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MOLECULAR PHYLOGENY OF JACANAS AND ITS IMPLICATIONS FOR MORPHOLOGIC AND BIOGEOGRAPHIC EVOLUTION

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ABSTRACT.—We compared sequences of mitochondrial cytochrome-*b* and ND5 genes in a phylogenetic analysis of seven species of jacanas, representing all six genera and including the Greater Painted-snipe (*Rostratula benghalensis*) as an outgroup. When analyzed separately by parsimony and maximum-likelihood bootstrapping, the two genes produced consistent trees, although the ND5 tree was better resolved than the cytochrome-*b* tree. When combined, the data from the two genes produced a fully resolved tree that was identical to the ND5 tree. This tree had the following form: ((((*Irediparra, Microparra*), *Metopidius*), *Actophilornis*), ((*Jacana jacana, J. spinosa*), *Hydrophasianus*)), *Rostratula*. The phylogeny consists of two major clades that were known to traditional and phylogenetic taxonomists. It also contains sister taxa that are geographically disjunct: the New World *Jacana* and Asian *Hydrophasianus*, and the African *Microparra* and Australian *Irediparra*. We postulate that this biogeographic pattern results from the extinction of intervening African and Asian taxa, respectively. *Received 31 May 1998, accepted 26 April 1999.*

JACANAS are worldwide inhabitants of tropical and subtropical open wetlands. Eight extant species in six genera are recognized in the family Jacanidae. Four genera are monotypic and occur on three continents: *Microparra* (Africa), *Irediparra* (Australia), *Hydrophasianus* (Asia), and *Metopidius* (Asia). Two other genera consist of two species: *Actophilornis africanus* (Africa) and *A. albinucha* (Madagascar), and *Jacana jacana* (South America) and *J. spinosa* (Central America). The genera of jacanas are mainly allopatric, but some co-occur in portions of Asia (*Hydrophasianus* and *Metopidius*) and Africa (*Actophilornis* and *Microparra*). The two *Jacana* species co-occur and hybridize only in a small area of western Panama (Wetmore 1965).

Until recently, relationships of the jacanas to other groups of birds were poorly known. It is now clear that they are members of the Charadriiformes and probably are most closely related to painted-snipes, family Rostratulidae (Kitto and Wilson 1966, Strauch 1978, Sibley and Ahlquist 1990). Within the Jacanidae, there have been no thorough modern studies of intergeneric relationships. The only systematic studies have been tangents of larger efforts to reconstruct shorebird phylogeny (Strauch 1978, Sibley and Ahlquist 1990, Chu 1995). Strauch's (1978) character-compatibility (clique) analysis of 63 skeletal and seven muscular characters of shorebirds defined two groups of jacana genera: (1) *Hydrophasianus* and *Jacana*, and (2) *Metopidius, Actophilornis, Irediparra,* and *Microparra.* Following a critical appraisal of Strauch's shorebird study (Michevich and Parenti 1980), Chu (1995) reanalyzed Strauch's data using cladistic parsimony. His reanalysis indicated the monophyly of jacanas but was not designed to resolve relationships within the family. Sibley and Ahlquist's (1990) DNA-DNA hybridization study indicated that *Actophilornis* was much closer to *Irediparra* than to *Jacana.* Unfortunately, Sibley and Ahlquist did not include other jacana taxa in their comparisons.

To shed more light on intergeneric relationships in jacanas, we compared DNA sequences of portions of the mitochondrial cytochrome-*b* gene and the fifth subunit of nicotinamide adenine dinucleotide dehydrogenase gene (ND5) of all six genera. These comparisons resolve the jacana phylogeny and provide an initial assessment of the utility of ND5 sequences for studying avian phylogeny. Knowledge of the phylogeny of jacanas permits us to speculate on the evolutionary forces that shaped their morphology and distribution.

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METHODS

One individual from each of seven species representing all six genera of the Jacanidae were sampled (Table 1): African Jacana (*Actophilornis africanus*), Lesser Jacana (*Microparra capensis*), Comb-crested Jacana (*Irediparra gallinacea*), Pheasant-tailed Jacana (*Hydrophasianus chirurgus*), Bronze-winged Jacana (*Metopidius indicus*), Wattled Jacana (*Jacana jacana*), and Northern Jacana (*J. spinosa*). The Greater Painted-snipe (*Rostratula benghalensis*) was used as an outgroup because of its apparent close relationship to the jacanas (Sibley and Ahlquist 1990).

Laboratory methods.—In an attempt to avoid amplification of nuclear pseudogenes, we extracted DNA from tissue, which has a relatively high mitochondrial DNA content compared with blood (Sorenson and Fleischer 1996, Quinn 1997). DNA was extracted from 0.1 g of heart, liver, or muscle tissue following the standard phenol/chloroform extraction method (Hillis et al. 1990). A portion of the mitochondrial cytochrome-*b* and ND5 genes was isolated and amplified via the polymerase chain reaction (PCR). We used the following primers for cytochrome *b* (H16065: GGAGTCTTCAGTCTCTGGTTTACAAGAC and L15656: AACCTACTAGGAGACCCAGA; Helm-Bychowski and Cracraft 1993) and ND5 (H14149: CCTATTTTGCGGATGTCTTGTTC and L 13753: CAGGAAAATCCGCTCAATTCGG; Garciá-Moreno et al. 1999). Primer sequences are listed 5′ to 3′ and numbers refer to the 3′ base of the primer referenced to the complete mtDNA sequence of the chicken (*Gallus gallus*; Desjardins and Morais 1990). H and L refer to primers located on the heavy and light strands of the mitochondrial genome, respectively.

