

The impact of variable egg incubation regimes on hatchling American alligator genital differentiation

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Abstract: Adult crocodylian genital morphology substantially differs by sex. However, the sexually dimorphic differences of clitero-penis (CTP) anatomy are subtle at hatching, with male genitals slightly larger than female. While egg incubation temperature determines American alligator (*Alligator mississippiensis*) gonadal sex, genitalia development (either clitoris or penis outcome) is predominantly delayed until post-hatching and is androgen-dependent. A constant 33°C nest incubation temperature determines testis differentiation in *A. mississippiensis* and subsequent male penis development. In contrast, 30°C results in ovary development and clitoral formation. However, it is unclear how intermediate and/or fluctuating incubation temperatures influence external genitalia differentiations and development. We investigated this question in week-old hatchlings incubated under the following conditions: 1) constant male producing temperature (33°C), 2) constant female producing temperature (30°C), 3) intermediate promoting temperature (31.2°C), and 4) intermediate promoting fluctuating temperatures one (31.2°C +/- 0.6°C) and 4) intermediate promoting fluctuating temperature two (31.2 +/- 2.8°C). To better characterize CTP morphological differences across incubation temperature regimes, we quantified: linear measures- lateral and ventral CTP dimensions of glands height (maximum vertical width at mid-section of the head), glands width (widest point of glands), and curve distance (from beginning of sulcus to end of tip)- and the geometric morphometry of landmarks and semi-landmarks of the CTP lateral aspect. We compared these results to total body morphometrics and circulating steroid hormone concentrations at tissue collection. The results we present here expand our understanding of “normal” alligator CTP sexual dimorphisms. Our project illustrates how the environment intimately shapes crocodylian biology and increases our ability to detect altered development in wild populations.

Keywords: Genitalia, Sexual dimorphism, American Alligator, Clitero-penis

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