Individual differences in boldness positively correlate with heart rate in orb-weaving spiders of genus *Larinioides*

Taylor A. SHEARER¹, Jonathan N. PRUITT*
Department of Biological Sciences, University of Pittsburgh, Pittsburgh, Pennsylvania 15260, USA

Abstract Recent years have seen a dramatic increase in the number of studies devoted to consistent individual differences in behavior, termed “behavioral syndromes” or animal “personality”. In particular, ecologists and evolutionary biologists alike seek to explore the functional consequences of individuals’ personalities and explanations for their general maintenance. Understanding the mechanistic underpinnings to personality is arguably key to a deeper understanding of their functional consequences and maintenance. In particular, consistent individual differences in metabolic rate have been proposed as a general explanation for the maintenance of consistent individual differences in boldness, aggressiveness, and activity level. In this paper we explore whether consistent individual differences in boldness are associated with heart rate, a proxy for metabolic rate, in two species of orb-weaving spider: *Larinioides cornutus* and *L. patagiatus*. We first documented consistent individual differences in boldness in *L. cornutus* but not *L. patagiatus*, as estimated by individuals’ latency to resume movement following an aversive stimulus. We also measured individuals’ heart rate twice in a separate situation. We then tested for an association between individuals’ boldness and their heart rate during these two events. We found that increased boldness was associated with increased heart. Thus, individuals that resumed movement quickly following an aversive stimulus also exhibited faster heart rates. Our studies provide further evidence that consistent individual differences in key physiological attributes, like metabolic rate, might be a general mechanism underlying consistent individual differences in behavior [Current Zoology 60(3): 387–391, 2014].

Keywords Araneae, Aggression, Pace of life syndrome, Temperament

Consistent individual differences in behavior that occur across time, situation, or ecological context have received a surge of attention in recent years (Sih et al., 2004; Sih et al., 2012). These “behavioral syndromes” are of ecological significance because of their potential to generate performance trade-offs across contexts (Johnson and Sih, 2005), and because they may provide a general mechanisms for maintaining trait variation within populations (Riechert and Hedrick, 1993). In addition, these consistent individual differences can have enormous influences on higher-order ecological phenomena, ranging from individual fitness (Reale and Festa-Bianchet, 2003; Biro et al., 2004; Biro et al., 2006), to species interactions (Pruitt and Ferrari, 2011; Pruitt et al., 2012a; McGhee et al., in press), to invasion success (Fogarty et al., 2011), or extinction risk (Pruitt, 2012, 2013). Thus, research aimed at exploring the underlying morphological and/or physiological correlates of these differences is of general significant for ecology and evolutionary biology alike. Mechanisms hypothesized to underlie consistent individual differences in behavior include, correlated selection, physical linkage, pleiotropy, neurocirculatory hormones, body state, and individual differences in metabolic rate (McElreath and Strimling, 2006; Bell and Sih, 2007; Biro and Stamps, 2010).

One mechanism that may underlie consistent individual differences in behavior is individuals’ metabolic rate (Careau et al., 2008; Biro and Stamps, 2010). For instance, a more active metabolic rate may require behavioral traits that can keep up with and fuel higher energy expenditures. This means that behaviors linked to exploration, aggressiveness, and boldness should correlate positively with energy metabolism. This relationship of cause and effect, where higher metabolic rates drive the need for more aggressive/bold behavior is termed the Performance Model. In contrast, the Allocation Model argues that behaviors linked to exploration, aggressiveness, and boldness should correlate negatively with metabolic rate. In this model, an organism allocates much of its energy to maintaining a high resting metabolic rate and is thus limited in expending its energy to behaviors associated with activity, and vice versa. This is because any energy allocated to one need...
(metabolic rate or personality) cannot be spent on the other. Thus, these models predict opposing correlations between individuals’ metabolic rate and their aggressiveness, boldness, exploration, and/or activity level. A third less-frequently considered alternative is that activity-levels and/or aggressiveness are the driver of metabolic rate, since individuals with higher activity levels will tend to have higher metabolic rates, since they spend more of their time being active. This alternative again predicts a positive association between individuals’ metabolic rate and their aggressiveness, boldness, and/or activity level. One of the most common methods of estimating individuals metabolic rate is through the measurement of heart and/or respiration rate (Carere and van Oers, 2004; McGhee et al., in press), which is the approach we take in this study.