PCR reactions were done in 50-μL volumes using 0.5 μM of each primer, 2.5 mM $MgCl_2$, 10 mM of each dNTP, and 3 U of *Taq* polymerase. Thermal cycling began with 3.0 min at 94°C for initial denaturation, followed by 35 cycles of denaturation (94°C, 1 min), annealing (46 to 50°C, 1.5 min) and extension (72°C, 1 min), and a final extension of 10 min at 72°C. PCR products were run on a 2% low-melt agarose gel that was stained with ethidium bromide, allowing the amplified DNA fragment to be visualized under UV illumination and excised from the gel. PCR products were purified and concentrated with Wizard PCR Preps Kit (Promega A7170) and then sequenced directly on an ABI 373 Stretch automated sequencer. Sequences are deposited in GenBank under accession numbers AF146616 to AF146631.

Data analysis.—Sequences were aligned by eye relative to the chicken sequence (Desjardins and Morais 1990) using the editor and translator in MEGA (Kumar et al. 1993). Base frequencies, variation, pairwise transition and transversion values, and distances were determined with MEGA. Overall transition to transversion ratio (ti:tv) was estimated by plotting the pairwise proportion of transitions versus the proportion of transversions (Moore and DeFillipis 1997); ti:tv was also estimated via the maximum-likelihood option in PAUP* (Swofford 1998). Phylogenetic relationships were estimated using maximum-likelihood (randomized heuristic search) and maximum-parsimony (branch and bound search) methods with PAUP*. Bootstrapping values (100 replicates) were determined for both maximum-likelihood and parsimony methods to assess support for internal branches. Cytochrome-*b* and ND5 data were combined and analyzed separately to observe the behavior of the genes.

RESULTS

Characteristics of sequence data.—We examined a total of 705 nucleotides (345 of cytochrome *b* and 360 of ND5) for seven species of jacanas and the outgroup (*Rostratula*). Average nucleotide compositions for both genes and all positions were as follows: G = 11.3%, A = 30.2%, T = 24.3% and C = 34.2%. Nucleotide compositional bias was highest at third-codon positions, intermediate at second positions, and lowest at first positions (Table 2). First positions were biased slightly, being G-poor and C-rich for cytochrome *b* and slightly T-poor and A-rich for ND5 (Table 2). Second positions were G-poor and T-rich for both cytochrome *b* and ND5. Third positions of both genes were most heavily biased toward G- and T-poor and A- and C-rich. The total number of variable and potentially phylogenetically informative sites was similar for both cytochrome-*b* and ND5 fragments (Table 3). Although the number of variable amino acids appeared lower for ND5 than for cytochrome *b*, the number of potentially informative amino acids was similar (Table 3). The numbers of variable and informative sites were highest at third- codon positions, intermediate at first positions, and lowest at second positions (Table 2). These trends were evident whether or not the outgroup was included with the jacanas.

The percent divergence (using the Tamura and Nei [1993] correction) among taxa for cytochrome *b* and ND5 is summarized in Table 4 and Figure 1. For cytochrome-*b* sequences, ingroup divergence ranged from 2% (*Jacana jacana* to *J. spinosa*) to 17.2% (*Irediparra* to *Jacana jacana*). Similarly, ND5 ingroup divergence ranged from 1.7% (*Jacana jacana* to *J. spinosa*) to 17.4% (*Microparra* to *Actophilornis* and *Microparra* to *Hydrophasianus*). For the combined

TABLE 1. Distributions and collection localities of jacana species sampled plus the outgroup.

Species[a]	Distribution	Sample number[b]	Collection locality
African Jacana (*Actophilornis africanus*)	Central and southern Africa	LSUMNS B19187	Dallas Zoo
Lesser Jacana (*Microparra capensis*)	Central and southeastern Africa	LSUMNS B23806	South Africa
Comb-crested Jacana *Irediparra gallinacea*)	Malaysia, northeastern Australia	LSUMNS B14045	Australia
Pheasant-tailed Jacana (*Hydrophasianus chirurgus*)	India, Southeast Asia	LSUMNS B14041	India
Bronze-winged Jacana (*Metopidius indicus*)	India, Southeast Asia	LSUMNS B14040	India
Northern Jacana (*Jacana spinosa*)	Mexico, Central America	LSUMNS B28956	Mexico
Wattled Jacana (*Jacana jacana*)	Panama, northeastern South America	LSUMNS B15237	Bolivia
Greater Painted-snipe (*Rostratula benghalensis*)	Africa, India, Southeast Asia	71 (C. Sibley)	Eastern Malaysia

[a] Classification follows Sibley and Monroe (1990).
[b] LSUMNS = Louisiana State University Museum of Natural Science.

data, ingroup sequence divergence ranged from 2% (*Jacana jacana* to *J. spinosa*) to 16.5% (*Microparra* to *Actophilornis* and *Microparra* to *Jacana jacana*). It was not surprising that the congeneric New World species (*Jacana*) exhibited the lowest divergence. However, we did not expect to find the greatest sequence divergence between the two African species (*Microparra* and *Actophilornis*). The divergence pattern in Figure 1 suggests that cytochrome *b* evolves slightly faster, overall, than ND5 (17 out of 28 points lie below the diagonal).

To examine patterns of nucleotide change, we plotted pairwise proportions of transitions and transversions against overall distance (Tamura and Nei 1993). For cytochrome *b*, both first- and second-position transitions increased slightly with overall distance (Fig. 2A), whereas first- and second-position transversions did not (Fig. 2B). Third-position transitions (Fig. 2A) and transversions (Fig. 2B) increased more substantially with overall distance than any other partition. Of ND5 partitions, third-position transitions (Fig. 3A) and transversions (Fig. 3B) increased most dramatically in relation to overall distance. ND5 first- and second-position divergence did not change markedly in relation to overall distance (Figs. 3A, B).

