were used in the heterozygosity analyses.

Results Larval and adult body size increased with latitude, but decreased with elevation. Average heterozygosity, measured at five polymorphic loci, also increased significantly with latitude. Minimum temperature variance was the best predictor of body size, whereas precipitation and maximum temperature were the best predictors of heterozygosity. Populations were genetically differentiated from one another and showed a pattern of isolation by distance, as measured by Wright's F_{st} values and Nei's genetic distances.

Main conclusions Sampling artifacts, heat conservation, character displacement, cell-size variation, density-dependent mortality, and differential dispersal probably cannot account for latitudinal variation in ant lion body size. Our results implicate the importance of diurnal photoperiod, which varies with latitude, but not with elevation. Because photoperiod often controls growth, diapause, and metamorphosis, it may be an important determinant of latitudinal clines in body size and life history of insects.

Keywords
Ant lion, Bergmann's rule, body size, elevation, heterozygosity, latitude, photoperiod.

INTRODUCTION

Bergmann's rule, an intraspecific increase in body size towards higher latitudes, is a common geographic pattern in homeotherms (Brown & Lee, 1969; Brodie, 1975; Forsman, 1991; Scharples, Fa & Bell, 1996; but see Zink & Remsen, 1986 and Geist, 1987). In ectotherms, Bergmann's rule has been documented among species or families of insects (Cushmann,

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Lawton & Manly, 1993; Hawkins & Lawton, 1995). However, these patterns are difficult to interpret because species may not be statistically independent of one another (Felsenstein, 1985), and there may be confounding influences of speciation history (Taylor & Gotelli, 1994) and geographical range size (Brown, 1995). Masaki's (1967, 1978) detailed studies of intraspecific variation in cricket body size document the opposite pattern—body size was greatest at low latitudes and at low elevations. Traits often vary in the same way with increasing latitude and increasing elevation, presumably because of similar climatic trends (Rosenzweig, 1995).

Hypotheses to account for Bergmann's rule include character displacement (McNab, 1971), clines in prey body size (McNab, 1971; Forsman, 1991), starvation resistance (Kondoh, 1977; Lindstedt & Boyce, 1985), seasonality (Murphy, 1985), voltinism (Roff, 1992; Bradford & Roff, 1995), and genotypic and phenotypic responses to temperature, humidity, and photoperiod (Masaki, 1967; Schoener & Janzen, 1968; James, 1970; Furunishi & Masaki, 1983; Levinton & Monahan, 1983; Conover & Present, 1990). Characteristics of populations, such as density and habitat patchiness, may also influence clines in body size (Iba, Nagao & Urano, 1995; Brunkow & Collins, 1996; Hutchings, 1996).

Finally, Bergmann's rule may arise through life history correlates of adult mortality (Stearns & Koella, 1986; Roff, 1992) and egg size (Berven, 1982). Genetic variation has been correlated with variation in life history traits (Ledig, Guries & Bonefield, 1983; Mitton & Grant, 1984; Koehn, Diehl & Scott, 1988; Strauss, 1991), and may be necessary for individuals to survive heterogeneous environments at high latitudes (Hedrick, Ginevan & Ewing, 1976; Liu, Soong & Chen, 1995).

To distinguish among these mechanisms, it is necessary to first describe the broad climatic, as opposed to strictly geographic, correlates of body size (Power, 1969). In addition, it is necessary to understand the genetic structure of populations, and the extent to which local populations are genetically distinct from one another (James, 1982; Graves, 1991).

In this paper, we document a pattern of increasing body size and increasing heterozygosity with latitude in the ant lion, *Myrmeleon immaculatus* DeGeer (Neuroptera: Myrmeleontidae). Our results suggest that heterozygosity and body size can be predicted on the basis of climate and annual climatic variance, and that local populations are genetically distinct from one another. Because body size also decreases with elevation, our results implicate the importance of photoperiod as a mechanism generating geographic clines in insect body size.

METHODS

Ant lion life history

Myrmeleon immaculatus is commonly found in large populations throughout the southern and eastern Unites States (Wheeler, 1930). The larvae are sit-and-wait predators that dig pits for prey capture. They live in sandy, sheltered habitat and occur in discrete populations. *M. immaculatus* has three larval instars, which are distinguished on the basis of head-capsule width and pigmentation patterns (Stange, 1980). Larvae live up to 2 years, construct silk cocoons, and emerge as adults after approximately 1 month (Furunishi & Masaki, 1981, 1982). Adults are weak fliers and live for approximately 1 month (Arnett, unpub. data).

Larval body size

To address the question of latitudinal variation in body size, we collected 872 *M. immaculatus* larvae from thirty-four collecting sites ranging from Oklahoma to New York. We began collecting in Oklahoma in mid-June, 1994, and spent 1–2 days at each site. We visited sheltered sites with dry soil (overhangs, cliff ledges, old barns) that were likely to support ant lion populations (Gotelli, 1993). At each site, we randomly sampled ant lion pits and collected between fifteen and fifty-two larvae in total. Depending on the density and patchiness of the population, we collected specimens by walking a path of 0.1 to 1.0 km. Larvae were preserved in 95% ETOH for morphological analysis. For each preserved larva, we measured maximum head width, a common indicator of body size in insects (Lutz, 1974; Hawking & New, 1996), with a micrometer to the nearest 0.01 mm.

Adult body size

Does adult body size vary with latitude? To address this question, we measured body size of eighty-five preserved adult *M. immaculatus* obtained from museum collections (University of Kansas, University of Georgia, Academy of Natural Sciences in Philadelphia, Illinois Natural History Survey, Florida Department of Agriculture, and the Museum of Comparative Zoology at Harvard). The geographic extent of these collections was from Michigan and New Hampshire in the north to Oklahoma and Florida in the south. The specimens were originally collected by a variety of researchers between 1877 and 1938. Species identifications were confirmed by the criteria in Banks (1927). Sexes were determined by the presence of oviposition setae on females and the pilula axillaris at the base of the hind wing on males (Stange, 1980). For each museum specimen, the length of the front wings was measured with calipers to the 0.1 mm. We also measured distance between the eyes, a common indicator of insect body size (Hogue & Hawkins, 1991). Inter-eye distance was measured to the 0.01 mm with a micrometer.