Here we consider whether there is an association between boldness and heart rate in the spiders Larinioides cornutus and L. patagiatus (Araneae, Araneidae). Spiders have long served as a model for behavioral syndromes research. Behavioral syndromes have been noted in more than twenty species in ten families (Pruitt and Riechert, 2012; Pruitt et al., 2013), and generally involve an association between high exploratory behavior, activity level, aggressiveness, boldness, and low sociability (Riechert and Hedrick, 1993; Johnson and Sih, 2007; Pruitt et al., 2008; Kralj-Fiser and Schneider, 2012). Here we test whether spiders, like various species of vertebrate, exhibit a significant association between their behavioral tendencies (i.e., boldness) and their heart rate under a standardized conditions. Specifically, we ask the following questions: (I) Do Larinioides exhibit consistent individual differences in boldness? (II) If yes, are consistent individual differences associated with body size or body condition? (III) Is boldness associate with individuals’ heart rate under standardized laboratory conditions?

1 Materials and Methods

1.1 Collection & laboratory maintenance

Adult Larinioides cornutus (n = 29) and Larinioides patagiatus (n = 9) were collected from around housing structures on the western side of Gibraltar Island in the Bass Island region of Lake Erie. We conducted our assays in an Ohio State University teaching laboratory space which lacked air conditioning between 1200–1600 hr each day (27.7–32°C). Our experiments took place during May 2013, when both adult males and females were present, and our assays occurred one day after a mayfly emergence. Thus, spiders received an ad libitum meal of mayflies immediately before collection. After 24 hours in captivity, the mass of each spider was measured on a digital balance to the nearest hundredth of a gram and their prosoma width was measured using digital calipers to the nearest tenth of a millimeter. Spiders were housed individually in strawberry containers (15 cm × 15 cm × 13 cm) between behavioral assays.

1.2 Boldness assay: latency to resume movement

To assess spiders’ boldness towards predator cues, we use a “puff test” following the protocol of Riechert and Hedrick (1993). Similar protocols have been utilized in numerous studies on web-building spiders (Kralj-Fiser and Schneider, 2012; Grinsted et al., 2013; Pruitt et al., 2013). Boldness assays occurred 6h after weighing and measuring spiders and again 24 h after that. Trials are initiated by lifting off the lid to spiders’ containers and moving them to a novel open field environment. The open field environment was a 50 cm × 50 cm square plastic enclosure lined with computer paper, which was replaced between trials. Spiders were permitted 60s acclimation time before the application of an aversive stimulus. We then “puffed” the spider’s anterior, dorsal side with two rapid jets of air using an infant ear-cleaning bulb. Spiders universally responded by huddling, with their legs drawn toward their cephalothorax. Boldness was estimated as the time taken for spiders to resume movement following this cue. Spiders with shorter latencies to resume movement are deemed more “bold”, whereas spiders with longer latencies are termed more “shy”. Trials ended when the spider initiated movements or after 10 minutes, whichever happened first. To obtain more intuitive values for “boldness”, individuals’ latencies to resume movement in seconds were subtracted from the maximum value of 600 seconds. Thus, a greater latency to resume movement corresponds with a smaller boldness value.