Because the third-position transitions and transversions increased steadily in comparison with overall genetic divergence, these partitions appear generally unsaturated. However, other studies of cytochrome *b* indicate that third-position transitions, if not transversions, often are saturated above the 10% divergence level (Moore and DeFillipis 1997, Sheldon et al. 1999). It is difficult to tell the extent of saturation in Figure 2 because data on the x-axis and y-axis are not independent, and Tamura-Nei distances (x-axis) can be foreshortened by saturation, despite correction for back mutations (Sheldon et al. 1999). Similarly, the ND5 data (Figs. 3A, B) do not appear saturated, but again it is difficult to tell in these plots. Moreover, we cannot rely on the findings of other studies using ND5 because there has been no comparative work on ND5 in birds. To examine potential saturation of the data using an alternative method, we plotted the proportion of transitions versus transversions in Figure 4, as suggested by Moore and DeFillipis (1997). That plot indicates that the ti:tv ratio decreases with increasing transversion, suggesting that saturation slows the apparent rate of transitional change at higher levels of divergence, at least to some degree.

Determining initial ti:tv was difficult because only one pairwise comparison (*Jacana jacana* with *J. spinosa*) was outside the likely zone of saturation (on the left-hand side of Fig. 4). Thus, there were not enough taxa for a regression analysis. Moreover, the short sequence lengths of the two genes made it difficult to determine an accurate ratio even for the single *Jacana-Jacana* pair. There were no ND5 transversions between them, and only three (synony-

TABLE 2. Percent nucleotide composition, number of variable sites, and number of informative sites by codon position in cytochrome-*b* and ND5 genes of the Jacanidae (excluding *Rostratula*).

	First position				Second position				Third position			
	G	A	T	C	G	A	T	C	G	A	T	C
Cytochrome *b*												
% Nucelotides	19.1	26.7	19.5	34.7	10.6	17.3	43.5	28.7	1.6	47.1	10.3	40.9
No. variable sites		21				6				66		
No. informative sites		10				2				39		
ND5												
% Nucelotides	23.0	32.7	18.6	25.7	12.5	16.9	39.2	31.4	1.0	40.3	14.9	43.9
No. variable sites		21				2				67		
No. informative sites		12				1				44		
Total												
% Nucelotides	21.1	29.8	19.0	30.1	11.6	17.1	41.3	30.1	1.3	43.6	12.7	42.4
No. variable sites		42				8				133		
No. informative sites		22				3				83		

TABLE 3. Summary of variable sites and amino acids in cytochrome *b* and ND5 for the seven species of jacanas. Numbers in parentheses include the outgroup (*Rostratula benghalensis*).

	Cytochrome *b*	ND5	Total
Sites			
Total	345	360	705
No. variable	93 (102)	90 (101)	183 (203)
No. informative	51 (59)	57 (67)	108 (126)
Amino acids			
Total	115	120	235
No. variable	19 (23)	13 (14)	32 (37)
No. informative	7 (7)	5 (5)	12 (12)

mous) cytochrome-*b* transversions. By including the other fairly closely related pairs of taxa (e.g. *Jacana* with *Hydrophasianus*), it was possible to estimate approximate initial ti:tv of 6:1 for ND5 and 3:1 for cytochrome *b*. The combined initial ti:tv appears to be about 3:1 (Fig. 4). Maximum-likelihood searches by PAUP* indicated a combined ti:tv of 3.48.

One concern in this study was that because the DNA sequences were short, they might represent accidentally amplified nuclear pseudogenes. However, the characteristics of the sequences indicated that they were not pseudogenes. The cytochrome-*b* sequences exhibited nucleotide composition, codon-position rates of change, and transition and transversion patterns that are typical of the bird mitochondrial cytochrome *b* (e.g. Edwards et al. 1991, Nunn and Cracraft 1996, Slikas 1997). It is more difficult to tell whether the ND5 sequences are typical because no published ND5 data sets exist for birds. However, the ND5 sequences exhibited patterns of base composition and change that were very similar to the cytochrome-*b* sequences and showed no signs of pseudogene behavior (Sorenson and Fleischer 1996, Quinn 1997).

PHYLOGENETIC ANALYSIS

Total sequence data.—In the combined analysis of cytochrome-*b* and ND5 nucleotide sequences, maximum-parsimony (MP) and maximum-likelihood (ML) methods produced a single topology (Fig. 5). The ML tree had a ln of −2,437.1475, an estimated ti:tv at 3.48, and a site- specific rate factor for the gamma distribution (alpha) of 0.17. Weighted MP analysis

TABLE 4. Percent divergence (corrected for back mutations [Timura and Nei 1993]) between taxa for cytochrome-*b* (above diagonal) and ND5 (below diagonal) sequences.

