ELSEVIER

Some like it hot: Body and weapon size affect thermoregulation in horned beetles

Brittany L. Shepherd[a], Henry D. Prange[a,b], Armin P. Moczek[a,*]

[a]Department of Biology, Indiana University, 915 East, Third Street, Myers Hall 150, Bloomington, IN 47405-7107, USA
[b]Medical Sciences Program, Indiana University, Bloomington, IN 47405, USA

Abstract

Body size and shape affect thermoregulatory properties of organisms, and in turn are believed to have shaped macroevolutionary patterns of morphological diversity across many taxa. However, it is less clear whether thermoregulation plays a role in shaping intraspecific morphological diversity such as sexual dimorphisms or the conditional expression of exaggerated secondary sexual traits. Here, we investigate individual thermoregulatory properties in two species of horned beetles that share similar ecologies and body size ranges, but differ substantially in degree of sexual and male dimorphism. We find that intraspecific variation in body size had an unexpectedly large effect on thermal preference behavior and the ability to passively regulate body temperature. Furthermore, we find that the presence or absence of exaggerated secondary sexual traits dramatically altered thermal preference behavior, consistent with a thermoregulatory cost of horn possession. Lastly, we show that the increase in surface area associated with the expression of enlarged horns is, by itself, insufficient to account for the radically altered thermoregulatory behavior observed in horn-bearing males, and discuss possible alternative, physiological explanations. These findings are among the first to link intra-and interspecific variation in body- and weapon size to thermal preferences within and between insect species.
© 2007 Elsevier Ltd. All rights reserved.

Keywords: Evolutionary constraint; Horned beetle; *Onthophagus*; Polyphenism; Thermoregulation

1. Introduction

Temperature fluctuations are a nearly ubiquitous, and at the same time critical, environmental variable in the lives of most organisms, and a diverse array of cellular, developmental, physiological, and behavioral mechanisms exist that permit organisms to respond adaptively to changes in temperature (Schmidt-Nielsen, 1997). Simple physical properties, foremost size and shape, contribute heavily in determining a given organism's ability to maintain constant internal temperature in the face of environmental fluctuations, and are believed to have shaped macroevolutionary patterns of morphological diversity in many taxa, as well as delineate ecological niche boundaries in many communities (Schmidt-Nielsen, 1994, 1997). However, it is less clear whether size and shape also affect thermoregulatory capacities within species or sexes, particularly in invertebrates, and whether thermoregulation may have played a role in shaping, or limiting, intraspecific morphological diversity such as sexual dimorphisms or the expression of exaggerated secondary sexual traits. Here, we explore individual thermoregulatory properties in two polyphenic beetle species as a function of intra- and interspecific variation in size and shape.

Polyphenism can be defined as a single genotype's ability to express two or more discrete, alternative phenotypes in response to changes in environmental conditions (Nijhout, 1994). Well studied examples of polyphenic development involve castes in social insects (Nijhout and Wheeler, 1982), seasonal morphs in butterflies (Shapiro, 1976), phase polyphenisms in locusts (Pener, 1991), and alternative male morphologies in a variety of arthropods (Crespi, 1988; Radwan, 1993; Moczek, 1998). In the context of

---

*Corresponding author. Tel.: +1 812 856 1468.
E-mail address: armin@indiana.edu (A.P. Moczek).

horned beetles, polyphenic development has been best studied in *Onthophagus* dung beetles, and involves the size-dependent expression of alternative horned (major) and hornless (minor) male morphs in response to differences in larval nutrition (Emlen, 1994; Moczek, 1998; Moczek and Emlen, 1999). Male larvae with access to optimal food amounts reach larger body sizes and in response express disproportionately long horns, whereas male larvae with access to suboptimal feeding conditions eclose to smaller adult sizes and remain largely or entirely hornless. Horned males use their horns as weapons in male combat over access to females, whereas hornless males rely primarily on sneaking behaviors (Moczek and Emlen, 2000; Madewell and Moczek, 2006). Female larvae exhibit the same nutrition dependence of final adult size as males, but unlike males do not express horns as adults. Polyphenic development in horned beetles therefore generates substantial variation in individual body sizes in males and females, as well as dramatic variation in male shape due to the condition-dependent expression of exaggerated secondary sexual traits. As a rough benchmark, adult body mass typically varies by a factor > 3 among conspecifics, and the size and mass of horns expressed by the largest males often far exceed that of regular appendages such as legs, wings, or mouthparts (Moczek, 1998). Here, we investigate individual thermoregulatory properties in two species of horn polyphenic beetles that differ in the magnitude of horn expression observed in large males. Specifically, we hypothesize (1) that larger animals are better able to withstand extreme temperatures by virtue of their slower rates of temperature change; (2) that because of their higher rates of temperature change, smaller animals would select lower temperatures overall to minimize the risk of overheating; and (3) that horn possession in large males results in an increase in the surface available for heat dissipation, leading to an altered temperature preference of these males compared to same-sized females. Lastly, since degree of male horn exaggeration differs between the two species studied here, we predict (4) that differences in temperature preferences between sexes should be restricted