1.3 Heart rate

Spiders’ heart rates were measured 2 hours after spiders’ second boldness measure. Trials were initiated by removing individuals from their home containers and transferring them to a clear, plastic, circular enclosure (radius=4 cm), with their dorsal side up. This container was then placed beneath a dissecting microscope (Leica M80) and the spiders were given 60 seconds to acclimate. We then counted the number of heart beats that the spiders exhibited over a 15 second interval, waited 60 seconds longer, and measured their heart rate for another 15 seconds. In spiders, the heart runs along the dorsal anterior portion of the abdomen back posteriorly. Individual heart beats were evidenced by the acute ex-
pansion and retraction of the pigmented cells on the dorsal anterior portion of the abdomen (Carrel and Heathcote, 1976, Bromhall, 1987). The number of beats over 15 seconds was used to calculate the number of beats per minute, and the first and second measures (separated by 60 seconds) were compared in order to confirm that individuals had reached a relatively stable heart rate during our procedure. We assume that our protocols were stressful to the animal, and thus, we interpret our heart rate measure as having occurred during an acutely stressful event.

1.4 Statistical methods

To assess repeatability in heart rate and individual boldness we used nested ANOVAs to partition variances into the proportion of variation explained by within-versus between individual differences. Repeatability is defined herein as the proportion of total variation explained by between individual differences (Boake, 1989; Falconer and McKay, 1996). We tested the repeatabilities for each species independently. To test for an association between boldness and heart rate we performed species-wise linear mixed effect models, where individual intercept and slope were included and only the average metabolic rate was used to explain boldness.

2 Results

We detected significant repeatabilities of both boldness and heart rate in L. cornutus but not L. patagiatus: L. cornutus boldness ($F_{28,29} = 2.64, r = 0.77, P = 0.006$) and heart rate ($F_{28,29} = 5.23, r = 0.84, P<0.0001$), L. patagiatus boldness ($F_{8,9} = 1.31, r = 0.53, P = 0.35$) and heart rate ($F_{8,9} = 2.70, r = 0.71, P = 0.08$). And, we detected no significant differences in individuals’ heart rates in our first versus second measurement ($t = 0.08$, $df = 56, P = 0.77$). The two species also differed in their average boldness, where L. cornutus was 200%–400% bolder than L. patagiatus ($F_{1,66.11} = 1.31, R^2 = 0.62, P = 0.009$).

Our species-wise linear mixed effect models detected a strong positive correlation between boldness and heart rate in L. cornutus ($F_{1,27} = 14.73, R^2 = 0.56, P<0.0001$, Fig. 1). However, we failed to detect an association between boldness and heart rate in L. patagiatus ($F_{1,7} = 1.31, R^2 = 0.20, P = 0.28$, Fig. 1), perhaps owing to low sample sizes.

3 Discussion

In this study, the heart rates of individual L. cornutus of differing degrees of boldness were compared by measuring their heart rates during an acute, stressful event. First, we found that our estimates of individuals’ boldness were highly repeatable in L. cornutus, consistent with data collected on dozens of other species of spider (Pruitt and Riechert 2012). Second, we found that our two species differed in their average boldness. Third, we found a strong, positive association between individuals’ boldness and their heart rates for L. cornutus. Thus, we reason that individual differences in key metabolic functions (e.g., metabolic rate, heart rate) may underlie individual differences in boldness in these species.

Inter-individual variation in boldness was found to be repeatable in L. cornutus. Consistent individual differences in boldness are widespread among many species of spiders, and the repeatability estimates of L. cornutus ($r = 0.77$) are of a similar magnitude to those obtained from other species of web-building spiders (Bell et al., 2009; Pruitt and Riechert, 2012). This is a fortuitous result for the use of L. cornutus as a general model spider for the studies herein, because the strength of the signature of personality in this species is typical for these animals. However, we failed to detect a significant repeatability for L. patagiatus, presumably due to very low sample sizes ($n = 9$). Though, even for this species, our estimate of repeatability was high as compared to most study systems ($r = 0.53$).

