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Different incubation temperatures result in differences in mass in female red-eared slider turtle hatchlings

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Abstract

The Charnov–Bull model states that environmentally determined sex will prevail in patchy environments where males may fare best in one patch type, whereas females may fare well in a different patch type. To investigate whether or not potential differences manifest early in the life of a turtle with temperature-dependent sex determination, I assessed mass, carapace width and length, and plastron length of hatchlings from three different incubation temperature regimes. Differences in incubation temperature affected mass in turtles of the same sex; the difference appears to support a sex-based rationale for the phenotype. The results bear out an assumption of Charnov–Bull that neonates of the same sex from different temperatures may manifest different attributes (i.e., mass) that could affect their relative fitness.

Keywords: Temperature-dependent sex determination; Reptile; Development; Fitness

1. Introduction

All crocodilians, many turtles, the tuatara, and some lizards employ temperature as a cue for sex determination. Reptiles have served as the primary focus of studies of temperature-dependent sex determination and are often used as the animal model for testing hypotheses to explain its persistence. Probably the best-known model to explain the persistence of environmental sex determination is the Charnov–Bull model (Charnov and Bull, 1977), which predicts that environmental will be favored over chromosomal determination when an organism in early life benefits by being male under some conditions and female under others. Under this model, an organism inhabits a patchy environment in which individuals in some patches fare better than individuals in others, and the differential success is related to the sex of the organism. Thus, an offspring's entrance into a given patch must influence male fitness differentially compared to female fitness.

In some differential-fitness assessments using reptiles, temperature and gonadal sex interact to produce sex-specific phenotypes that affect fitness (Tousignant and Crews, 1995; Rhen and Crews, 1999; Reece et al., 2002); that is, instead of temperature's being the only influence, gonadal sex and temperature both exert influence on phenotype, supporting the Charnov–Bull model. The result is that although two sexes may occur under a single incubation temperature regime, they will exhibit different phenotypes as a result of the interaction of sex and temperature.

To assess the interaction of sex and phenotype in a different design, I examined the potential for different incubation regimes to exert differential effects in turtles

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of the same sex, measuring mass, carapace length and width, and plastron length in the red-eared slider turtle, *Trachemys scripta elegans*, a species with temperature-dependent sex determination. In this species, low incubation temperatures result in males, mid-range temperatures result in mixed-sex ratios, and higher temperatures cause female development (Crews, 1996). Instead of producing two sexes under one temperature regime, this study examined the effects of different temperature regimes on the same sex, looking at parameters in earliest life.

2. Materials and Methods

Turtle eggs were laid and collected the same day and incubated at the commercial turtle supplier (Kliebert Alligator and Turtle Farms, Hammond, LA) at 29.4 °C for 17d before being picked up and transported back to the laboratory. At all times, eggs underwent the same protocols and processing procedures. Using a random placement approach, I placed eggs in plastic boxes on beds of vermiculite:water (w/v; g mL), bagged the boxes loosely in plastic, and placed them in incubators (Brinsea, Titusville, Fla.) at one of three temperatures, 26 °C, 29.2 °C, or 31 °C. Because eggs were kept at 29.4 °C until stage 17 (Yntema 1968), the beginning of the temperature-sensitive period, some females were produced at a temperature that normally produces all males (26 °C), providing a comparison to females from higher temperatures. At 26 °C, the temperature-sensitive period lasts until about stage 21; it ends at about stage 19 at 31 °C.

Eggs were incubated to hatching; during incubation, temperatures were checked daily by digital readout on the incubators and by in-incubator thermometers, and boxes were rotated and turned daily to avoid temperature-gradient effects.

Within 24 h of hatching, each turtle was measured; mass was taken to 0.1 g, and carapace length and width and plastron length were taken using digital calipers. Turtles were killed via rapid decapitation and gonadal sex assessed using a dissection scope.

Results were analyzed using JMP (1989–2002) for Windows. Determinations were made about whether or not data for a group were normally distributed; if they were, then ANOVA was performed to compare groups. For data not normally distributed, the nonparametric Kruskal–Wallis was used. A full analysis comparing interactions of sex and each parameter was performed, as was an analysis of each sex vs. the same sex from another group; for example, females from 31 °C were compared to females from 26 °C. Sex ratio assessments were made using Fisher's exact analysis of two-by-two contingency tables.

3. Results

Turtles from 29.2 °C were significantly larger than those from 31 °C (ANOVA, df = 1; $F = 4.35$; $p = 0.04$) (Fig. 1A). Further investigation comparing only females from each group resulted in a significant difference; females from the 29.2 °C group (n = 15) were larger than females from the 31 °C group ($n = 28$) (Kruskal–Wallis test, $X2 = 4.7$, df = 1; $p = 0.03$) (Fig. 1B). No other comparison yielded a significant effect; e.g., females or males from 26 °C were no different from females or males in any other group, and there was no interaction of sex with any other parameter.

As expected, 31°C produced a significantly greater percentage of females (100 percent) than either of the lower temperatures ($p < 0.001$); the 26 and 29.2 °C groups did not differ significantly in number of females produced (37 and 52 percent, respectively).

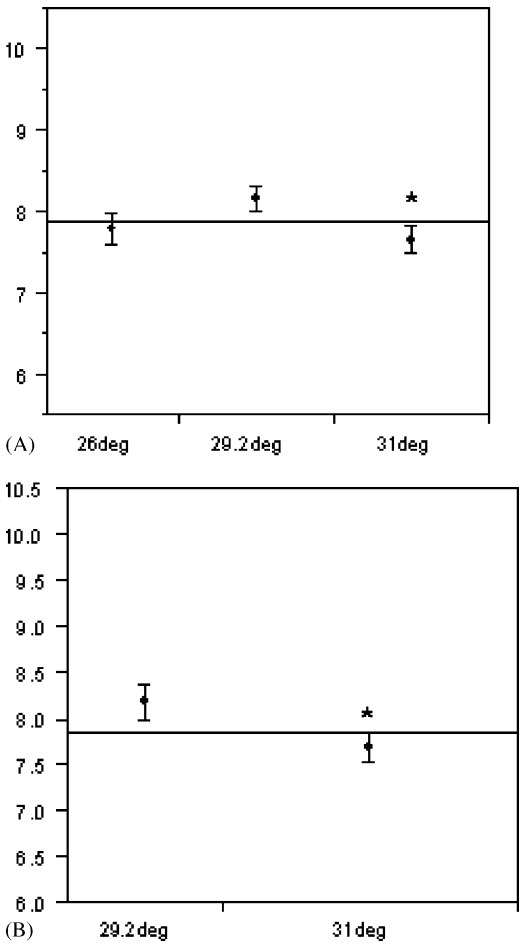

Fig. 1. (A) Mass of turtles from each temperature regime (26 °C, 29.2 °C, and 31 °C). (B) Mass of females from 29.2 °C and 31 °C. * indicates significant difference compared to 29.2 °C.