Population density and patchiness

In order to account for potential differences in local population density and habitat patchiness on body size and heterozygosity, we returned to ten of our collection sites in 1995. At each site, four occupied larval pits were randomly chosen. We measured the first four nearest-neighbour distances for each of these pits. The average of the first nearest-neighbour distance ($N = 4$) was used as surrogate for population density.

Next, we plotted the average nearest-neighbour distance as a function of neighbour number (1,2,3,4). We used the slope of this regression as a measure of population patchiness. We reasoned that if individuals were distributed homogenously in a population, the slope of this line would be relatively shallow. If individuals were clumped in space, the slope would be much steeper (see also Lewontin & Levins, 1989).

Climatic and geographic variables

To examine correlations of body size, heterozygosity, and climate, we obtained maximum and minimum temperature (°C) and precipitation (mm) from the National Climatic Data Center (1992). Each variable represents a 30-year average of annual means (1961–1990) from the weather station closest to each population. Maximum and minimum temperature variance was obtained by calculating standard deviation and variance of the 30-year averages. Distance from weather stations to population sites ranged from approximately 5 to 100 km. Latitude and longitude (min) of each collection site were measured from detailed state maps. Latitudes ranged from 33°43′ N to 41°53′ N. Longitudes ranged from 71°43′ W to 97°05′ W. Elevation (m) was measured with USGS topographic maps, and ranged from 0 to 457.3 m above sea level.

Allozyme survey

Protein electrophoresis was used to measure genetic differentiation among *M. immaculatus* populations. During the summer of 1994, we collected a minimum of thirty third-instar larvae from each of seventeen collecting sites for genetic analyses. These larvae were mailed overnight in individual cryotubes to the University of Vermont, Burlington, where they were stored at −80°C. To ensure that we collected *M. immaculatus*, larvae were identified to species using the key of Lucas & Stange (1981).

To create extractions, we homogenized frozen larvae in chilled, distilled water. This homogenate was analysed using standard starch-gel electrophoretic techniques (May, 1991). Initially we surveyed twenty-four enzymes known to be polymorphic in related insect species (B. May, pers. comm.). Eight of these enzymes were polymorphic. Three of them resolved poorly: ME (malic enzyme), G_3P (glycerol-3-phosphate dehydrogenase), and MPI (mannose phosphate isomerase). The remaining five enzymes were used in the final analyses: GPI (glucose phosphate isomerase), MDH (malate dehydrogenase), PEP (peptidase), DIA (diaphorase) and SOD (superoxide dismutase). A citrate-morpholine buffer was used for GPI, MDH and DIA (May, 1991), and a boric acid-lithium hydroxide buffer was used for SOD and PEP (May, 1991). Electrophoresis was conducted in 12% starch gels. Staining procedures followed those in May (1991). Loci nomenclature followed the protocols suggested by Shaklee *et al.* (1990).

Statistical analysis

Larval body size analyses were restricted to those sites for which five or more third-instar larvae were collected ($N=22$; Fig. 1). Between 36 and 40°N latitude (Virginia through New Jersey) we found no populations with enough third-instar larvae for inclusion in analyses (Fig. 1). Body size measurements of adults and larvae were natural log-transformed prior to analysis. Relationships among body size, observed heterozygosity, five climatic variables, three geographic variables, and density and patchiness were analysed with simple correlations. Stepwise forward multiple regression was also performed using all variables, except density and patchiness, due to differences in sample size. For multiple regression analyses, we used $P<0.25$ to enter variables into the model, and $P<0.05$ to retain them. For each dependent variable (larval body size, adult body size, heterozygosity), we conducted two regression analyses: (1) using the five climatic variables (maximum and minimum temperature, precipitation, and maximum and minimum temperature variance) as predictors; and (2) using the three geographic variables (latitude, longitude, and elevation) as predictors.

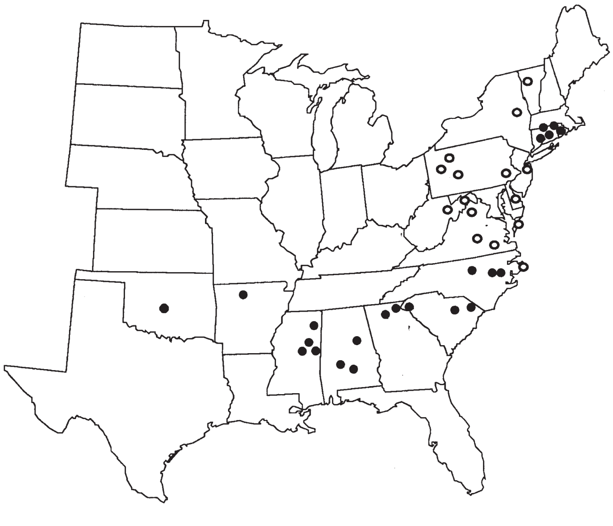

Figure 1 Locations of thirty-four *M. immaculatus* populations used for larval body size analyses. ●, Populations samples; ○, sites with no or few ant lions.

Genetic analysis

We calculated observed and expected heterozygosity for all five polymorphic loci for each population, then calculated average heterozygosity of all the populations (Hartl & Clark, 1989). We used Nei's genetic distance (Nei, 1972) as a measure of genetic distance between all possible pairs of populations. We used a Monte Carlo simulation of the Mantel test (Schnell, Watt & Douglas, 1985) to evaluate the correlation between genetic distance and geographic distance. Wright's *F*-statistics were calculated as measures of population differentiation: F_{is} (inbreeding of an individual relative to a subpopulation); F_{it} (inbreeding of an individual relative to the entire population); and F_{st} (inbreeding of a subpopulation relative to the entire population). Heterozygosity, Wright's F_{st}, and Nei's genetic distances were calculated using the computer program Genes

Table 1 Simple correlation coefficients (r) for larval body size and observed heterozygosity with all predictor variables. (NS $= P > 0.05$; * $= P < 0.05$; ** $= P < 0.01$; *** $= P < 0.001$).