to, or be more extreme in, the species with the more exaggerated male horns. Our approach combines simple behavioral studies on individual temperature preferences with experimental examination of warming rates and we discuss our results in the context of the behavioral ecology and physiology of horned beetles.

2. Material and methods

2.1. Species choice

Both species were chosen because they share very similar ecologies and body size ranges (approximately 4.5–6.5 mm thorax width), yet differ substantially in the degree of sexual and male dimorphism. *Onthophagus binodis* exhibits a very moderate sexual dimorphism and only large males express a short, broad pronotal outgrowth while smaller males and all females do not (Fig. 1(A)). In contrast, *Onthophagus nigriventris* exhibits a dramatic sexual and male dimorphism. Males larger than a critical size threshold express an enormous curved thoracic horn which is absent in females or smaller males (Fig. 1(B), Madewell and Moczek, 2006). The thoracic horn of large male *O. nigriventris* is hollow and hemolymph filled over its entire length, although exactly how much hemolymph circulation occurs through the horn is unknown. Both species were collected from pastures near Waimea, Hawaii, and maintained as described in Moczek and Nijhout (2003).

2.2. Thermal preference behavior

Thermal preference was determined with an alternative chamber or shuttle box as described in Prange and Hamilton (1992) (see also supplemental Fig. 1) in which beetles were free to move between a cool (15 °C) side and a warm (35–38 °C) side. Temperatures were maintained by an air stream that passed through either a resistance heater or a radiator cooled by a water bath and rose through the mesh floor of the chambers. The temperatures were
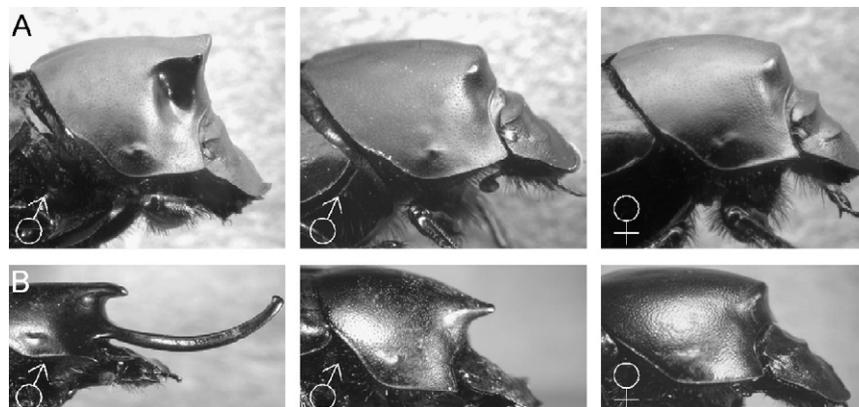

Fig. 1. Species used in the present study. (A) *Onthophagus binodis* and (B) *O. nigriventris*. Shown are large males (left), small males (center), and females (right). Note differences in sexual and male dimorphism between both species.

regulated by proportional controllers and thermistors placed in the air stream. By matching the flow rates it was possible to maintain discrete temperatures in each chamber without a physical barrier. Each individual was placed on the center line and its movements recorded with a video camera for 30 min. Beetles that failed to move between sides at least once in the first 15 min were excluded from the data. The video recordings were later scored to determine the time spent on each side and frequency of shuttling between sides. The preferred temperature was calculated from the weighted average of the time t spent in each temperature T according to the following equation:

$$T_{pref} = \frac{(t_{High}T_{High} + t_{Low}T_{Low})}{t_{Total}}.$$

High and low temperatures selected were presumed to be higher or lower, respectively, than the animal-preferred temperature, allowing us to calculate a time-weighted average as a proxy for a given individual's preferred temperature. The shuttle box technique was employed rather than a continuous gradient because it more closely resembles a natural environment with patches of sun and shade and because it provided a simple, repeatable, and standardized means of quantifying temperature preferences within and between species. Ten individuals of each sex and horn type were used. Body mass was determined to 0.1 mg with an analytical balance and body dimensions were measured with calipers to 0.1 mm.