Species	*Jj*	*Jsp*	*Hc*	*Mi*	*Aa*	*Mc*	*Ig*	*Rb*
Jacana jacana	—	2.06	11.58	15.40	14.19	16.64	17.20	18.02
Jacana spinosa	1.70	—	11.21	15.05	14.54	16.64	16.88	18.21
Hydrophasianus chirurgus	9.75	10.43	—	10.85	10.75	12.93	14.42	15.01
Metopidius indicus	14.47	15.97	13.08	—	15.28	12.94	14.85	16.17
Actophilornis africanus	13.05	12.96	11.29	12.16	—	15.75	15.01	17.41
Microparra capensis	16.48	14.95	17.36	14.68	17.35	—	11.44	18.48
Irediparra gallinacea	15.09	14.29	14.84	13.60	14.33	11.02	—	18.98
Rostratula benghalensis	14.51	14.45	15.30	17.05	14.13	22.29	17.27	—

(ti:tv = 3) produced a single most-parsimonious tree (length = 556 steps, CI excluding uninformative characters = 0.734). Bootstrap values (100 replicates) ranged from 72 to 100% for ML and 66 to 100% for MP (Fig. 5). In the ML/MP tree, two major clades were evident. The first consisted of four taxa: ((*Irediparra, Microparra*), *Metopidius*), *Actophilornis*. The second clade consisted of the two New World species, *Jacana jacana* and *J. spinosa*, plus *Hydrophasianus* as their sister group.

Cytochrome-b *data.*—The analysis of cytochrome-*b* sequences alone (Fig. 6) resulted in a single MP tree (length = 281 steps, CI excluding uninformative characters = 0.758, weighted transition:transversion = 3), which was identical to the ML tree (ln = −1,203.9366, PAUP* estimated ti:tv at 3.46, and the site-specific rate factor for the gamma distribution at 0.19). When bootstrapped, the cytochrome-*b* tree indicated a very strong association between *Jacana jacana* and *J. spinosa*, and between *Irediparra* and *Microparra*. In this respect, it was similar to the total sequence tree. However the

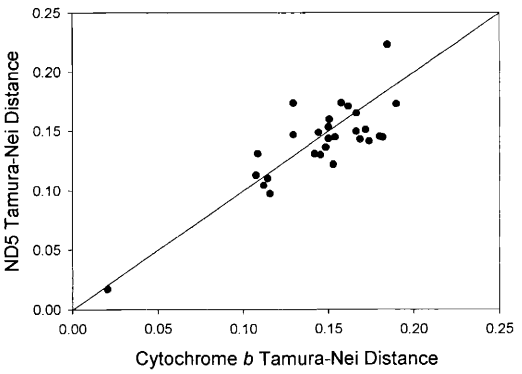

FIG. 1. Divergence of cytochrome *b* versus ND5 for jacanas and the *Rostratula* outgroup.

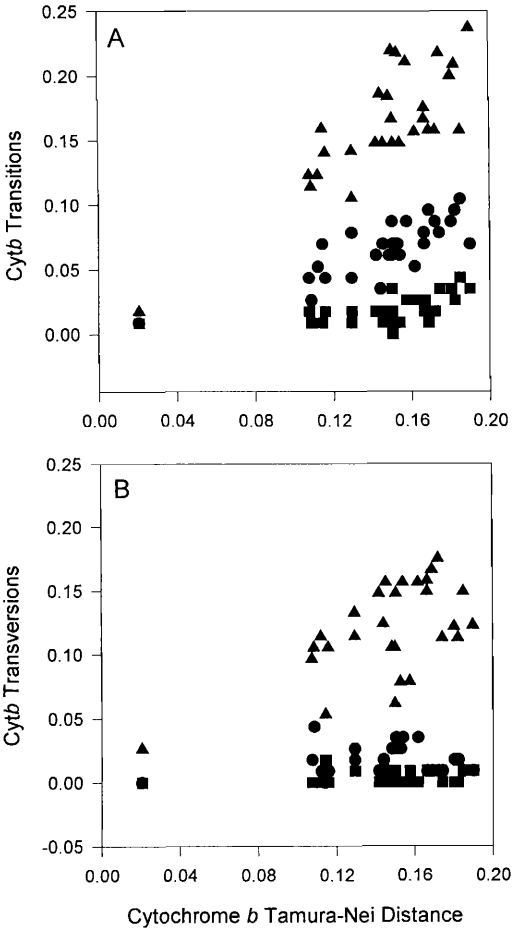

FIG. 2. Divergence of cytochrome-*b* transitions (A) and transversions (B) relative to corrected distance (Tamura and Nei 1993) at first- (circles), second- (squares), and third-codon (triangles) positions.

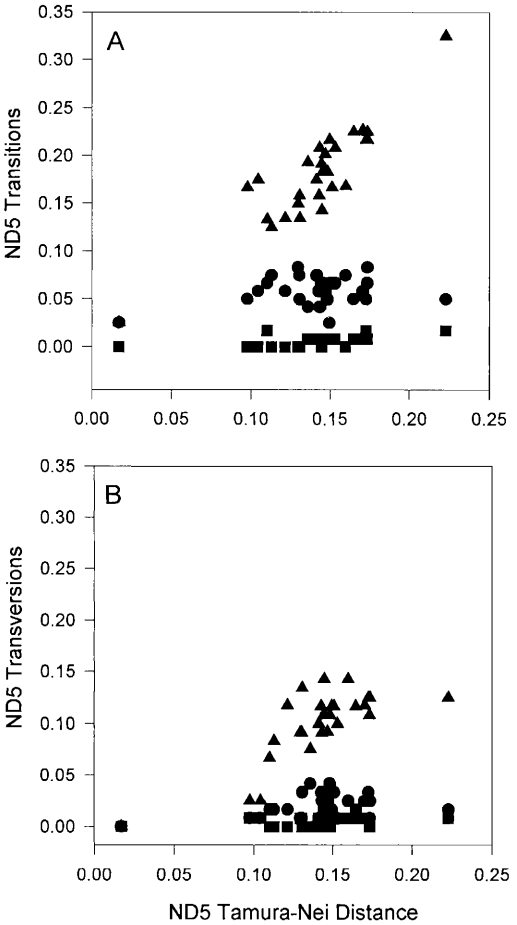

FIG. 3. Divergence of ND5 transitions (A) and transversions (B) relative to corrected distance (Tamura and Nei 1993) at first- (circles), second- (squares) and third-codon (triangles) positions.

