



# Thermoregulatory behavior and orientation preference in bearded dragons



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## ABSTRACT

The regulation of body temperature is a critical function for animals. Although reliant on ambient temperature as a heat source, reptiles, and especially lizards, make use of multiple voluntary and involuntary behaviors to thermoregulate, including postural changes in body orientation, either toward or away from solar sources of heat. This thermal orientation may also result from a thermoregulatory drive to maintain precise control over cranial temperatures or a rostrally-driven sensory bias. The purpose of this work was to examine thermal orientation behavior in adult and neonatal bearded dragons (*Pogona vitticeps*), to ascertain its prevalence across different life stages within a laboratory situation and its interaction with behavioral thermoregulation. Both adult and neonatal bearded dragons were placed in a thermal gradient and allowed to voluntarily select temperatures for up to 8 h to observe the presence and development of a thermoregulatory orientation preference. Both adult and neonatal dragons displayed a non-random orientation, preferring to face toward a heat source while achieving mean thermal preferences of ~ 33–34 °C. Specifically, adult dragons were more likely to face a heat source when at cooler ambient temperatures and less likely at warmer temperatures, suggesting that orientation behavior counter-balances local selected temperatures but contributes to their thermoregulatory response. Neonates were also more likely to select cooler temperatures when facing a heat source, but required more experience before this orientation behavior emerged. Combined, these results demonstrate the importance of orientation to behavioral thermoregulation in multiple life stages of bearded dragons.

## 1. Introduction

Animals commit time and energy to achieve and maintain thermally optimality, defined as the range of temperatures which maintains physiological processes operating at, or near, maximum conditions (Dewitt, 1967; Huey and Slatkin, 1976). For ectotherms which depend on environmental heat absorption, behaviors that adjust the rate of body temperature ( $T_b$ ) change are crucial to thermoregulation (Cowles and Bogert, 1944). Although thermoregulatory behaviors are known to present during early life (Blumberg et al., 2002; Lang, 1981; Stahlschmidt et al., 2015; Vollset et al., 2013; Zhao et al., 2013), how thermoregulatory behaviors change through ontogeny is not well studied. Shuttling and basking behaviors, which have a high impact on body temperature, are likely present from an animal's first exposure to a novel thermal environment, although these behaviors are also subject to change with experience. For example, shuttling behaviors show lower precision in bearded dragons that are naïve to an operant conditioning thermoregulatory paradigm compared to those with prior experience or when the locomotory costs of thermoregulation are increased (Cadena and Tattersall, 2009). Indeed, that lizards dedicate time to exploring and adjusting to a thermal gradient in the lab suggests that learning plays a role in behavioral thermoregulation, especially in

novel environments (Cadena and Tattersall, 2009). Subtle thermoregulatory responses might, therefore, also require time and may even need to be learned well after hatching.

Unlike birds and mammals, squamates do not typically display parental care behaviors after their young hatch (Reynolds et al., 2002). After hatching, neonates meet their needs on their own, but not all behaviors are present during every phase of an animal's life (Dawkins, 1995; Khan et al., 2010). Innate behaviors are responses that prepare an animal for adaptive reactions to the world around them, and are generally fully formed from the outset without the need for experience or learning; this is also referred to as inbuilt adaptiveness (Dawkins, 1995). Learned behaviors are responses that develop through the accumulation of experience and the retention of information (Barnard, 2003). For the purposes of this work, innate thermoregulatory behaviors are those present from the neonatal stage that do not generally require experience. Examples of both innate and learned responses can be found in voluntary, involuntary, and autonomic behaviors (Dawkins, 1995), and thus thermoregulatory behaviors can be expected to reflect both innate and learned responses.

Aside from morphological and developmental differences, neonatal bearded dragons differ from their adult counterparts, especially with respect to social behaviors (Khan et al., 2010). Neonates are more prone

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to gregarious behavior, and often observed in aggregations (Khan et al., 2010). Gregarious behavior, while common in mammals (Aureli et al., 2002), is less prevalent in reptiles and is not necessarily driven by sociality. Instead, aggregation could be related to other fitness demands, such as gestation, oviposition, parasitism, predation protection, or even thermoregulation (Gautier et al., 2006; Graves and Duvall, 1995; Wikelski, 1999). When solitary and groups of neonatal bearded dragons were allowed to thermoregulate, both isolated and aggregated neonates were observed to select similar temperatures, however, individuals within a group context showed lower thermoregulatory precision than those in isolation (Khan et al., 2010). Khan et al. (2010) suggested that this aggregation results from a mutual attraction to a limited and valuable resource (e.g., an optimal temperature), and that the drop in precision results from agonistic interactions (Khan et al., 2010). Agonistic behaviors over access to preferred temperatures are known to occur in other ectothermic species, and are also dependent on familiarity and experience (Tattersall et al., 2012b), suggesting that both social context and learning contribute to thermoregulatory behaviors. In short, although thermoregulatory behaviors are guided by innate, sensory driven mechanisms, they can be shaped and influenced by experience.