	Body size	Heterozygosity
Latitude	0.46*	0.84***
Longitude	−0.44*	−0.80***
Elevation	−0.46*	NS
Precipitation	−0.43*	−0.59**
Maximum temperature	−0.43*	−0.82***
Minimum temperature	NS	−0.77***
Density	NS	NS
Patchiness	NS	0.87***
Heterozygosity	NS	
Maximum temperature variance	0.41*	0.64**
Minimum temperature variance	0.46*	0.77***

Table 2 Multiple regression coefficients for larval body size and observed heterozygosity with geographic and climatic variables as predictors. Regression coefficients are shown for the best-fitting models. P-values are given in parentheses. NS = non-significant regression coefficient ($P > 0.05$).

	Body size*	Heterozygosity†
Geographic		
Latitude	0.00178 ($P = 0.02$)	0.01104 ($P = 0.03$)
Longitude	NS	NS
Elevation	−0.00001 ($P = 0.04$)	NS
Climatic		
Max. temperature	NS	−0.01855 ($P = 0.05$)
Min. temperature	NS	NS
Precipitation	NS	−0.00308 ($P = 0.03$)
Max. temp. variance	NS	NS
Min. temp. variance	0.05269 ($P = 0.02$)	NS

* Body size, d.f. = 19.
† Geographic, d.f. = 14; climatic, d.f. = 12.

in Populations (Eng & Paul, 1992). Gene flow was calculated by Wright's Island Model (Slatkin & Barton, 1989). We wrote a program in Turbo Pascal 7.0 for the Mantel test. A G-test was used to assess the significance of Wright's F_{st} (Sokal & Rohlf, 1995).

RESULTS

Larval body size

In a simple correlation analysis, larval body size was positively correlated with latitude and minimum and maximum temperature variance. Body size was negatively correlated with longitude, elevation, precipitation, and maximum annual temperature. Minimum annual temperature, heterozygosity, and population patchiness and density were not significant predictors of larval body size (Table 1).

In the best-fitting regression model with geographic variables, latitude and elevation were the only significant predictors of larval body size (Table 2). As in the simple regression models, body size was positively correlated with latitude (Fig. 2a), but negatively correlated with elevation (Fig. 2b). There was no significant correlation between latitude and elevation of the collection sites ($r = 0.07$; $P = 0.74$). In a regression model with climatic variables, minimum temperature variance was the only predictor of larval body size (Table 2).

Adult body size

Adult females were significantly larger than adult males for both inter-eye distance (females [$n = 53$]: mean ± 1 SD = 3.7 ± 0.2; males [$n = 29$]: 3.5 ± 0.3; $F_{1,80} = 17.96$, $P < 0.0001$) and average front-wing length (females: mean ± 1 SD = 35.0 ± 2.0; males: 32.5 ± 2.1; $F_{1,80} = 20.92$, $P < 0.0001$). For this reason, we used sex as a categorical variable in our multiple regression models.

For the simple correlations, the only significant relationship was a negative correlation between female body size and elevation (Fig. 2d). The correlation between body size of both sexes and latitude was positive, but non-significant, in both the simple and the multiple regression models (Fig. 2c). None of the climatic variables were significantly correlated with adult body size in either simple or multiple regression models.

Heterozygosity

Observed heterozygosity was positively correlated with latitude (Fig. 3), population patchiness, and maximum and minimum temperature variance (Fig. 4a). Latitude and heterozygosity are still positively correlated when three northern sites are removed ($P = 0.01$). Heterozygosity was negatively correlated with longitude, precipitation, maximum temperature (Fig. 4b), and minimum temperature. There was no significant correlation of heterozygosity with elevation or density (Table 1). In multiple regression models, only latitude, maximum temperature, and precipitation were significantly correlated with heterozygosity (Table 2).

Genetic differentiation

Expected and observed heterozygosities of the seventeen *M. immaculatus* populations surveyed are shown in Table 3. Nei's genetic distances between sites were significantly associated with geographic distances between sites ($r = 0.58$; Mantel test $P < 0.0001$). This result is seen when all populations are included in the analysis, but isolation by distance is not seen for the southern sites alone.

F_{st} analysis revealed significant differentiation among all populations, as well as among populations within regions (Table 4). Approximately 25% of the total variation in allele frequencies was due to the genetic differences among populations (mean $F_{st} = 0.254$). High F_{it} values (inbreeding of individuals relative to the total) and high F_{is} values (inbreeding of individuals to the subpopulation) both indicate a greater number of homozygous individuals than expected. Genetic differentiation within a region was less than among regions, except for the Georgia populations, which had an F_{st} similar to the total (Table 4). Gene flow, estimated from F_{st} values,

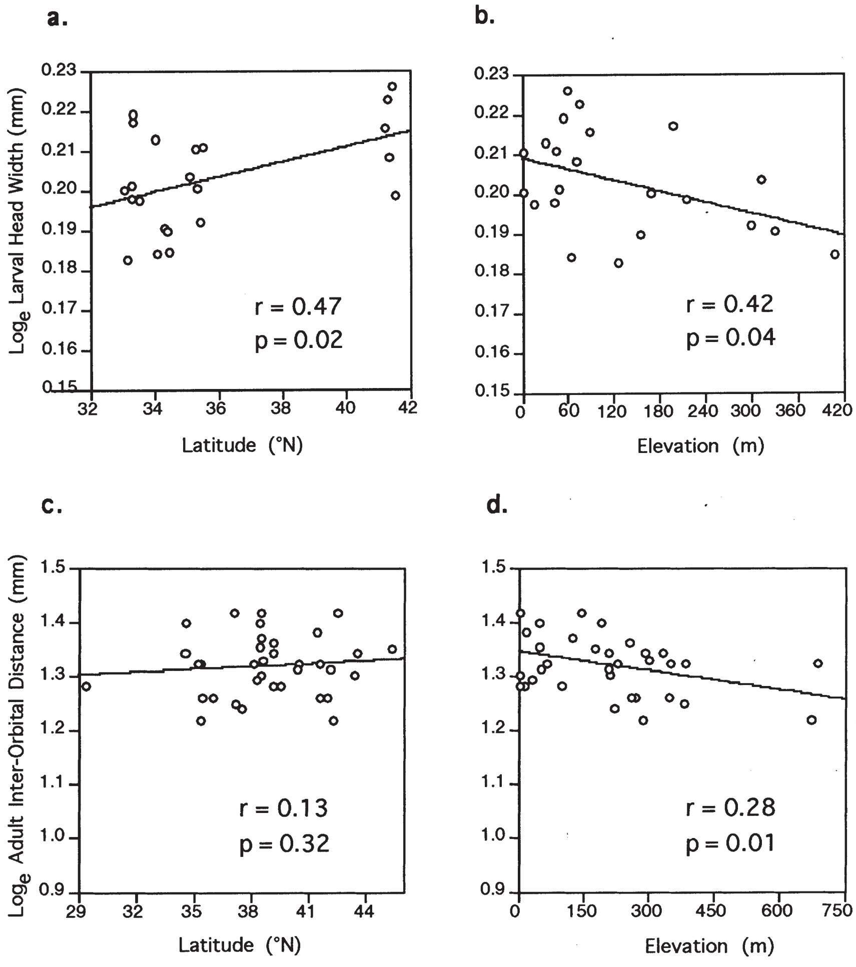

Figure 2 Average larval body size (a) increases with latitude and (b) decreases with elevation. Each point represents the average head-width of third-instar larvae from a population (*N*>5 larvae per population). Average adult body size, shown for females, (c) increases non-significantly with latitude, but (d) decreases significantly with elevation. Each point represents the inter-eye distance measured for a single adult female.

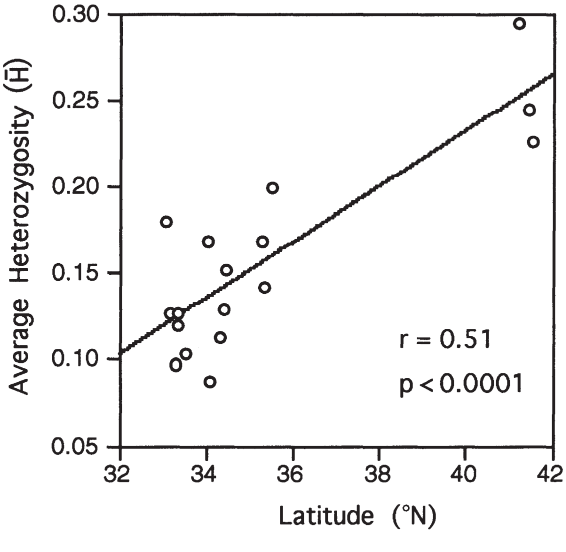

Figure 3 Average observed heterozygosity increases significantly with latitude ($N=17$).

was less than one migrant per generation among all populations, but varied from 0.74 to 8.4 migrants per generation among populations within a region (Table 4).

DISCUSSION

We found two consistent geographic patterns in ant lion body size: a weak positive correlation between body size and latitude (Fig. 2a,c) and a strong negative correlation between body size and elevation (Fig. 2b,d). The latitudinal correlation for adults is weak (both sexes: $P=0.10$; females only: $P=0.32$), but the combined probability for the adult and larval pattern is significant (both sexes: Fisher's combined probability value $\chi^2_4=11.8$, $P=0.02$; females only: $\chi^2_4=9.57$, $P=0.048$). The disparity between larvae and adults in the strength of the latitudinal correlation is most likely due to sample size differences, and sampling differences in space and time. This correlation with latitude appears to be the first documented case of Bergmann's rule *within* a single species of insect. The correlation of body size with elevation is a more common pattern in insects and other poikilotherms (Bizer, 1978; Berven, 1982; Berven & Gill, 1983).

What accounts for these geographic clines? We can eliminate

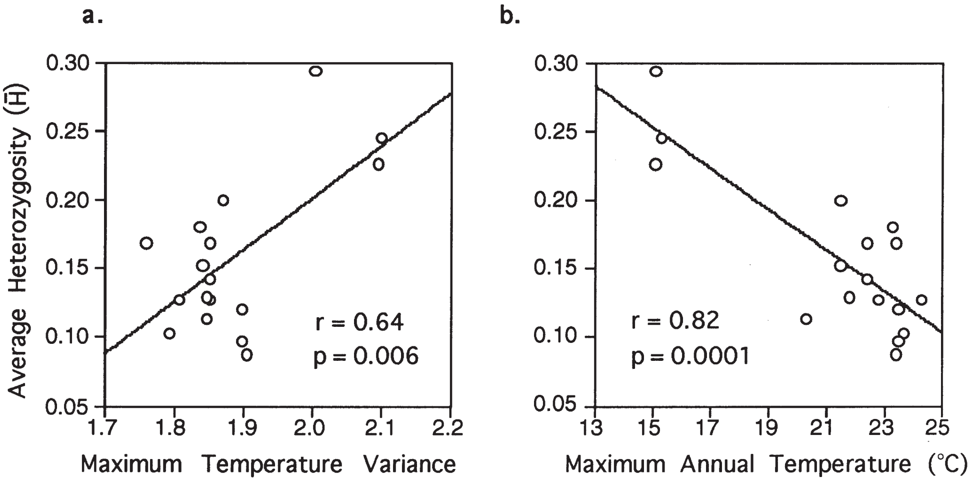

Figure 4 (a) Average observed heterozygosity increases significantly with maximum temperature variance. (b) Average observed heterozygosity decreases with increasing maximum temperature.

Table 3 Average expected and observed heterozygosities ± one standard deviation for seventeen *M. immaculatus* populations.

Population	*n*	Average expected heterozygosity	Average observed heterozygosity
1. Columbus, MS (1)	31	0.315 ± 0.112	0.097 ± 0.061
2. Columbus, MS (2)	30	0.434 ± 0.096	0.120 ± 0.057
3. Fulton, MS	32	0.313 ± 0.098	0.087 ± 0.033
4. Calera, AL	30	0.402 ± 0.103	0.180 ± 0.057
5. Childersburg, AL	30	0.368 ± 0.100	0.127 ± 0.078
6. Truissville, AL	30	0.364 ± 0.107	0.127 ± 0.037
7. Chattahoochee, GA	30	0.319 ± 0.118	0.113 ± 0.065
8. Tallulah, GA	33	0.504 ± 0.045	0.152 ± 0.066
9. Clemson, SC	31	0.385 ± 0.079	0.129 ± 0.037
10. Lake City, SC	33	0.396 ± 0.090	0.103 ± 0.065
11. Loris, SC	31	0.531 ± 0.044	0.168 ± 0.087
12. Washington, NC	34	0.403 ± 0.083	0.141 ± 0.050
13. Greenville, NC	31	0.434 ± 0.090	0.142 ± 0.063
14. Raleigh, NC	43	0.411 ± 0.111	0.200 ± 0.087
15. Beach Pond, RI	32	0.427 ± 0.064	0.294 ± 0.126
16. Mansfield Hollow, CT	31	0.408 ± 0.059	0.245 ± 0.073
17. Staffordville, CT	31	0.363 ± 0.065	0.226 ± 0.106

the hypothesis that the patterns are an artifact of our sampling protocol because the same patterns we described for our samples of third-instar larvae (Fig. 2a,b) were also seen in museum specimens of adult ant lions (Fig. 2c,d).

We recognize that our results are caused, in part, by the latitudinal 'hole' in our data between 36° and 40° N latitude (Fig. 1). However, this is not simply a sampling artifact. Instead, it represents the geographic pattern of population structure. We do not have an explanation for the puzzling absence or rarity of ant lion populations in the fifteen sites that we censused in the region. We returned to this region in the summer of 1997 and again found few third-instar larvae.

Two classic explanations for Bergmann's rule are heat conservation (Bergmann, 1847; Kendeigh, 1969), and ecological character displacement with competing species (McNab, 1971). Heat conservation is not relevant for poikilotherms. Character displacement also cannot account for the latitudinal cline in *M. immaculatus* because it was the only species occurring in most of our collection sites. *M. immaculatus* does occur with other species in the southern part of its range (*M. crudelis*, *M. carolinus*, and *M. mobilis*), but *M. crudelis* and *M. carolinus* are smaller in body size than *M. immaculatus*, and *M. mobilis* overlaps with *M. immaculatus* only along the Alabama and Georgia coast (Lucas & Stange, 1981).

Latitudinal clines in body size can also be explained by differential migration or historical dispersal patterns leading to larger-bodied individuals (or species) in the north (Cushman *et al.*, 1993). However, ant lions are poor dispersers, and populations are genetically well-differentiated from one another (Table 4). Moreover, heterozygosity was higher in the north than in the south (Fig. 3), which is not what we would expect if small populations of large-bodied individuals were dispersing from the south.

Recently, Van Voorhies (1996) suggested that Bergmann's rule in ectotherms simply reflects an increase in cell size with decreasing temperatures. However, this mechanism is unlikely to be a determinant of Bergmann's rule in ectotherms (see Mousseau, 1997 and Partridge & Coyne, 1997). Other studies on the effects of temperature on cell size of ectotherms have shown a diversity of results, with no consistent increase in cell size at cooler temperatures (Atkinson, 1994). Moreover, if the cell-size hypothesis were correct for ant lions, body size should have been positively correlated with elevation (Fig. 2b,d).

It is interesting that many of the environmental factors invoked to explain Bergmann's rule covary with elevation and latitude, and that many geographic clines in body size show parallel patterns at high elevations and latitudes (James, 1970; Masaki, 1978). In contrast, ant lion body size increases with latitude, but decreases with elevation. Therefore, we should look for environmental factors that vary with latitude, but not with elevation, as a causal explanation for these patterns.

Table 4 *F*-statistics (Wright, 1978) for each variable locus of *M. immaculatus* from populations within specific regions and among all populations. Gene flow (number of migrants per generation) is shown in parentheses for each region and among all populations. Significance of the F_{st} is indicated by the G-value. Locus names are given in Methods. (* $P<0.05$; ** $P<0.01$; *** $P<0.001$)

Region	Locus	F_{is}	F_{it}	F_{st}	G-value	d.f.
MS	GPI	0.356	0.361	0.008	5.498	4
(4.6)	MDH	0.884	0.888	0.030	28.816***	8
	DIA	0.916	0.922	0.079	40.416***	6
	PEP	0.388	0.423	0.057	10.801**	2
	SOD	1.000	1.000	0.069	14.002***	2
Mean		0.713	0.728	0.051	99.533***	22
AL	GPI	−0.064	−0.028	0.034	8.520	4
(5.7)	MDH	0.701	0.707	0.020	5.991	4
	DIA	0.865	0.876	0.077	25.653***	4
	PEP	0.510	0.523	0.027	7.537	4
	SOD	1.000	1.000	0.023	5.683	2
Mean		0.618	0.634	0.042	53.384***	18
GA	GPI	0.255	0.579	0.435	133.524***	4
(0.74)	MDH	0.712	0.760	0.167	77.386***	4
	DIA	0.844	0.868	0.155	65.201***	6
	PEP	0.566	0.572	0.013	2.476	2
	SOD	1.000	1.000	0.592	124.696***	2
Mean		0.674	0.756	0.252	403.283***	18
SC	GPI	0.080	0.161	0.089	22.272***	2
(4.1)	MDH	0.944	0.946	0.029	8.368*	2
	DIA	0.946	0.950	0.069	19.982***	2
	PEP	0.428	0.447	0.033	4.280*	1
	SOD	1.000	1.000	0.082	11.021***	1
Mean		0.708	0.725	0.057	65.924***	8
NC	GPI	0.102	0.110	0.008	8.724	4
(7.8)	MDH	0.822	0.825	0.016	12.703*	6
	DIA	0.885	0.894	0.077	52.180***	6
	PEP	0.316	0.321	0.007	8.762	4
	SOD	1.000	1.000	0.032	11.134**	2
Mean		0.613	0.625	0.031	93.502***	22
CT/RI	GPI	0.213	0.252	0.049	27.923***	6
(8.4)	MDH	−0.172	−0.105	0.058	30.361***	4
	DIA	0.625	0.626	0.003	1.167	4
	PEP	0.582	0.587	0.011	2.170	2
	SOD	1.000	1.000	0.007	1.364	2
Mean		0.362	0.380	0.029	62.984***	18
Overall Mean (0.73)		0.609	0.708	0.254	2422.774***	208

One such factor is photoperiod. Insect growth, metamorphosis, development, and body size are often tightly controlled by photoperiod (Philogene, 1982; Ruberson, Bush & Kring, 1991; Blanckenhorn & Fairbairn, 1995; Fantinou, Tsitsipis & Karandinos, 1996). In ant lions, Furunishi & Masaki (1983) found that photoperiod acts as a cue to slow or increase growth rate in all instars. They also found a high variance in instar and pupal weight that depended on the photoperiod in which larvae were raised. Thus, latitudinal differences in photoperiod may be responsible for reversing the negative correlation we found for ant lion body size and elevation. We suggest that photoperiod should be considered as a possible explanation for geographic gradients in body size of insects.

Our data cannot distinguish among a variety of other hypotheses that implicate variation in food or temperature as a cause of Bergmann's rule. These include geographic gradients in prey body size (McNab, 1971), starvation resistance of populations in seasonal enviroments (Kondoh, 1977; Lindstedt & Boyce, 1985), and effects of cooler temperatures (Berrigan & Charnov, 1994; Atkinson, 1994), reduced humidity (Aldrich & James, 1991; Bonato, Mapangoudivassa & Gutierrez, 1995), seasonality (Murphy, 1985), voltinism (Roff, 1992; Bradford & Roff, 1995) and food availability (Boyce, 1978; McNab, 1971) on survival and individual growth rates. Some of these hypotheses are also consistent with our finding that heterozygosity increases in the north (Fig. 3), and in populations that experience greater seasonal fluctuations in temperature (Fig. 4a).

Clines in body size may also result from geographic variation in juvenile or adult mortality (Rowe & Ludwig, 1991; Roff, 1992). We do not have data on this type of mortality in the field. However, local population density of ant lions did not vary with latitude ($r=0.09$; $P=0.80$) or elevation ($r=-0.18$, $P=0.61$), so mortality due to crowding is probably not important.

A complete understanding of Bergmann's rule requires information on the genetic and environmental components of body size (James, 1982). For our data, we cannot say how much of the geographic variation in body size is due to genetic differences between populations (Blanckenhorn & Fairbairn, 1995; Conover & Present, 1990), and how much is due to phenotypic plasticity of individuals growing in different environments (Via, 1985; Kodoh, Ishiguri & Kawano, 1995). However, our allozyme analyses (Table 4) do suggest that populations are relatively isolated and well-differentiated genetically from one another, and that heterozygosity increases in the north (Fig. 3). We are currently conducting common garden and reciprocal-transplant experiments (A. Arnett, unpublished data) to tease apart the environmental and genetic components of body size in larval ant lions and to directly test some of the hypotheses that have been proposed to account for Bergmann's rule.

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REFERENCES

Aldrich, J.W. & James, F. (1991) Ecogeographic variation in the American robin (*Turdus migratorius*). *The Auk*, **108**, 230–249.

Atkinson, D. (1994) Temperature and organism size—a biological law for ectotherms? *Adv. Ecol. Res.* **25**, 1–58.

Banks, B. (1927) Revision of the nearctic Myrmeleonidae. *Bull. Mus. Comp. Zool.* **68**, 72–79.

Bergmann, C. (1847) Ueber die Verhältnisse der Wärmeökonomie der Thiere zu ihrer Grösse. *Guttinger Stud.* **3**, 595.

Berrigan, D. & Charnov, E.L. (1994) Reaction norms for age and size at maturity in response to temperature: a puzzle for life historians. *Oikos*, **70**, 474–478.

Berven, K.A. (1982) The genetic basis of altitudinal variation in the wood frog *Rana sylvatica* II. An experimental analysis of larval development. *Oecologia*, **52**, 360–369.

Berven, K.A. & Gill, D.E. (1983) Interpreting geographic variation in life-history traits. *Am. Zool.* **23**, 85–97.

Bizer, J.R. (1978) Growth rates and size at metamorphosis of high elevation populations of *Ambystoma tigrinum*. *Oecologia*, **34**, 175–184.

Blanckenhorn, W.U. & Fairbairn, D.J. (1995) Life history adaptation along a latitudinal cline in the water strider *Aquarius remigis* (Heteroptera: Gerridae). *J. Evol. Biol.* **8**, 21–41.

Bonato, O., Mapangoudivassa, S. & Gutierrez, J. (1995) Influence of relative humidity on life-history parameters of *Mononychellus progresivus* and *Oligonychus gossypii* (Acari: Tetranychidae). *Environ. Entomol.* **24**, 841–845.

Boyce, M. (1978) Climatic variability and body size variation in the muskrats (*Ondatra zibethicus*) of North America. *Oecologia*, **36**, 1–19.

Bradford, M.J. & Roff, D.A. (1995) Genetic and phenotypic sources of life history variation along a cline in voltinism in the cricket *Allonemobius socius*. *Oecologia*, **103**, 319–326.

Brodie, P.F. (1975) Cetacean energetics, an overview of intraspecific size variation. *Ecology*, **56**, 152–161.

Brown, J. & Lee, A. (1969) Bergmann's rule and climatic adaptation in woodrats (*neotoma*). *Evolution*, **23**, 329–338.

Brown, J.H. (1995) *Macroecology.* University of Chicago Press, Chicago.

Brunkow, P.E. & Collins, J.P. (1996) Effects of individual variation in size on growth and development of larval salamanders. *Ecology*, **77**, 1483–1492.

Conover, D. & Present, T. (1990) Countergradient variation in growth rate: compensation for length of the growing season among Atlantic silversides from different latitudes. *Oecologia*, **83**, 316–324.

Cushman, J.H., Lawton, J.H. & Manly, B.F.J. (1993) Latitudinal patterns in European ant assemblages: variation in species richness and body size. *Oecologia*, **95**, 30–37.

Eng, W. & Paul, E. (1992) *Genes in populations*. Designed by B. May and C. Krueger. http://animalscience.ucdavis.edu/software/may.htm.

Fantinou, A.A., Tsitsipis, J.A. & Karandinos, M.G. (1996) Effects of short- and long-day photoperiods on growth and development of *Sesamia nonagrioides* (Lepidoptera: Noctuidae). *Environ. Entomol.* **25**, 1337–1343.

Felsenstein, J. (1985) Phylogenies and the comparative method. *Am. Nat.* **125**, 1–15.

Forsman, A. (1991) Variation in sexual size dimorphism and maximum body size among adder populations: effects of prey size. *J. anim. Ecol.* **60**, 253–267.

Furunishi, S. & Masaki, S. (1981) Photoperiodic response of the univoltine ant-lion *Myrmeleon formicarius* (Neuroptera, Myrmeleontidae). *Kontyu*, **49**, 653–667.

Furunishi, S. & Masaki, S. (1982) Seasonal life cycle in two species of ant-lion (Neuroptera: Myrmeleontidae). *Jap. J. Ecol.* **32**, 7–13.

Furunishi, S. & Masaki, S. (1983) Photoperiodic control of development in the ant-lion *Hagenomyia micans* (Neuroptera: Myrmeleontidae). *Entomol. Generalis*, **9**, 51–62.

Geist, V. (1987) Bergmann's rule is invalid. *Can. J. Zool.* **65**, 1035–1038.

Gotelli, N.J. (1993) Ant lion zones: causes of high-density predator aggregations. *Ecology*, **74**, 226–237.

Graves, G.R. (1991) Bergmann's rule near the equator: latitudinal clines in body size of an Andean passerine bird. *Proc. Natl Acad. Sci.* **88**, 2322–2325.

Hartl, D.L. & Clark, A.G. (1989) *Principles of population genetics*. Sinauer Associates, Inc., Sunderland, Mass.

Hawking, J.H. & New, T.R. (1996) The development of dragonfly larvae (Odonata: Anisoptera) from two streams in north-eastern Victoria, Australia. *Hydrobiologia*, **317**, 13–30.

Hawkins, B.A. & Lawton, J. (1995) Latitudinal gradients in butterfly body sizes: is there a general pattern? *Oecologia*, **102**, 31–36.

Hedrick, P.W., Ginevan, M.E. & Ewing, E.P. (1976) Genetic polymorphism in heterogeneous environments. *Ann. Rev. Ecol. Syst.* **7**, 1–32.

Hogue, J.N. & Hawkins, C.P. (1991) Morphological variation in adult aquatic insects: associations with developmental temperature and seasonal growth patterns. *J. N. Am. Benthol. Soc.* **10**, 309–321.

Hutchings, J.A. (1996) Adaptive phenotypic plasticity in brook trout, *Salvelinus fontinalis*, life histories. *Ecoscience*, **3**, 25–32.

Iba, M., Nagao, T. & Urano, A. (1995) Effects of population density on growth, behavior and levels of biogenic amines in the cricket, *Gryllus bimaculatus*. *Zool. Sci.* **12**, 695–702.

James, F. (1970) Geographic size variation in birds and its relationship to climate. *Ecology*, **51**, 365–390.

James, F. (1982) The ecological morphology of birds: a review. *Annales Zool. Fenn.* **19**, 265–275.

Kendeigh, S.C. (1969) Tolerance of cold and Bergmann's rule. *The Auk*, **86**, 13–25.

Kodoh, H., Ishiguri, Y. & Kawano, S. (1995) Phenotypic plasticity in *Cardamine flexuaosa*: variation among populations in plastic response to chilling treatments and photoperiods. *Oecologia*, **103**, 148–156.

Koehn, R.K., Diehl, W.J. & Scott, T.M. (1988) The differential contribution by individual enzymes of glycolysis and protein catabolism to the relationship between heterozygosity and growth rate in the coot clam *Mulinia lateralis*. *Genetics*, **118**, 121–130.

Kondoh, M. (1977) On the difference of vitality among worker ants under starvation. *8th international congress of the international union for the study of social insects*, pp. 69–70. Centre for Agricultural Publishing and Documentation, The Netherlands.

Ledig, F.T., Guries, R.P. & Bonefield, B.A. (1983) The relation of growth to heterozygosity in pitch pine. *Evolution*, **37**, 1227–1238.

Levinton, J.S. & Monahan, R.K. (1983) The latitudinal compensation hypothesis: growth data and a model of latitudinal growth differentiation based upon energy budgets. II. Intraspecific comparisons between subspecies of *Ophryotrocha puerilis* (Polychaeta: dorvilleidae). *Biol. Bull.* **165**, 699–707.

Lewontin, R.C. & Levins, R. (1989) On the characterization of density and resource availability. *Am. Nat.* **134**, 513–524.

Lindstedt, S.L. & Boyce, M.S. (1985) Seasonality, fasting endurance, and body size in mammals. *Am. Nat.* **125**, 873–878.

Liu, L.L., Soong, K. & Chen, C.P. (1995) Allozyme variation in the Pacific oyster *Crassostrea gigas* along the coast of Taiwan. *Zool. Stud*. **34**, 177–182.

Lucas, J. & Stange, L. (1981) Key and descriptions to the *Myrmeleon* larvae of Florida (Neuroptera: Myrmeleontidae). *Fla Entomol*. **64**, 208–216.

Lutz, P.E. (1974) Environmental factors controlling duration of larval instars in *Tetragoneuria cynosura* (Odonata). *Ecology*, **55**, 630–637.

Masaki, S. (1967) Geographic variation and climatic adaptation in a field cricket (Orthoptera: Gryllidae). *Evolution*, **21**, 725–741.

Masaki, S. (1978) Seasonal and latitudinal adaptations in the life cycles of crickets. *Evolution of insect migration and diapause* (ed. H. Dingle), pp. 72–100. Springer-Verlag, New York.

May, B. (1991) Starch gel electrophoresis of allozymes. *Molecular genetic analysis of populations: a practical approach* (ed. A.R. Hoelzel), pp. 1–27 and 271–280. Oxford University Press, Oxford.

McNab, B. (1971) On the ecological significance of Bergmann's rule. *Ecology*, **52**, 845–854.

Mitton, J.B. & Grant, M.C. (1984) Associations among protein heterozygosity, growth rate and developmental homeostasis. *Ann. Rev. Ecol. Syst*. **15**, 479–499.

Mousseau, T.A. (1997) Ectotherms follow the converse to Bergmann's rule. *Evolution*, **51**, 630–632.

Murphy, E.C. (1985) Bergmann's rule, seasonality, and geographic variation in body size of house sparrows. *Evolution*, **39**, 1327–1334.

National Climatic Data Center. (1992) *Climatography of the United States*, *No. 81*. Asheville, North Carolina.

Nei, M. (1972) Genetic distance between populations. *Am. Nat*. **106**, 283–292.

Partridge, L. & Coyne, J.A. (1997) Bergmann's rule in ectotherms: is it adaptive? *Evolution*, **51**, 632–635.

Philogene, B.J.R. (1982) Development rate changes in insects: the importance of photoperiod. *Am. Nat*. **120**, 269–272.

Power, D.M. (1969) Evolutionary implications of wing and size variation in the Red-winged Blackbird in relation to geographic and climatic factors: a multiple regression analysis. *Syst. Zool*. **18**, 363–373.

Roff, D.A. (1992) *The evolution of life histories: theory and analysis*. Chapman & Hall, New York.

Rowe, L. & Ludwig, D. (1991) Size and timing of metamorphosis in complex life cycles: time constraints and variation. *Ecology*, **72**, 413–427.

Rosenzweig, M.L. (1995) *Species diversity in space and time*. Cambridge University Press, Cambridge.

Ruberson, J.R., Bush, L. & Kring, T.J. (1991) Photoperiodic effect on diapause induction and development in the predator *Orius insidiosus*. *Environ. Entomol*. **20**, 786–789.

Scharples, C.M., Fa, J.E. & Bell, D.J. (1996) Geographical variation in size in the European rabbit *Oryctolagus cuniculus* (Lagomorpha: Leporidae) in western Europe and North Africa. *Zool. J. Linnean Soc*. **117**, 141–158.

Schnell, G.D., Watt, D.J. & Douglas, M.E. (1985) Statistical comparison of proximity matrices: applications in animal behaviour. *Anim. Behav*. **33**, 239–253.

Schoener, T.W. & Janzen, D.H. (1968) Notes on environmental determinants of tropical versus temperate insect size patterns. *Am. Nat*. **102**, 207–244.

Shaklee, J.B., Allendork, F.W., Morizot, D.C. & Whitt, G.S. (1990) Gene nomenclature for protein-coding in fish. *Trans. Am. Fish. Soc*. **119**, 2–15.

Slatkin, M. & Barton, N.H. (1989) A comparison of three indirect methods for estimating average levels of gene flow. *Evolution*, **43**, 1349–1368.

Sokal, R.R. & Rohlf, F.J. (1995) *Biometry, 3rd edn*. W.H. Freeman and Company, New York.

Stange, L.S. (1980) The ant-lions of Florida. I. Genera (Neuroptera: myrmeleontidae). Florida Department of Agriculture and Consumer Services, Division of Plant Industry, *Entomol. Circ*. **215**, 1–4.

Stearns, S. & Koella, J.C. (1986) The evolution of phenotypic plasticity in life-history traits: Predictions of reaction norms for age and size at maturity. *Evolution*, **40**, 893–913.

Strauss, R.E. (1991) Correlations between heterozygosity and phenotypic variability in *Cottus* (teleostei: cottidae): character components. *Evolution*, **45**, 1950–1956.

Taylor, C.M. & Gotelli, N.J. (1994) The macroecology of *Cyprinella*: correlates of phylogeny, body size and geographic range. *Am. Nat*. **112**, 583–593.

Van Voorhies, W.A. (1996) Bergmann size clines: a simple explanation for their occurrence in ectotherms. *Evolution*, **50**, 1259–1264.

Via, S. (1985) Genotype-environment interaction and the evolution of phenotypic plasticity. *Evolution*, **39**, 505–522.

Wheeler, W.M. (1930) *Demons of the dust*. Norton & Company, New York.

Wright, S. (1978) *Evolution and the genetics of populations. Vol. 4: Variability within and among natural populations*. University of Chicago Press, Chicago.

Zink, R.M. & Remsen, J.V. (1986) Evolutionary processes and patterns of geographic variation in birds. *Current Ornithology, Vol. 4* (ed. by R.F. Johnston), pp. 1–69. Plenum Press, New York.

BIOSKETCHES

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