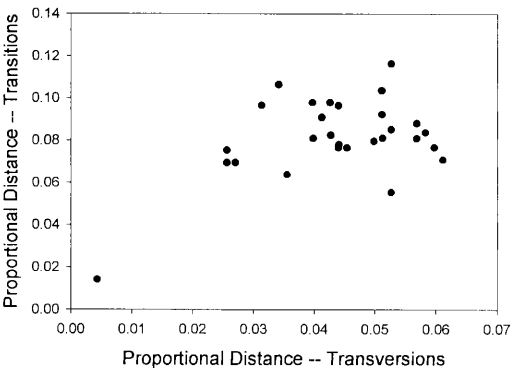

FIG. 4. Plot of transitions versus transversions for combined cytochrome-*b* and ND5 sequences.

positions of *Actophilornis* vis-a-vis *Metopidius* was unclear, as was the position of *Hydrophasianus* in regard to the *Jacana* species. Compared with the total sequence tree and the ND5 tree (see below), the cytochrome-*b* tree was poorly resolved.

ND5 data.—Analysis of ND5 sequences using both MP and ML produced a single tree (Fig. 5) identical to the total sequence tree (MP: length = 296 steps, CI excluding uninformative characters = 0.725, weighted ti:tv = 3; ln = −1,227.2669, PAUP* estimated ti:tv at 3.78, and the site-specific rate factor for the gamma distribution [alpha] at 0.15). The cytochrome-*b*, ND5, and combined trees were congruent, given the low bootstrap values for positions of *Actophilornis*, *Metopidius*, and *Hydrophasianus* in the cytochrome-*b* tree.

DISCUSSION

Our analysis of cytochrome-*b* and ND5 sequence data produced a fully resolved phylogeny indicating the division of the Jacanidae into two clades: (1) Australian *Irediparra*, African *Microparra*, Asian *Metopidius*, African *Actophilornis*, and (2) Asian *Hydrophasianus* and New World *Jacana*. The phylogeny of these taxa is as follows: (((*Irediparra*, *Microparra*), *Metopidius*), *Actophilornis*), ((*Jacana jacana*, *J. spinosa*), *Hydrophasianus*).

In general, previous classifications of jacanas have been consistent with this phylogeny. Wagler (1832 *in* Johnsgard 1981) recognized seven species of jacanas and placed them in three genera: *Jacana*, *Hydrophasianus*, and *Metopidius*. *Jacana* and *Hydrophasianus* were monotypic genera and thought to be each others' closest relatives. *Metopidius* included the five species of Old World "typical" jacanas: *capensis*, *africanus*, *albinucha*, *gallinacea*, and *indicus*. In classifications subsequent to Wagler's, the five "typical" Old World jacanas were split into four separate, and mostly monotypic, genera (e.g. Peters 1934): *Microparra*, *Actophilornis*, *Irediparra gallinacea*, and *Metopidius indicus*.

Previous phylogenetic studies also produced results largely consistent with ours. Strauch's (1976, 1978) character-compatibility analysis of Charadriiformes revealed two groups of jacanas corresponding to those that we found. One included *Hydrophasianus* and *Jacana*, and the

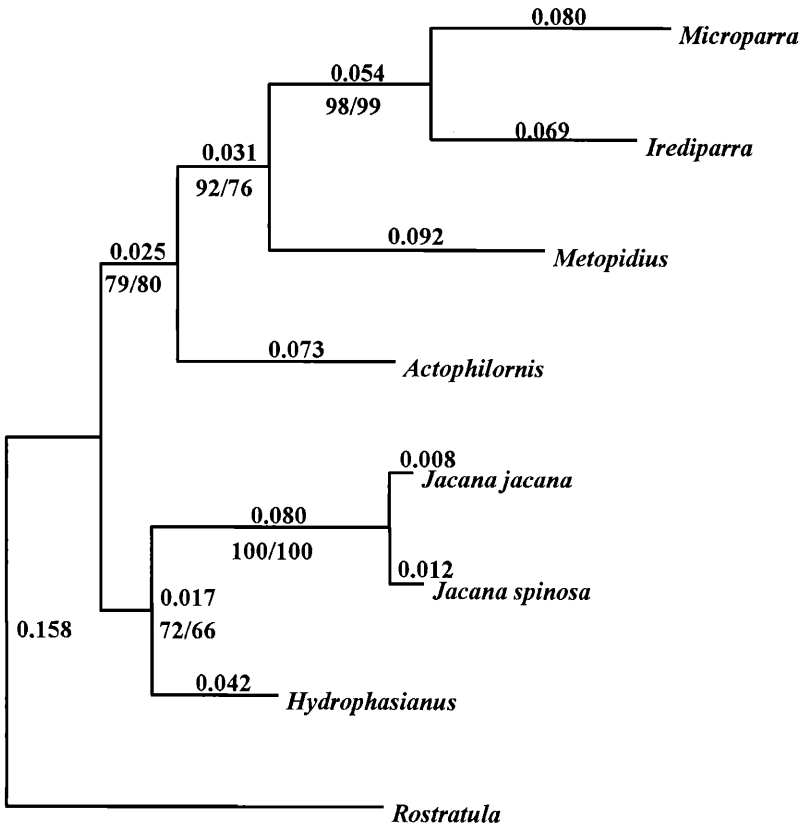

FIG. 5. Phylogenetic relationships among the Jacanidae based on maximum-likelihood (ML) and maximum-parsimony (MP) analyses of 345 nucleotides of cytochrome *b* and 360 nucleotides of ND5. Identical branching patterns were determined by both analyses (MP single most- parsimonious tree length = 556 steps, CI excluding uninformative characters = 0.734; ML tree ln = −2,437.1475). See text for more details. Percentage bootstrap support from both methods of phylogenetic analysis is shown to the left of internal branches (ML / MP).

