

MITOGENOME ANNOUNCEMENT

Complete mitochondrial genomes of the New World jacanas: *Jacana spinosa* and *Jacana jacana*Matthew J. Miller^{1,2}, Celestino Aguilar², Luis Fernando De León², José R. Loaiza², and W. Owen McMillan¹¹Smithsonian Tropical Research Institute, Panamá, República de Panamá and ²Centro de Biodiversidad y Descubrimiento de Drogas, Instituto de Investigaciones Científicas y Servicios de Alta Tecnología, Panamá, República de Panamá**Abstract**

The New World jacanas, *Jacana spinosa* (Mexico to Panama and also the West Indies) and *Jacana jacana* (Panama and South America), are polyandrous freshwater waders that are common throughout the Neotropics. These two species hybridize narrowly at their contact zone in Panama, and as part of a study of the hybrid zone dynamics, we present complete, annotated mitochondrial genomes for both species. The two species have very similar mitochondrial genomes, showing identical gene orders, and differing in size in only two RNA features and the control region, and among protein-coding genes, the two genomes had average uncorrected pairwise divergence of 1.8%, ranging from 0.7% for ND4L and 3.6% for ATP8. However, control region divergence is high (~16%). These mitochondrial genome sequences may be useful tools for understanding jacana hybridization dynamics, especially regarding potential mitonuclear incompatibilities.

KeywordsGenome, *Jacana*, mitochondrion, waders**History**

Received 30 March 2014

Revised 6 April 2014

Accepted 13 April 2014

Published online 20 May 2014

Genomic DNA was isolated from Smithsonian Tropical Research Institute Bird Collection (STRIBC) vouchered tissues: *Jacana jacana* (STRIBC 4055: Panama, Darién) and *Jacana spinosa* (STRIBC 3332: Panama, Chiriquí). Specimens were collected under Panamanian ANAM permit: SE/A-137-10 (Notif: DAPVS-0628-11) and IACUC permit 2011-0927-2014-03. Whole mitochondrial genomes were sequenced as off-target captures generated during next-generation sequencing of ultra-conserved elements libraries (UCEs, e.g. Faircloth et al., 2012; protocols at: <http://ultraconserved.org/>). Whole mtDNA genomes are routinely recovered from avian UCE sequencing efforts (Smith et al., 2014). Briefly, we assembled contigs from paired-end 150 base-pair MiSeq (Illumina, San Diego, CA) reads using Velvet 1.2 (Zerbino & Birney, 2008) via the phyluce package (<https://github.com/faircloth-lab/phyluce>) using a k-mer of 95. Each sample had just one contig of approximately 16,000 basepairs. A BLAST (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>) search confirmed that these contigs referred to mitochondrial DNA. Initial draft annotations were generated in DOGMA (Wyman et al., 2004; <http://dogma.ccbb.utexas.edu/>), and subsequently tuned by eye. The complete control region was not initially recovered during Velvet assembly, so we re-mapped all reads onto the draft genomes using Bowtie 2.2.1 (Langmead & Salzberg, 2012) implemented in Geneious 7.0.6 (Biomatters, <http://www.geneious.com>). Thus we recovered the consensus sequence of the control region and confirmed the validity of the complete mitochondrial sequence. A

total of 14,741 reads were mapped to *J. jacana* (average coverage = 126 reads) while 32,168 reads were mapped to *J. spinosa* (average coverage = 274). Genbank accessions are: KJ631049 (*J. jacana*) and KJ631048 (*J. spinosa*). We calculated uncorrected pairwise divergence between features of the two genomes via Muscle alignments executed in Geneious.

Both genomes are comprised of 2 rRNAs, 22 tRNAs, 13 protein-coding genes, and 1 displacement loop (control region) in the standard avian gene order (Desjardins & Morais, 1990). Between the two species, all genes and RNA sequences were of identical size except for single additional nucleotides in both 12S and tRNA^{Lys} in *Jacana jacana*. In both species, the ND3 gene had a base pair at position 174 that results in a stop codon in the open reading frame; this almost certainly represents another example of a non-translated nucleotide at this position in ND3, which occurs in over 50 bird and turtle species (Mindell et al., 1998). Among the 38 mtDNA features, pairwise sequence differences varied from 0.0% (14 tRNAs) to 15.9% (control region). Among protein-coding regions, ND4L had the shallowest pairwise divergence (0.7%) and ATP8 had the largest divergence (3.6%; Table 1 provides all pairwise divergences).

The two New World jacana species are conspicuous for their species-specific ornamental facial shields, and have an extremely narrow hybrid zone in western Panama (Miller et al., in review). Recently, Hill & Johnson (2013) hypothesized that avian ornaments such as jacana facial shields, are “honest signals” allowing birds to choose mates that maximize compatibility between mitochondrial genes and associated nuclear cellular respiration genes. We note that hybrid jacanas typically have aberrant facial ornaments, and hope that these mitochondrial genomes aid studies of mitonuclear incompatibilities in hybridization dynamics.

Table 1. Characteristics of the *J. jacana* mitochondrial genome.

Code	Amino Acid	Start	Stop	Size	Spacer (+) or Overlap (–)	Direction	% Pairwise Divergence	Start Codon	Stop Codon
F	tRNA-Phe	1	67	67	0	F	0.0%		
	12S rRNA	68	1036 (1035)	969 (968)	0	F	1.4%		
V	tRNA-Val	1037 (1036)	1109 (1108)	73	0	F	1.4%		
	16S rRNA	1110 (1109)	2697 (2696)	1588	0	F	1.1%		
L	tRNA-Leu	2698 (2697)	2771 (2770)	104	10	F	2.7%		
	ND1	2782 (2781)	3757 (3756)	976	0	F	1.9%	ATG	TAA
I	tRNA-Ile	3758 (3757)	3829 (3828)	72	11	F	1.4%		
Q	tRNA-Gln	3841 (3840)	3911 (3910)	71	–1	R	0.0%		
M	tRNA-Met	3911 (3910)	3979 (3978)	69	0	F	1.4%		
	ND2	3980 (3979)	5018 (5017)	1039	0	F	1.5%	ATG	T–
W	tRNA-Trp	5019 (5018)	5092 (5091)	74	1	F	0.0%		
A	tRNA-Ala	5094 (5093)	5162 (5161)	69	1	R	0.0%		
N	tRNA-Asn	5164 (5163)	5236 (5235)	73	2	R	0.0%		
C	tRNA-Cys	5239 (5238)	5305 (5304)	67	–1	R	0.0%		
Y	tRNA-Tyr	5305 (5304)	5375 (5374)	71	1	R	0.0%		
	COX I	5377 (5376)	6927 (6926)	1551	–9	F	1.4%	ATG	AGG
S	tRNA-Ser (UCN)	6919 (6918)	6992 (6991)	74	3	R	1.4%		
D	tRNA-Asp	6995 (6994)	7065 (7064)	71	1	F	0.0%		
	COX II	7067 (7066)	7750 (7749)	684	0 (1)	F	2.2%	ATG	TAA
L	tRNA-Lys	7751	7821 (7820)	71 (70)	1	F	1.4%		
	ATP 8	7823 (7822)	7990 (7989)	168	–10	F	3.6%	ATG	TAA
	ATP 6	7981 (7980)	8664 (8663)	684	–1	F	2.3%	ATG	TAA
	COX III	8664 (8663)	9447 (9446)	784	0	F	0.8%	ATG	T–
G	tRNA-Gly	9448 (9447)	9516 (9515)	69	0	F	0.0%		
	ND3	9517 (9516)	9868 (9867)	352	2	F	1.7%	ATA	TAA
R	tRNA-Arg	9871 (9870)	9940 (9939)	70	1	F	0.0%		
	ND4L	9942 (9941)	10,238 (10,237)	297	–7	F	0.7%	ATG	TAA
	ND4	10,232 (10,231)	11,606 (11,605)	1375	3	F	1.7%	ATG	T–
H	tRNA-His	11,610 (11,609)	11,678 (11,677)	69	1	F	1.4%		
S2	tRNA-Ser	11,680 (11,679)	11,743 (11,742)	64	0	F	0.0%		
L	tRNA-Leu 2	11,744 (11,743)	11,814 (11,813)	71	0	F	1.4%		
	ND5	11,815 (11,814)	13,629 (13,628)	1815	10	F	2.3%	ATG	AGA
	Cytb	13,640 (13,639)	14,782 (14,781)	1143	0	F	1.8%	ATG	TAA
T	tRNA-Thr	14,783 (14,782)	14,852 (14,851)	70	5	F	0.0%		
P	tRNA-Pro	14,858 (14,857)	14,927 (14,926)	70	9	R	0.0%		
	ND6	14,937 (14,936)	15,458 (15,457)	522	0	R	1.7%	ATG	TAG
E	tRNA-Glu	15,459 (15,458)	15,529 (15,528)	71	0	R	0.0%		
	Control Region	15,530 (15,529)	16,975 (17,079)	1446 (1551)	0	F	15.9%		

Parentheses indicate feature differences for *J. spinosa*; otherwise the features between the two species were identical.

Declaration of interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the article. This work was funded by the Smithsonian Institution and the Smithsonian Tropical Research Institute Office of Academic Programs. We thank Panama's Environmental Ministry (ANAM) for continued support of scientific collecting, without which, this study would not have been possible. The jacana specimens were collected by the STRIBC with the support of an NIH/NSF "Ecology and Evolution of Infectious Diseases" award from the Fogarty International Center 3R01-TW005869-05S1.

References

Desjardins P, Morais R. (1990). Sequence and gene organization of the chicken mitochondrial genome. *J Mol Biol* 212:599–634.
 Faircloth BC, McCormack JE, Crawford NG, Harvey MG, Brumfield RT, Glenn TC. (2012). Ultraconserved elements anchor thousands of genetic markers spanning multiple evolutionary timescales. *Syst Biol* 61:717–26.

Hill GE, Johnson JD. (2013). The mitonuclear compatibility hypothesis of sexual selection. *Proc Biol Sci* 280:20131314. doi: 10.1098/rspb.2013.1314.
 Langmead B, Salzberg S. (2012). Fast gapped-read alignment with Bowtie 2. *Nat Methods* 9:357–9.
 Miller MJ, Lipshutz SE, Smith NG, Bermingham E. Genetic and phenotypic characterization of a hybrid zone between polyandrous Northern and Wattled Jacanas in western Panama. *BMC Evol Biol*, in review.
 Mindell DP, Sorenson MD, Dimcheff DE. (1998). An extra nucleotide is not translated in mitochondrial ND3 of some birds and turtles. *Mol Biol Evol* 15:1568–71.
 Smith BT, Harvey MG, Faircloth BC, Glenn TC, Brumfield RT. (2014). Target capture and massively parallel sequencing of ultraconserved elements for comparative studies at shallow evolutionary time scales. *Syst Biol* 63:83–95.
 Wyman SK, Jansen RK, Boore JL. (2004). Automatic annotation of organellar genomes with DOGMA. *Bioinformatics* 20:3252–5.
 Zerbino DR, Birney E. (2008). Velvet: Algorithms for de novo short read assembly using de Bruijn graphs. *Genome Res* 18:821–9.