2.3. Passive warming rate

Beetles freshly killed with ethyl acetate were weighed and a 36 ga. thermocouple was inserted into the center of the thorax. When the hemolymph hardened around the thermocouple, the beetle was placed in the cool side of the shuttle box until it reached a steady temperature. The beetle was then transferred quickly to the warm side and the temperature change was recorded from the output of the thermocouple reader on a calibrated chart recorder. Data were analyzed from a semilogarithmic plot to obtain the warming constant. We chose this method for consistency with our behavioral measurements. Because our apparatus employed forced convection, the rate of temperature change we measured is not directly comparable to measurements that were presumably taken in free convection, as e.g., in Bartholomew and Epting (1975).

2.4. Statistical analyses

Following earlier studies on morphological and physiological correlates of size and shape in horned beetles (e.g., Emlen, 1996, 2001; Nijhout and Emlen, 1998; Simmons and Emlen, 2006) we employed a residual analysis to examine the effects of species, sex, size, and horn possession on thermoregulatory behavior. We first employed simple linear regressions to explore the effect of adult mass on thermoregulatory behavior. We initially pooled all individuals regardless of sex to maximize statistical power. Once we obtained a significant effect, we then separated the sexes and employed a residual analysis to determine (i) whether males and females differed in thermoregulatory behavior (both species), and if so (ii) whether these differences could be attributed to the presence or absence of large horns (*O. nigriventris* only). To do so we used measurements collected from females to fit a linear regression describing the average relationship between female body mass and the mean preferred temperature T_{pref}. This then allowed us to calculate the expected T_{pref} for a given body mass in females and males, and to characterize each individual by the difference between the observed and expected T_{pref} given its individual body mass, or its *residual* T_{pref}. We then contrasted male and female residual T_{pref} estimates in both species using two-tailed, non-parametric Mann Whitney U-tests. Using the same test, we contrasted horned and hornless male morphs (*O. nigriventris* only) by comparing their T_{pref} estimates separately to that of females, as well as to each other. We used an identical approach to examine the effects of body mass and horn possession on shuttling frequency and passive warming rates, respectively. All analyses were replicated using surface/volume ratios as independent variable instead of body mass. This, however, neither improved fit of regressions nor changed the outcome of comparisons, and for ease of interpretation we therefore graphed our results using body mass as the independent variable. Unless otherwise noted, all results are presented as mean ± standard error.

3. Results

3.1. Effect of size, sex, and morph

Body mass had a dramatic and highly significant effect on the average weighted temperature in both species (Figs. 2 and 3). Small individuals consistently sought out much cooler temperatures than their larger counterparts (*O.binodis*, $p < 0.0001$; *O. nigriventris*, $p = 0.039$). Male and female *O. binodis* did not differ in their thermoregulatory behavior (Fig. 2 insert), however, we detected a complex relationship between thermal preferences and mass, sex, and horn possession in *O. nigriventris* (Fig. 3). In this species, small, hornless males showed the same absolute temperature preferences, and the same increase in temperature preferences with body mass, as their female counterparts. However, large, horned males did not show the further increase in average weighted temperatures that was predicted given their body size and instead sought out much cooler temperatures relative to their size compared to their female ($z = 2.34$; $p = 0.019$) or hornless male ($z = 2.94$; $p = 0.0032$) counterparts (Fig 3. insert). In fact, if horned males were analyzed separately, preferred temperatures exhibited a strong trend to *decline*, rather than increase, with male body size ($p = 0.06$; after removal of a single outlier $p = 0.031$). If hornless and horned males
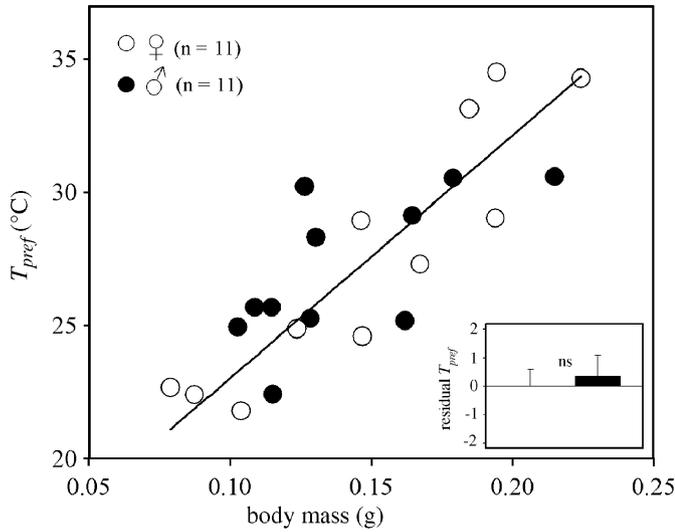
Fig. 2. Temperature preference (T_{pref}) as a function of body mass and sex in *O. binodis*. T_{pref} increased significantly with body mass in the combined sample ($p < 0.0001$) as well as when females ($p = 0.0001$) and males ($p = 0.02$) were analyzed separately. Reference line indicates the best-fit linear regression of female T_{pref} on body mass used to calculate T_{pref} residuals for both sexes ($y = 13.91 + 91.2x$; $r^2 = 0.83$). Insert: T_{pref} residuals in males and females ($p = $ ns). Sample sizes are indicated in parentheses.
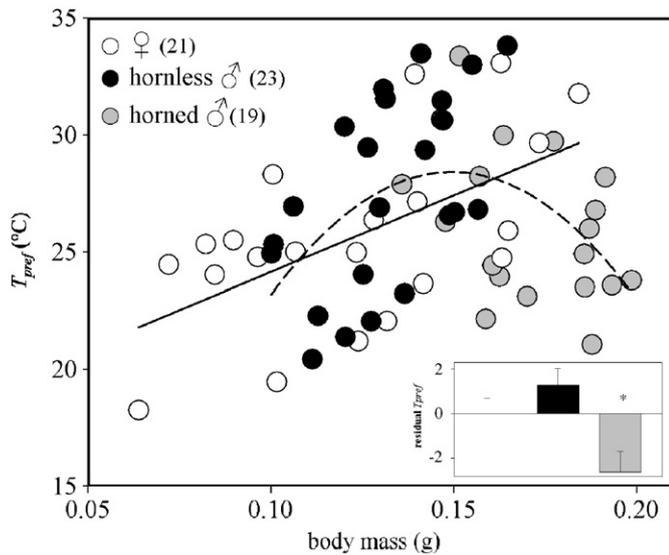