Two commonly used models for explaining the connection between behavior and metabolic rate are the Performance Model, which predicts a positive correlation between metabolic rate and boldness, aggressiveness, and activity-level, and the Allocations Model, which predicts negative correlations (Careau et al., 2008; Biro and Stamps, 2010; Reale et al., 2010). Results from this experiment reveal a positive correlation between
boldness and heart rate for *L. cornutus*, which is consistent with the Performance Model. This model argues that high metabolic rates actually require than animals exhibit greater boldness, aggressiveness, and activity-level in order to procure the resources required to sustain their metabolisms. This result is intriguing, because it may provide a mechanism for maintaining variation in personality and metabolic rates within populations, because of spatiotemporal variation in resource availability. Specifically, we argue that metabolic rates may be favorable under high resource conditions, because boldness, aggressiveness, and activity-level can be amply fueled and could even be advantageous for securing resources against rivals. Consistent with this hypothesis, boldness has been associated the ability to secure resources in numerous species of spiders (Pruitt et al., 2008; Pruitt and Riechert, 2009; Pruitt et al., 2012b).

However, spiders’ foraging ecology is characterized by boom-and-bust, feast-or-famine resource dynamics, where most individuals suffer extended periods of starvation between foraging bouts (Foelix, 1996; Maupin and Riechert, 2001). Under these conditions, we reason excessively high metabolic rates may limit individuals’ ability to survive these intervening periods, and instead favor individuals with lower metabolic rates. Consistent with this interpretation, studies on the spider *A. aperta* have found that aggressive individuals are less selective in terms of the prey they accept, potentially reflecting the need to fuel a higher metabolic rates for these individuals (Riechert, 1991).

The argument that some phenotypes will be favored because of their low resource needs bears thematic resemblance to classic resource competition theory (i.e., R*)(Tilman, 1982). Importantly, the success of R* Theory as a general explanation for competitive dominance in community ecology has been marred by insufficient field tests. Thus, we caution that although the ideas outlined above have intuitive appeal, future studies that orthogonally manipulate resource availability, population density, and ratios of different personality types are badly needed to address whether these models hold true. As a growing number of studies document basic associations between metabolic traits and personality (like this one), the most important, illuminating studies will be those that directly and explicitly test these models’ predictions either in laboratory mesocosms or (preferably) in situ. For our study, we must recognize that all of our measurements were taken immediately following an intense feeding bout. It, thus, remains unclear whether the relationship between boldness and heart rate observed here would hold under severe food limitation. Although, the idea that the relationship between basic physiological measurements and animal personality will emerge/disappear as a consequence of system’s resource regimes is itself an exciting and little-explored hypothesis.

**Acknowledgements** We are indebted to Andreas Modlmeier, C. Nick Keiser, Colin Wright, Matthias Foellmer, the executive editor Zhi-Yun Jia and two anonymous reviewers whose comments improved the quality of this manuscript.

**References**

Bell AM, Hankison SJ, Laskowski KL, 2009. The repeatability of behaviour: A meta-analysis. Animal Behaviour 77: 771–783.

Bell AM, Sih A, 2007. Exposure to predation generates personality in threespined sticklebacks *Gasterosteus aculeatus*. Ecology Letters. 10: 828–834.

Biro PA, Abrahams MV, Post JR, Parkinson EA, 2004. Predators select against high growth rates and risk-taking behaviour in domestic trout populations. Proceedings of the Royal Society Biological Sciences Series B. 271: 2233–2237.

Biro PA, Abrahams MV, Post JR, Parkinson EA, 2006. Behavioural trade-offs between growth and mortality explain evolution of submaximal growth rates. Journal of Animal Ecology 75: 1165–1171.

Biro PA, Stamps JA, 2010. Do consistent individual differences in metabolic rate promote consistent individual differences in behavior? Trends in Ecology & Evolution 25: 653–659.

Boake CRB, 1989. Repeatability: Its role in evolutionary studies of mating-behavior. Evolutionary Ecology 3: 173–182.

Bromhall C, 1987. Spider heart-rates and locomotion. Journal of Comparative Physiology B-Biochemical Systemic and Environmental Physiology 157: 451–460.

Careau V, Thomas D, Humphries MM, Reale D, 2008. Energy metabolism and animal personality. Oikos 117: 641–653.

Carrel JE, Heathcote RD, 1976. Heart-rate in spiders: Influence of body size and foraging energetics. Science 193: 148–150.

Falconer DS, McKay TF, 1996. Introduction to quantitative genetics. Longman, New York: Prentice Hall.

Foelix RF, 1996. Biology of Spiders. New York: Oxford University Press.

Fogarty S, Cote J, Sih A, 2011. Social personality polymorphism and the spread of invasive species: A model. American Naturalist 177: 273–287.

Grinsted L, Pruitt JN, Settepani V, Bilde T, 2013. Individual personalities drive task differentiation in a social spider. Proceedings of the Royal Society B-Biological Sciences 280: 20131407.

Johnson JC, Sih A, 2005. Precopulatory sexual cannibalism in fishing spiders *Dolomedes triton*: A role for behavioral syndromes. Behavioral Ecology and Sociobiology 58: 390–396.

Johnson JC, Sih A, 2007. Fear, food, sex and parental care: A syndrome of boldness in the fishing spider *Dolomedes triton*. Animal Behaviour 74: 1131–1138.

Kralj-Fiser S, Schneider JM, 2012. Individual behavioural consis-
tency and plasticity in an urban spider. Animal Behaviour 84: 197–204.

Maupin JL, Riechert SE, 2001. Superfluous killing in spiders: A consequence of adaptation to food-limited environments? Behavioral Ecology 12: 569–576.

McElreath R, Strimling P, 2006. How noisy information and individual asymmetries can make 'personality' an adaptation: A simple model. Animal Behaviour 72: 1135–1139.

McGhee KE, Pintor LM, Bell AM, in press. Reciprocal behavioral plasticity and behavioral types during predator-prey interactions. The American Naturalist.

Pruitt JN, 2013. A real-time eco-evolutionary dead-end strategy is mediated by the traits of lineage progenitors and interactions with colony invaders. Ecology Letters 16: 879–886.

Pruitt JN, Ferrari MCO, 2011. Intraspecific trait variants determine the nature of interspecific interactions in habitat forming species. Ecology 92: 1902–1908.

Pruitt JN, Grinsted L, Settepani V, 2013. Linking levels of personality: Colony-level personality is associated with the average body size and behavioral tendencies of the colony constituents. Animal Behaviour.

Pruitt JN, Riechert SE, 2012. The ecological consequences of temperament in spiders. Current Zoology 58: 589–596.

Pruitt JN, Riechert SE, Jones TC, 2008. Behavioural syndromes and their fitness consequences in a socially polymorphic spider Anelosimus studiosus. Animal Behaviour 76: 871–879.

Reale D, Garant D, Humphries MM, Bergeron P, Careau V et al., 2010. Personality and the emergence of the pace-of-life syndrome concept at the population level. Philosophical Transactions of the Royal Society B-Biological Sciences 365: 4051–4063.

Riechert SE, 1991. Prey abundance vs diet breadth in a spider test system. Evolutionary Ecology 5: 327–338.

Riechert SE, Hedrick AV, 1993. A test for correlations among fitness-linked behavioral traits in the spider Agelenopsis aperta (Araneae, Agelenidae). Animal Behaviour 46: 669–675.

Sih A, Bell A, Johnson JC, 2004. Behavioral syndromes: An ecological and evolutionary overview. Trends in Ecology & Evolution 19: 372–378.

Sih A, Cote J, Evans M, Fogarty S, Pruitt JN, 2012. Ecological implications of behavioral syndromes. Ecology Letters 15: 278–289.

Tilman D, 1982. Resource competition and community structure. Princeton: Princeton University Press.