other included *Microparra, Irediparra, Metopidius,* and *Actophilornis.* The *Hydrophasianus-Jacana* group was defined by shared-derived wing spurs and the other group (the "typical" Old World species) by shared-derived wing knobs and a blade-like radius. These two groups also are distinguished by plumage characters (Strauch 1976). Both *Hydrophasianus* and *Jacana* have light-colored wing stripes in all plumages (except for neonatal plumage), and a black eye stripe is present in young birds. None of the other genera of jacanas has either of these character states in any plumage. However, within the "typical" Old World group, the Australian and two African genera (*Irediparra, Microparra,* and *Actophilornis*) share a derived character (fusion of the ischium and pubis) not found in *Metopidius* (Strauch 1976). This led Strauch (1976) to place *Metopidius* basal to *Irediparra, Microparra,* and *Actophilornis.* In contrast, our molecular analysis places *Actophilornis* basal to *Irediparra, Microparra,* and *Metopidius.* Based on his results, Strauch (1976) recommended recognizing only two genera of jacanas: *Actophilornis* (including *Actophilornis, Irediparra, Microparra,* and *Metopidius*), and *Jacana* (including *Jacana* and *Hydrophasianus*).

Strauch's (1976, 1978) method—character compatibility or clique analysis—was criticized by Mickevich and Parenti (1980), who preferred a parsimony approach. However, a recent reanalysis of Strauch's data using modern cladistic parsimony failed to resolve any relationships within the Jacanidae (Chu 1995). In con-

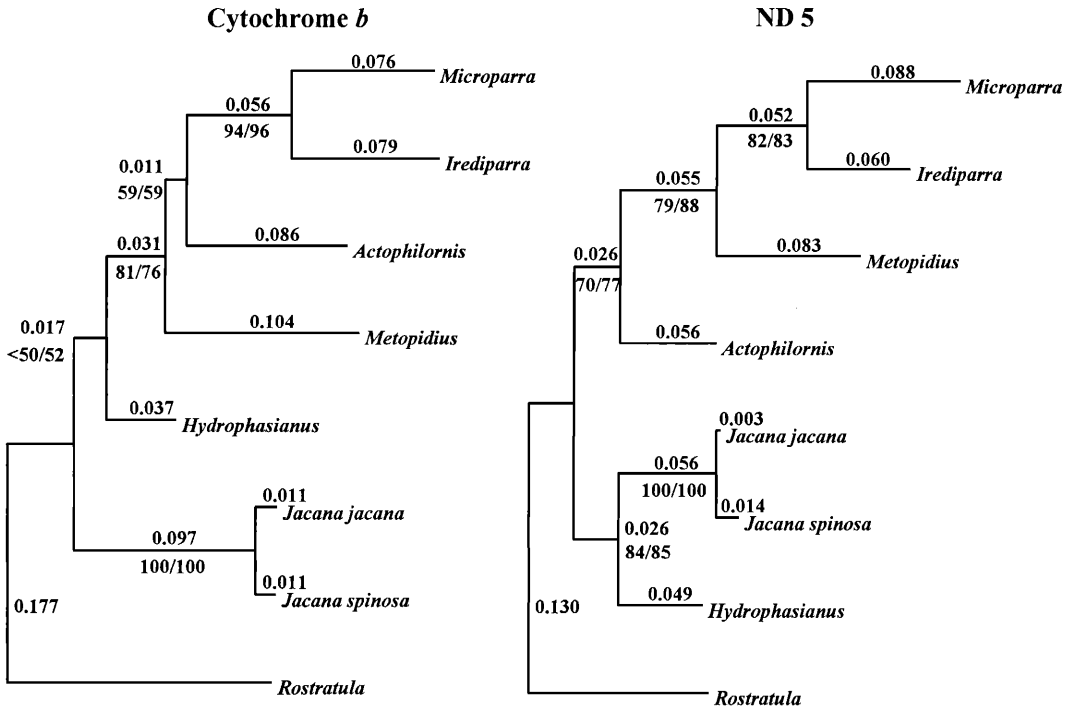

FIG. 6. Phylogenetic relationships among the Jacanidae based independently on the analysis of cytochrome *b* (345 nucleotides ; MP single most-parsimonious tree length = 281 steps, CI excluding uniformative characters = 0.758; ML tree ln = −1,203.9366) and ND5 (360 nucleotides; MP single most-parsimonious tree length = 296 steps, CI excluding uniformative characters = 0.725; ML tree ln = −1,227.2669) mitochondrial genes. For details, see text. Percentage bootstrap support from both methods of phylogenetic analysis is shown to the left of internal branches (ML / MP).

trast, the tree produced by clique analysis substantially resembles our molecular tree. So, at least for the Jacanidae, if not for the Charadriiformes, clique analysis appears to be an effective approach.

Sibley and Ahlquist (1990) compared three jacana genera by DNA-DNA hybridization (*Irediparra, Jacana,* and *Actophilornis*) using *Irediparra* as the radio-labeled control and *Rostratula* as the radio-labeled outgroup. The pairwise distances in their tree were ΔT50H 4.9 for *Irediparra-Actophilornis* (three comparisons) and ΔT50H 8.0 for *Irediparra-Jacana* (two comparisons). Their findings are consistent with ours and Strauch's (1976, 1978), and their data also suggest that a substantial divergence gap separates the two main groups of jacanas.

Fry's (1983) study appears at first to contradict ours. Using an evolutionary-taxonomy approach, he examined various characteristics of the jacanas and concluded that there were three genera: *Jacana* (for *Jacana, Actophilornis, Metopidius,* and *Irediparra*), *Hydrophasianus,* and *Microparra.* The main reason he lumped the first four genera into *Jacana* was that they are "less heterogeneous" than other groups of shorebirds that have been lumped (e.g. lapwings). He kept *Hydrophasianus chirurgus* and *Microparra capensis* in their own genera because of the distinctiveness of these two species. Such an approach does not meet modern (cladistic) standards. It groups taxa by overall similarity and ignores potential information in shared-derived characters (e.g. wing spurs for *Jacana* and *Hydrophasianus*). Nevertheless, Fry's summary of jacana characters contains several interesting observations about their evolution. He observed that only *Actophilornis, Metopidius,* and *Irediparra* share a unique blade-like radius, which presumably is an adaptation for wing-brooding of eggs and downy young, whereas *Jacana, Hydrophasianus,* and *Microparra* have a

typical avian radius. He also noted that the plumage of adult *Microparra* is similar to that of juveniles in the other species. The only other adult jacana to have such plumage is the nonbreeding *Hydrophasianus*. From these observations, and others (e.g. patterns of radius development in juveniles), he concluded that *Microparra* is neotenic, having retained the juvenile radius and plumage. This conclusion makes excellent sense and also provides an explanation for homoplasy in jacana morphology. For example, the "typical" avian radius described by Fry (1983) is a synapomorphy uniting *Jacana* and *Hydrophasianus* and a convergence due to neoteny in *Microparra*. Similarly, the retention of juvenile character states also explains how *Microparra* obtained its plumage and the possible causes of the unique nonbreeding plumage of *Hydrophasianus* and the appearance and disappearance of frontal shields in various species.

Biogeography.—The jacanas have a pantropical distribution. Like other groups of birds with similar distributions, such as storks, tiger-herons, finfoots, trogons, and barbets (Houde 1994, Lanyon and Hall 1994, Houde et al. 1995, Sheldon et al. 1995, Slikas 1997), jacanas are too young to be Gondwanan (Sibley and Ahlquist 1990) but are old enough to have developed idiosyncratic biogeographic patterns. Thus, there is no simple geographic scenario (e.g. common vicariance pattern or dispersal route) to explain how jacanas and other pantropical bird groups became distributed in the manner we observe today. However, given new phylogenetic information, a few recurrent patterns among some pantropical groups are beginning to emerge. These patterns are enough to suggest some large-scale processes behind current distributions. For example, two of the more unusual phylogenetic relationships among the jacana genera are the sister groupings of (1) the New World *Jacana* and the Asian *Hydrophasianus* and (2) the African *Microparra* and Australian *Irediparra*. These distributions are reminiscent of the distributions of other groups, such as storks. We know that the Jabiru (*Jabiru mycteria*) of the New World is most closely related to the Saddlebill (*Ephippiorhynchus senegalensis*) of Africa and the Black-necked Stork (*E. asiaticus*) of Asia and Australia (Slikas 1997). This pattern is repeated, in some respects, by other clades of storks (e.g. *Mycteria* in the New World, Africa, and Asia). The New World-Asian (*Jacana-Hydrophasianus*) distribution is similar to the stork distribution without an African representative. Furthermore, the African-Australian (*Microparra-Irediparra*) distribution is similar to the stork distribution without an Asian or New World representative. Quite likely, the jacana distributions are relicts of wider distributions in which the intervening taxa became extinct. Such a pattern applies to several nonpasserine groups. For example, the tiger-herons, which are represented by three species in the New World (*Tigrisoma*), one in Africa (*Tigriornis*), and one in New Guinea (*Zonerodius*), would be an example of a group that may have undergone extinction in Asia.

This pattern of extinction of intervening species is most obvious in depauperate groups, such as jacanas, but it occurs in speciose groups as well. We have found, for example, closely related sister species of swallows in Africa (*Pseudhirundo griseopyga*) and Australia (*Cheramoeca leucosternus*) without intervening Asian relatives (Sheldon and Winkler 1993). Moreover, this pattern seems to apply mainly to relatively vagile open-country or marsh-dwelling taxa, such as jacanas, storks, herons, and swallows. These groups have redundant distributions, indicating multiple invasions of different continents. The forest-dwelling groups, such as trogons and barbets, with monophyletic New World representatives (Sibley and Ahlquist 1990, Lanyon and Hall 1994, Espinosa de los Monteros 1998), have largely nonredundant distributions. These distributions suggest these groups have moved unidirectionally around the globe. The difference in dispersal patterns among groups suggests that we must factor behavior (e.g. dispersal potential), ecology (e.g. habitat preferences), and geography (e.g. routes of dispersal), at the very least, into a general discussion of patterns and causes of pantropical bird distribution.

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Literature Cited

Chu, P. C. 1995. Phylogenetic reanalysis of Strauch's osteological data set for the Charadriiformes. Condor 97:174–196.

Desjardins, P., and R. Morais. 1990. Sequence and gene organization of the chicken mitochondrial genome. A novel order in higher vertebrates. Journal of Molecular Biology 212:599–634.

Edwards, S. V., P. Arctander, and A. C. Wilson. 1991. Mitochondrial resolution of a deep branch in the genealogical tree for perching birds. Proceedings of the Royal Society of London Series B 243:99–107.

Espinosa de los Monteros, A. 1998. Phylogenetic relationships among the trogon. Auk 115:937–954.

Fry, C. H. 1983. The jacanid radius and *Microparra*, a neotenic genus. Gerfaut 73:173–184.

Garciá-Moreno, J., P. Arctander, and J. Fjeldså. 1999. Strong diversification at the treeline among *Metallura* hummingbirds. Auk 116:702–711.

Helm-Bychowski, K., and J. Cracraft. 1993. Recovering phylogenetic signal from DNA sequences: Relationships within the corvine assemblage (class Aves) as inferred from complete sequences of the mitochondrial DNA cytochrome *b* gene. Molecular Biology and Evolution 10:1196–1214.

Hillis, D. M., A. Larson, S. K. Davis, and E. A. Zimmer. 1990. Nucleic acids III: Sequencing. Pages 318–370 *in* Molecular systematics (D. M. Hillis and C. Moritz, Eds.). Sinauer Associates, Sunderland, Massachusetts.

Houde, P. 1994. Evolution of the Heliornithidae: Reciprocal illumination by morphology, biogeography and DNA hybridization (Aves: Gruiformes). Cladistics 10:1–19.

Houde, P., F. H. Sheldon, and M. Kreitman. 1995. A comparison of solution and membrane-based DNA × DNA hybridization, as used to infer phylogeny. Journal of Molecular Evolution 40: 678–688.

Johnsgard, P. A. 1981. Plovers, sandpipers and snipes of the world. University of Nebraska Press, Lincoln, Nebraska.

Kitto, G. B., and A. C. Wilson. 1966. Evolution of malate dehydrogenase in birds. Science 153: 1408–1410.

Kumar, S., K. Tamura, and M. Nei. 1993. MEGA: Molecular Evolutionary Genetics Analysis, version 1.01. Institute of Molecular Evolutionary Genetics, Pennsylvania State University, University Park.

Lanyon, S. M., and J. G. Hall. 1994. Reexamination of barbet monophyly using mitochondrial-DNA sequence data. Auk 111:389–397.

Mickevich, M. F., and L. R. Parenti. 1980. Review of "The phylogeny of the Charadriiformes (Aves): A new estimate using the method of character compatability analysis." Systematic Zoology 29:108–113.

Moore, W. S., and V. R. DeFillipis. 1997. The window of taxonomic resolution for phylogenies based on mitochondrial cytochrome *b*. Pages 83–119 *in* Avian molecular evolution and systematics (D. P. Mindell, Ed.). Academic Press, New York.

Nunn, G. B., and J. Cracraft. 1996. Phylogenetic relationships among the major lineages of the birds-of-paradise (Paradisaeidae) using mitochondrial gene sequences. Molecular Phylogenetics and Evolution 5:445–459.

Peters, J. L. 1934. Check-list of the birds of the world, vol.2, Harvard University Press, Cambridge, Massachusetts.

Quinn, T. W. 1997. Molecular evolution of the mitochondrial genomes. Pages 3–28 *in* Avian molecular evolution and systematics (D. P. Mindell, Ed.). Academic Press, New York.

Sheldon, F. H., K. G. McCracken, and K. D. Stuebing. 1995. The phylogeny of the Zigzag Heron (*Zebrilus undulatus*) and White-crested Bittern (*Tigriornis leucolophus*) estimated by DNA-DNA hybridization. Auk 112:672–679.

Sheldon, F. H., L. A. Whittingham, and D. W. Winkler. 1999. Comparison of cytochrome *b* and DNA-hybridization data bearing on the phylogeny of swallows (Aves: Hirundinidae). Molecular Phylogenetics and Evolution 11:320–331.

Sheldon, F. H., and D. W. Winkler. 1993. Intergeneric phylogenetic relationships of swallows estimated by DNA-DNA hybridization. Auk 110: 798–824.

Sibley, C. G., and J. E. Ahlquist. 1990. Phylogeny and classification of birds. Yale University Press, New Haven, Connecticut.

Sibley, C. G., and B. L. Monroe, Jr. 1990. Distribution and taxonomy of birds of the world. Yale University Press, New Haven, Connecticut.

Slikas, B. 1997. Phylogeny of the avian family Ciconiidae (storks) based on cytochrome *b* sequences and DNA-DNA hybridization distances. Molecular Phylogenetics and Evolution 8:275–300.

Sorenson, M. D., and R. C. Fleischer. 1996. Multiple independent transpositions of mitochondrial DNA control region sequences to the nucleus. Proceedings of the National Academy of Sciences USA 93:15239–15243.

Strauch, J. G., Jr. 1976. The cladistic relationships of

the Charadriiformes. Ph.D. dissertation, University of Michigan, Ann Arbor.

STRAUCH, J. G., Jr. 1978. The phylogeny of the Charadriiformes: A new estimate using the method of character compatibility analysis. Transactions of the Zoological Society of London 34:263–345.

SWOFFORD, D. L. 1998. PAUP*: Phylogenetic Analysis Using Parsimony, version 4. Sinauer Associates, Sunderland, Massachusetts.

TAMURA, K., AND M. NEI. 1993. Estimation of the number of nucleotide substitutions in the control region of mitochondrial DNA in humans and chimpanzees. Molecular Biology and Evolution 10:512–526.

WETMORE, A. 1965. The birds of the republic of Panama. Part 1. