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## Complete mitochondrial genomes for three lizards (*Anolis punctatus*, *Sceloporus woodi*, and *S. grammicus*): a contribution to mitochondrial phylogenomics of Iguanoidea

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### ABSTRACT

The complete mitochondrial genomes of the Spotted Anole (*Anolis punctatus*), Florida Scrub Lizard (*Sceloporus woodi*), and Mesquite Lizard (*S. grammicus*) were assembled and annotated. Genomic DNA of *A. punctatus* was sequenced in one-sixth of a lane in an Illumina HiSeq while comparable sequencing data for *S. woodi* and *S. grammicus* were retrieved from the Short Read Archive (entries SRR1286259 and SRR1145766, respectively). We assembled these mitogenomes using MIRA, NovoPlasty, and/or MITObim software. The resulting whole and circularized mitogenome assemblies exhibited typical vertebrate sequence length and gene organization. We deposited these mitogenomes into public databases under accession numbers MK091854, BK010487, and BK010486, respectively. To verify the taxonomic identifications of our mitogenomes (as iguanian lizards), we conducted maximum likelihood analyses with 14 published mitogenomes from the lizard superfamily Iguanoidea and two outgroups. Our analysis of concatenated gene sequences from mitochondria confirmed the taxonomic identities of our mitogenomes to the genus level.

### ARTICLE HISTORY

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Anolis lizards; *Sceloporus* lizards; mitochondrial genome; Iguanoidea; phylogenomics; mitogenome

With more than 400 species described, the genus *Anolis* represents one of the most speciose vertebrate genera (Losos 2009). However, to date, only one complete mitogenome is available for *Anolis*. The Spotted Anole, *Anolis punctatus* is widespread in Amazonia and coastal Atlantic Forests of South America (Ribeiro-Júnior 2015; Prates et al. 2016). The sample sequenced here (MNRJ 23455) was obtained from the reptile collection at the Museu Nacional, Universidade Federal do Rio de Janeiro. The partial genome of *A. punctatus* was sequenced in one-sixth of a lane in an Illumina HiSeq and produced ~50 million 100-bp paired-end sequencing reads. Spiny lizards from the genus *Sceloporus* are represented by ~90 species in North America (Leaché 2010). Like Anoles, *Sceloporus* is only represented by one mitogenome in Genbank. We assembled the mitogenome of the Florida Scrub Lizard (*Sceloporus woodi*) endemic to Florida (Demarco 1992) and the Mesquite Lizard *Sceloporus grammicus* from Northern and Central Mexico (Arévalo et al. 1991; Ochoa-Ochoa and Vilella 2006). Illumina datasets were downloaded from the Short Read Archive (SRR1286259 for *S. woodi* and SRR1145766 for *S. grammicus*) originally obtained from specimens UWBM 7265 and UWBM 6585 in the Burke Museum of