Fig. 3. Temperature preference (T_{pref}) as a function of body mass and sex in *O. nigriventris*. T_{pref} increased significantly with body mass in the combined sample ($p = 0.039$), in females ($p = 0.005$), hornless males ($p = 0.0054$), but not in large, horned males, which instead exhibited the opposite trend ($p = 0.06$). If hornless and horned males were combined, temperature preferences could be modeled best by a quadratic function (dashed line; $y = -19.45 + 639.96x - 2139x^2$; $r^2 = 0.19$) including a highly significant, negative quadratic term ($p = 0.0043$). In contrast, a linear regression yielded no significant fit ($p = 0.89$; $r^2 = 0.00$). Solid line indicates the best-fit linear regression of female T_{pref} on body mass ($y = 17.6 + 65x$; $r^2 = 0.35$) used to calculate T_{pref} residuals for females and both male morphs. Insert: T_{pref} residuals in females, and horned and hornless males. T_{pref} residuals of horned males are significantly different compared to hornless males ($z = 2.94$; $p = 0.0032$) and females ($z = 2.34$; $p = 0.019$). All other comparisons are non-significant. Sample sizes are indicated in parentheses.

were combined, male temperature preferences could be modeled best by a quadratic function (dashed line; $y = -19.45 + 639.96x - 2139x^2$; $r^2 = 0.19$), including a highly significant, negative quadratic term ($p = 0.0043$). In contrast, a linear regression yielded no significant fit ($p = 0.89$; $r^2 = 0.00$). Temperature preferences of male *O. nigriventris* therefore appear to exhibit a peak at medium body mass and then declined significantly to both smaller and larger body sizes.

3.2. Mobility

Body mass strongly affected shuttling frequency in both species, with small adults shuttling approximately twice as much as large adults (*O. nigriventrism*, $p < 0.0001$, Fig. 4; *O.binodis*, $p = 0.055$; after removal of one outlier $p = 0.01$; not shown). However, in both species males and females showed a very similar decline in shuttling frequency with increasing body mass. Specifically, in *O. nigriventris*, large horned males showed the same reduced level of shuttling than their same-sized, but hornless, female counterparts, suggesting that even though horned males exhibit very different temperature preferences compared to same-sized females, these do not appear to be brought about by differences in overall levels of activity.