Due to the importance of temperature regulation, it is plausible that thermoregulatory behaviors will show some level of innateness and therefore be conserved between adults and neonates. Behaviors such as aggregation are likely to be lost at the adult stages due to increased competition between individuals that have reached sexual maturity. Critical thermoregulatory behaviors, such as shuttling and basking are likely observed across all life stages. Subtler, yet equally important thermoregulatory behaviors, such as body orientation toward a heat source (referred to hereafter as orientation preference), may instead be partially learned through growth and development, since body size is strongly implicated in the effectiveness of orientation (Lactin and Johnson, 1997). Orientation preference, as a thermoregulatory behavior, has been documented in select mammals as an energy savings strategy (Hetem et al., 2011), but most notably in several invertebrates and certain reptiles (Bartholomew, 1966; Fraser et al., 2016; Martin et al., 1995; McMaster and Downs, 2006; Tattersall et al., 2012b). Adult lizards of a species well known to exhibit distinct thermoregulatory behaviors, such as the bearded dragon, are the best option for examining the presence of any orientation preference, as they are larger than neonates and therefore orientation is more likely to have a direct impact on heating and cooling rates, and therefore thermoregulation. Additionally, adults have had time to acquire any learned aspects of the behavior. On the other hand, cutaneous thermosensory feedback may be over-emphasized in the cephalic region, resulting in an innate, sensory bias-based preference to face toward or away from the source of the stimulus. Ion channels implicated in thermotaxis are highly expressed in the rostral margin of the head in flatworms, compared to lateral margins or the rest of the body (Inoue et al., 2014), emphasizing the role of rostral sensory information to locomotion in bilateral animals. Furthermore, sensory representation and organization within the somatosensory cortex often reflects adaptive functionality and may contribute to enhanced sensory capacity (Catania, 2005); whether similar overrepresentation with respect to temperature exists in an animal that relies heavily on thermosensation is unknown.

This work seeks to address the presence of a thermoregulatory orientation bias in bearded dragons. By observing orientation behavior at an early and mature life stage, a better understanding of the innateness of orientation as a thermoregulatory strategy will be gained. We hypothesized that orientation would present as a thermoregulatory mechanism in bearded dragons. Therefore, we expected that bearded dragons would display non-random orientation behavior in a thermal gradient, choosing to face towards a heat source rather than towards a cold source. We also hypothesized that a preference for orienting towards a heat source would be conserved across multiple life stages, which would support an innate hypothesis for orientation behavior.

Since orientation also informs about the direction in which an animal has been moving while selecting preferred temperatures, we examined orientation in combination with movement and selected temperature in a thermal gradient.

## 2. Methods

### 2.1. Animal husbandry

All animals were captive reared and kept at Brock University in an environmentally controlled room dedicated exclusively to bearded dragons (*Pogona vitticeps*) and a small cockroach colony. A total of 43 dragons were maintained and used during this time. Of the 43 dragons, 17 were adults (10 Male and 7 Female of 1–4 years of age) and 26 were neonates (less than 2 weeks old). Each adult dragon was housed singly in a terrarium (custom built by Brock University Machine Shop; dimensions 76 cm × 76 cm × 42 cm) with either a corn cob or a coconut husk bedding and equipped with a 40 W light bulb set over a stone basking plate, which provided a maximum temperature of 45 °C compared to a minimum temperature of 25 °C at the far side of the terrarium. In addition to a standard 40 W light bulb, a UV light (13 W Reptisun® 10.0 mini compact fluorescent) was also installed to provide UV required for vitamin D synthesis. All cages were given extra enrichment in the form of cardboard packing material, which was used for shelter or as a climbing surface, and Polyvinyl chloride (PVC) pipe sections, used as hiding spots. The neonatal animals were kept in small cages until they grew large enough to move to an adult cage. These smaller terraria were 45 cm × 24 cm × 20 cm and had paper towel bedding. Heating pads were laid down under the cages and set to provide half of each enclosure with floor temperatures from 25 to 45 °C. Smaller PVC pipe sections were also supplied as additional enrichment. In addition, neonates were placed two to a cage unless they showed aggressive behaviors, e.g., biting or attacking cage mates, in which case they were placed in individual cages.

All lizards were under a 12:12 light:dark cycle, such that cage temperature declined to 24 °C in the dark. Animals were fed a diet of assorted chopped fruits and vegetables which they ate *ad libitum* and which were replaced three times a week and were fed cockroaches once or twice a week. Once a week all lizards received a water bath for sanitation and hydration.

### 2.2. Series I: Thermal preference in adult bearded dragons

Thermal and orientation preferences were determined using video footage and temperature sensors. During Series I (see below), data loggers (iButton Thermochron™) were affixed to the lizards and gave a measurement of ambient/skin temperature preference. All iButtons were calibrated to the computer's clock. Each iButton was set to record every 30 s starting immediately. Once the iButtons was programed, it was attached to the lizard using 3 M Transpore™ tape, to the ventral side of the abdomen.

To examine orientation and any other behaviors, animals were placed in a thermal gradient. The thermal gradient had two water baths connected to either end to create a range of temperatures, from 15 °C to 45 °C along the floor of the experimental chamber (~ 0.18 °C/cm). In addition to the floor, an air gradient was also created by using fans, connected to radiators receiving water from the water baths, at either end of the sheet. The gradient had walls set 18 cm apart and 10 cm high while the length of the gradient itself was 163 cm. This created a somewhat narrow lane that encouraged each individual lizard to choose a direction to face during the experiment, either cold or hot. The animal was then placed in the center of the gradient facing either left or right, and was allowed to move freely. When placing animals in the gradient, the direction they were facing was randomly alternated, as was the warm and cold sides of the gradient itself.

To ensure a more consistent air gradient within the experiment,

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