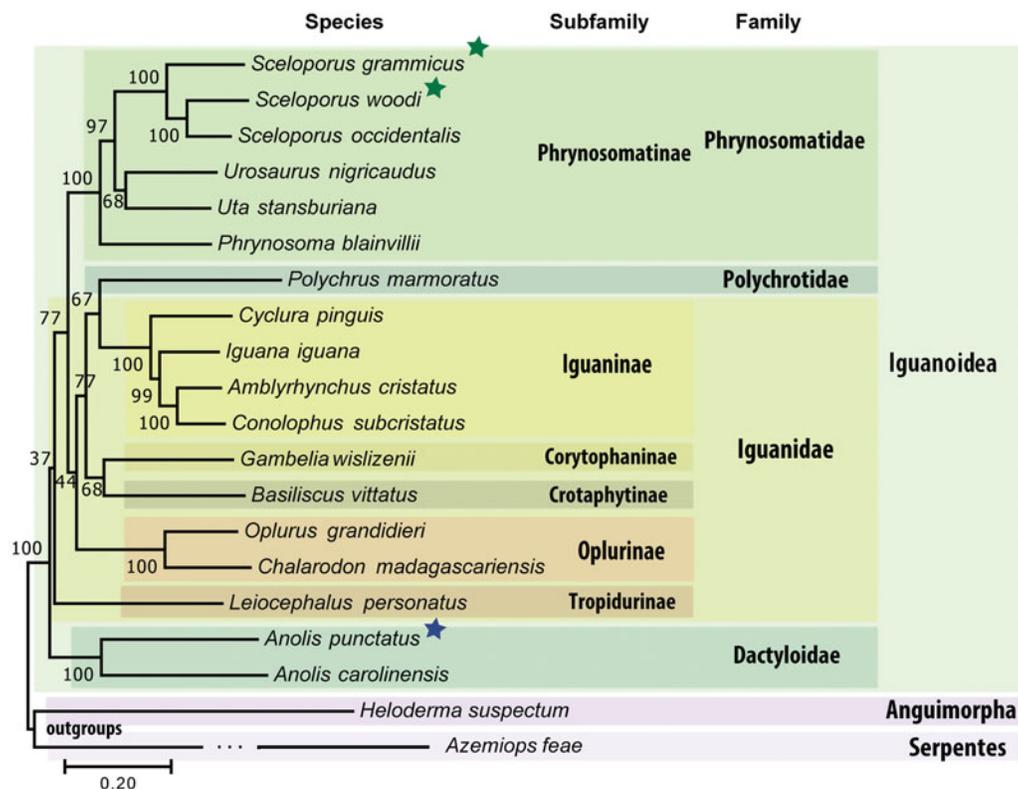
Natural History and Culture, University of Washington (Leaché et al. 2013; Genomic Resources Development Consortium 2015). For the Spotted Anole, a *de novo* assembly using MIRA software (Chevreux et al. 1999) produced a nearly complete version for the mitogenome. An additional assembly step was needed to confirm circularization. The complete *A. punctatus* mitochondrial genome is 17,132 bp and was produced using 18,623 sequencing reads with an average read coverage of 91× according to Tablet software (Milne et al. 2013). For *S. woodi*, the mitogenome was produced using a reference-based MIRA job guided by the mitochondrial genome of *S. occidentalis* (NC\_005960.1) before being completed by MITObim (Hahn et al. 2013). For *S. grammicus*, the raw sequence reads were used as input for Novoplasty 2.6.3 (Dierckxsens et al. 2016) and finalized using MITObim. The complete and circularized mitogenome for *S. woodi* is 17,301 bp based on 26,225 reads while the *S. grammicus* mitogenome is 16,830 bp based on 5832 reads. Tablet indicated that the average coverages of *S. woodi* and *S. grammicus* mitogenomes were ~483× and ~203×. Notably, we found several high-coverage regions in our mitogenome assemblies, which may indicate the presence of

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**Figure 1.** Phylogenomics tree inferred using maximum likelihood based on the General Time Reversible model in MEGA7. Genes from complete mitochondrial genomes from all available species from the Iguanoidea superfamily were downloaded, aligned, and concatenated into a dataset composed of 11,505 nucleotide positions. Bootstrap values (1000 replicates) are shown at the corresponding nodes. Colored boxes indicate clades described, green asterisks represent new mitochondrial genomes described here using public data while blue asterisk represent the new mitochondrial genome described here using our own data. The accession numbers of the mitogenomes used in the tree are BK010486.1, BK010487.1, NC\_005960.1, NC\_026308.1, NC\_027261.1, NC\_036492.1, NC\_012839.1, NC\_027089.1, NC\_002793.1, NC\_028031.1, NC\_028030.1, NC\_012831.1, NC\_012829.1, NC\_012827.1, NC\_012836.1, NC\_012834.1, MK091854.1, NC\_010972.2, NC\_008776.1, and NC\_030781.1.

mitochondrial pseudogenes (NuMTs). These high-coverage regions are: 13,650-13,914 and 15,173-15,542 in *A. punctatus*; 15,419-15,568 in *S. grammicus*; and 5,114-7,020 and 11,611-13,216 in *S. woodi*. Automatic annotation of mitogenomes was performed using the MITOS Web Server (Bernt et al. 2013) and *geneCheker* (Schomaker-Bastos and Prosdocimi, 2018), followed by manual curation. The annotated mitogenomes were submitted to GenBank and TPA databases (*A. punctatus*: MK091854; *S. woodi*: BK010487; *S. grammicus*: BK010486). To confirm the taxonomic identities, we performed a phylogenetic analysis using all 14 complete mitogenomes available for the Iguanoidea superfamily and two outgroups. Phylomito software (<https://github.com/igorrcoستا/phylomito>) extracted the protein-coding genes, performed sequence alignments, and concatenations. A maximum likelihood analysis confirmed monophyly for *Anolis* and *Sceloporus* genera (Figure 1).

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We would like to thank Adam Leaché for providing helpful information about the origins of the *Sceloporus* sequences used in this study. We cherish the inspiring presence of Alex Schomaker (*in memoriam*) that worked in initial versions of the anole mitogenome: thank you, Alex.

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No potential conflict of interest was reported by the authors.

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