3.3. Warming rate

Body mass dramatically affected passive heat loss in adult *O. nigriventris*. Small individuals lost heat at up to
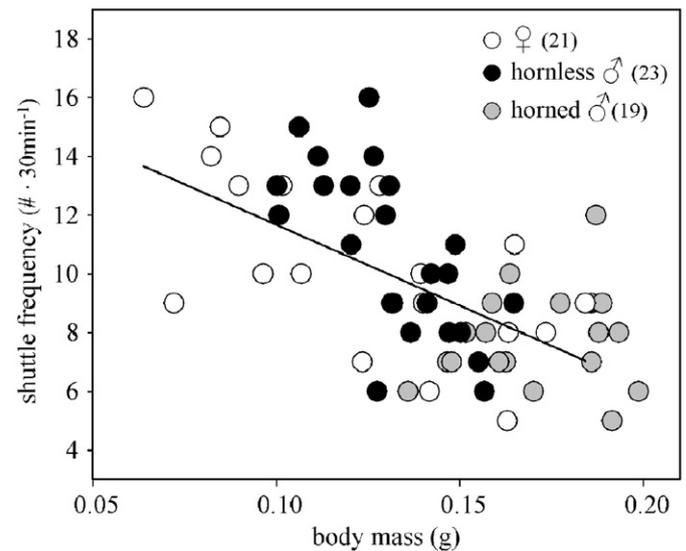

Fig. 4. Shuttle frequency during 30 min observation as a function of body mass, sex, and male morph in *O. nigriventris*. Shuttle frequency decreased significantly with body mass in the combined sample ($p < 0.0001$) as well as when females ($p = 0.0012$) and males ($p < 0.0001$) were analyzed separately. Solid line indicates the best-fit linear regression of female shuttle frequency on body mass ($y = 17.17 - 55x$; $r^2 = 0.43$) used to calculate frequency residuals for females and both male morphs. No significant differences were detected. Sample sizes are indicated in parentheses.

five times the rate compared to large individuals, which is consistent with predictions given their much increased surface/volume ratio. Importantly, horned males exhibited significantly lower warming rates compared to hornless males ($z = 2.49$, $p = 0.013$), but very similar rates as their same-sized, but hornless, female counterparts ($p = $ n.s.). Thus, we were unable to find any effect of horn possession on passive warming rate, which rejects the hypothesis that horn possession via the associated increase in surface area could, by itself, explain the discrepancy in thermal preference observed between large males and females of this species.

4. Discussion

Body size plays a fundamental role in defining thermoregulatory challenges to, and limits of, organisms (Schmidt-Nielsen, 1994, 1997). With the exception of very large species and active flyers, insects are generally incapable of generating enough heat through physiological processes to maintain a constant body temperature and are therefore heavily impacted by changes in external temperature (May, 1979). Both species used in the present study are much smaller than the body size above which the regulation of body temperature through internal processes is thought to be physiologically possible (Bartholomew and Heinrich, 1978; Prange, 1996; Roxburgh et al., 1996) and therefore have to rely on other mechanisms to respond to changes in external temperature. One mechanism to respond to such changes is to actively seek out locations with appropriate temperatures, or to move between warm and cold temperature locations so as to achieve a preferred average temperature. A large body of work contrasting species across size magnitudes has shown that the ability, and need, to thermoregulate by changing locations is heavily dependent on body mass, and closely associated, the surface/volume ratio of an individual. Larger individuals, by virtue of their reduced surface to volume ratio, retain their internal temperature relatively longer if challenged with a change in external temperature compared to smaller individuals (Bartholomew and Heinrich, 1978). Here we contribute to a growing body of evidence that suggests that this pattern also manifests itself on a much finer scale, namely among differently sized conspecifics, resulting in dramatically different temperature preferences among members of the same species or sex. Moreover, we show that this pattern is further complicated by the nature and degree of intraspecific diversity in the expression of secondary sexual traits, and we discuss the implications of our findings below.

4.1. Body mass affects temperature preference within species

Our results suggest that even though body mass only changed by a factor of 2–3 within both species studied here, it nevertheless dramatically affected individual thermal preferences. We show in two species that large individuals

consistently preferred higher temperatures than smaller individuals. This observation is consistent with the hypothesis that larger individuals, by virtue of their size, can operate at higher temperatures, whereas smaller individuals, possibly due to increased risk of overheating, prefer to operate at lower temperatures. Regardless of the exact underlying causes, our results are in line with the prediction that increases in body size increase thermoregulatory capacity, a notion further supported by our estimates of passive warming rates, which decreased highly significantly with increasing body size (Fig. 5). Furthermore, our results are in line with several earlier studies on butterflies, which also documented an effect of intraspecific variation in size on heating capacity (Gilchrist, 1990; Berwaerts et al., 2001), suggesting that such effects are not restricted to any particular insect order. What was unexpected, however, was the sheer magnitude of differences in temperature preferences. In both species, small and large individuals differed by as much as 15 °C (Figs. 2 and 3), which suggests that thermal preferences might play a large, and previously ignored, role in behavioral niche partitioning within species. *Onthophagus* beetles reproduce by flying and walking to fresh dung pads. Once located, adults dig tunnels underneath dung pads and pull pieces of dung into the tunnels, which are then shaped into brood balls, and provisioned with a single egg (reviewed in Moczek, 1998). Copulation, mate guarding, and male contests all take place inside tunnels, which vary in depth from a few to up
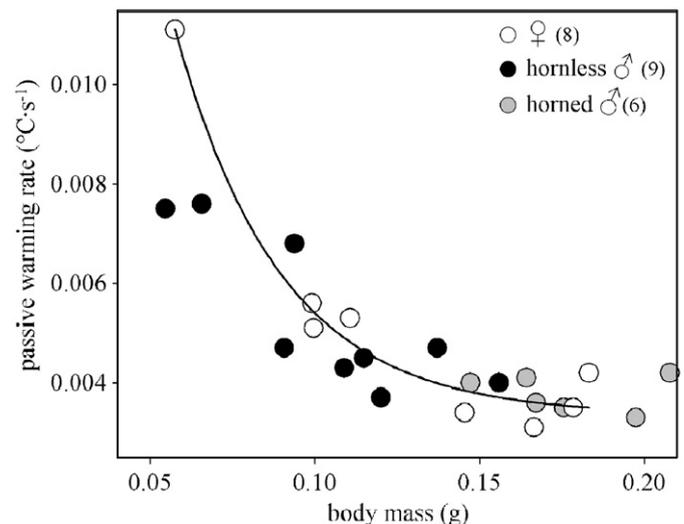

Fig. 5. Passive warming rate as a function of body mass, sex, and male morph in *O. nigriventris*. High rates indicate reduced ability to retain heat. Warming rate decreased exponentially with body mass regardless of sex. Horned males exhibited significantly lower warming rates compared to hornless males ($z = 2.49$; $p = 0.013$), but similar warming rates as their same-sized, but hornless, female counterparts. Solid line indicates the best-fit exponential decay regression of female warming rate on body mass ($y = 0.0034 + 0.0472\,e^{(-31.347x)}$; $r^2 = 0.97$) used to calculate rate residuals for females and both male morphs. No significant differences were detected.

to 120 cm depending on species and digging substrate (Halffter and Edmonds, 1982). Both species tested here regularly reproduce in the lab by generating tunnels with an approximate depth of 30 cm. Given their above- and below-ground ecology, *Onthophagus* beetles are likely to experience a wide range of temperatures, and the temperature range used in this study most likely captures an important fraction of the biologically relevant temperature range these species experience in nature. Interestingly, even though both species exhibit similar body size ranges, they nevertheless differed in the exact relationship between body size and individual thermal preferences, which may be a reflection of adaptations to different microhabitats.

4.2. Degree of sexual dimorphism affects temperature preference

Females of both species, as well as all males of the mildly sexually dimorphic *O. binodis*, and hornless males of the pronounced sexually dimorphic *O. nigriventris*, exhibited very similar body size-dependent thermal preference behavior. Large, horned male *O. nigriventris*, however, exhibited the opposite pattern, and average weighted temperatures actually declined with increasing size (Fig. 3). If all male *O. nigriventris* were analyzed together, their temperature preferences as a function of body mass could be modeled best by a quadratic function, including a highly significant, negative quadratic term, while a linear regression model yielded no significant fit. Combined, these results therefore support that T_{pref} of male *O. nigriventris*, but not male *O. binodis* or females of either species, peak at medium body mass and then decline significantly to both smaller and larger body sizes. Interestingly, the body mass correlated with the highest T_{pref} coincided with the body size threshold, above which males express the horned morph (Madewell and Moczek, 2006). An effect of secondary sexual traits on thermoregulatory properties has previously been documented in several elegant studies on butterflies, which showed that sex- or morph-specific differences in pigmentation patterns influence warming rates, activity patterns, and flight performance of individuals (e.g., Kingsolver and Wiernasz, 1991; Van Dyck and Matthysen, 1998; Berwaerts et al., 2001, 2002). However, to the best of our knowledge this is the first study that links the possession of weapons used in male combat to thermal preference behavior. Specifically, our results suggest that horn possession somehow alters thermal preferences among males, ultimately causing very large, horned males to exhibit preferred temperatures similar to those of very small males, rather than their same-sized, but hornless, female counterparts.

4.3. How do horns affect thermoregulation?

Analyses of shuttling frequencies ruled out that horn *O. nigriventris* show different behavioral activity patterns compared to same-sized females. Horned males shuttled just as frequently between temperature chambers as females of comparable size, but unlike females, horned males spent more time in colder temperatures. This observation hinted at the possibility that rather than affecting mobility, horn possession may affect temperature retention of adults. Horns, as in *O. nigriventris*, are similar or larger in size as regular appendages such as legs. As such they have the potential to increase surface area and, thus, heat transfer. If correct, passive warming rates should be increased in large horned males compared to similar-sized, but hornless females. This was not supported by our data, instead horned males warmed up at the same rate as same-sized females, and a much lower rate than smaller, hornless males (Fig. 5). This suggests that the increase in surface area associated with the expression of exaggerated horns is, by itself, insufficient to explain the decline in average weighted temperatures observed among horned male *O. nigriventris*. It is important to emphasize that passive warming rates were, as per definition, collected on recently euthanized individuals. Given the data, we now propose that for horn possession to affect heat transfer, individuals must be physiologically active, via one of two possible mechanisms.

One mechanism by which this could be achieved is the active circulation of hemolymph through the horn, which would permit the horn to act both as an absorber and radiator of heat in the living animal without leaving a signature of this effect in dead beetles. Increased heat absorption through the horn would place large horned males, but not their same-sized female counterparts, at greater risk of overheating and thus may explain the tendency of the former to seek out lower preferred temperatures. It is worth emphasizing that in *O. nigriventris*, thoracic horns are hollow over their *entire* length and filled with hemolymph. Exactly how much hemolymph circulation occurs through the horn is unknown; however, removal of the horn tip results in immediate release of appreciable quantities of hemolymph, suggesting that at least some circulation is likely.

An alternative mechanisms would involve active heat transfer across other body regions such as from the thorax to the abdomen, as has recently been documented for a species in the same subfamily (Verdu et al., 2004). Here, individuals actively transmit excess heat from the thorax to the abdomen, where it is then given up to the environment. The exact mechanism of heat transfer across tagmata has yet to be documented, but is likely to involve muscle powered directional pumping of hemolymph and/or tracheal air volume (Prange, 1996). If such a mechanism also exists in *Onthophagus*, large individuals, by virtue of their larger size, greater hemolymph, and tracheal volume, and most importantly greater cross-sectional area of muscles, should be more efficient in using this mechanisms to

regulate their temperature than smaller individuals, *unless* the development of an exaggerated secondary sexual trait, such as the enormous horn in *O. nigriventris*, diverts resources away from structures crucial for active heat transfer, such as thoracic muscles. If correct, such a scenario would cause horned males, but not females or hornless males, to end up with the active thermoregulatory capacity of smaller individuals despite their larger size, yet without leaving a signature of this effect during examination of passive warming rates. Tradeoffs arising from the development of horns have already been documented for wings, antennae, and mouthparts (Emlen, 2001), copulatory organs (Moczek and Nijhout, 2004), and testes (Simmons and Emlen, 2006), and preliminary data support the existence of a tradeoff between thoracic horns and flight muscles in *O. nigriventris* (A. Gibbs and A. Moczek, unpublished). Experiments are clearly needed to obtain heat transfer estimates *in vivo*, and to estimate relative tissue composition of male and female body segments as a function of adult body mass. Regardless of the exact outcome of these experiments, however, our data presented here are among the first to show that intra- and interspecific variation in body- and weapon size in horned beetles shape thermal preferences, with the potential to bias behavioral niche partitioning within and between species, and to constrain the further elaboration of exaggerated secondary sexual traits.

Acknowledgments

Appendix A. Supplementary materials

The online version of this article contains additional supplementary data. Please visit doi:10.1016/j.jinsphys.2007.12.007

References

Bartholomew, G.A., Epting, R.J., 1975. Allometry of post-flight cooling rates in moths: a comparison with vertebrate homeotherms. Journal of Experimental Biology 63, 603–613.

Bartholomew, G.A., Heinrich, B., 1978. Endothermy in African dung beetles during flight, ball making, and ball rolling. Journal of Experimental Biology 73, 65–83.

Berwaerts, K., Van Dyck, H., Vints, E., Matthysen, E., 2001. Effect of manipulated wing characteristics and basking posture on thermal properties of the butterfly *Pararge aegeria* (L.). Journal of Zoology 255, 261–267.

Berwaerts, K., Van Dyck, H., Aerts, P., 2002. Does flight morphology relate to flight performance? An experimental test with the butterfly *Pararge aegeria*. Functional Ecology 16, 484–491.

Crespi, B.J., 1988. Adaptation, compromise and constraint: the development, morphometrics, and behavioral basis of a fighter-flier polymorphism in male *Hoplothrips karni* (Insecta: thysanoptera). Behavioral Ecology and Sociobiology 23, 93–104.

Emlen, D.J., 1994. Environmental control of horn length dimorphism in the beetle *Onthophagus acuminatus* (Coleoptera: scarabaeidae). Proceedings of the Royal Society of London B 256, 131–136.

Emlen, D.J., 1996. Artificial selection on horn length–body size allometry in the horned beetle *Onthophagus acuminatus*. Evolution 50, 1219–1230.

Emlen, D.J., 2001. Costs and the diversification of exaggerated animal structures. Science 291, 1534–1536.

Gilchrist, G.W., 1990. The consequences of sexual dimorphism in body size for butterfly flight and thermoregulation. Functional Ecology 4, 475–487.

Halffter, G., Edmonds, W.G., 1982. The Nesting Behavior of Dung Beetles *(Scarabaeinae)*. An Ecological and Evolutive Approach. Instituto de Ecologica Mexico, Federal District, Mexico.

Kingsolver, J.G., Wiernasz, D.C., 1991. Seasonal polyphenism in wing-melanin pattern and thermoregulatory adaptation in Pierid butterflies. American Naturalist 137, 816–830.

Madewell, R., Moczek, A.P., 2006. Horn possession reduces maneuverability in a horn-polyphenic beetle. Journal of Insect Science 6, 21.

May, M.L., 1979. Insect thermoregulation. Annual Review of Entomology 24, 313–349.

Moczek, A.P., 1998. Horn polyphenism in the beetle *Onthophagus taurus*: larval diet quality and plasticity in parental investment determine adult body size and male horn morphology. Behavioral Ecology 9, 636–641.

Moczek, A.P., Emlen, D.J., 1999. Proximate determination of male horn dimorphism in the beetle *Onthophagus taurus* (Coleoptera: scarabaeidae). Journal of Evolutionary Biology 12, 27–37.

Moczek, A.P., Emlen, D.J., 2000. Male horn dimorphism in the scarab beetle *Onthophagus taurus*: do alternative tactics favor alternative phenotypes? Animal Behavior 59, 459–466.

Moczek, A.P., Nijhout, H.F., 2003. Rapid evolution of a polyphenic threshold. Evolution and Development 5, 259–268.

Moczek, A.P., Nijhout, H.F., 2004. Tradeoffs during the development of primary and secondary sexual traits in a dimorphic beetle. American Naturalist 163, 184–191.

Nijhout, H.F., 1994. Insect Hormones. Princeton University Press, Princeton.

Nijhout, H.F., Emlen, D.J., 1998. Competition among body parts in the development and evolution of insect morphology. Proceedings of the National Academy of Sciences 95, 3685–3689.

Nijhout, H.F., Wheeler, D.E., 1982. Juvenile hormone and the physiological basis of insect polymorphisms. Quarterly Review of Biology 57, 109–133.

Pener, M.P., 1991. Locust phase polymorphism and its endocrine relations. Advances in Insect Physiology 23, 1–79.

Prange, H.D., 1996. Evaporative cooling in insects. Journal of Insect Physiology 42, 493–499.

Prange, H.D., Hamilton, G.B., 1992. Humidity selection by thermoregulating grasshoppers. Journal of Thermal Biology 17, 353–355.

Radwan, J., 1993. The adaptive significance of male polymorphism in the acarid mite Caloglyphus berlesei. Behavioral Ecology and Sociobiology 33, 201–208.

Roxburgh, L., Pinshow, B., Prange, H.D., 1996. Temperature regulation by evaporative cooling in a desert grasshopper *Calliptamus barbarus* Ramme 1951. Journal of Thermal Biology 21, 331–337.

Schmidt-Nielsen, K., 1994. Scaling: Why is Body Size so Important. Cambridge University Press, Cambridge.

Schmidt-Nielsen, K., 1997. Animal Physiology: Adaptation and Environment. Cambridge University Press, Cambridge.

Shapiro, A.M., 1976. Seasonal polyphenism. Evolutionary Biology 9, 259–333.

Simmons, L.W., Emlen, D.J., 2006. Evolutionary trade-off between weapons and testes. Proceedings of the National Academy of Sciences 103, 16346–16351.

Van Dyck, H., Matthysen, E., 1998. Thermoregulatory differences between phenotypes in the speckled wood butterfly: hot perchers and cold patrollers? Oecologia 114, 326–334.

Verdu, J.R., Diaz, A., Galante, E., 2004. Thermoregulatory strategies in two closely related sympatric *Scarabaeus* species (Coleoptera: scarabaeinae). Physiological Entomology 29, 32–38.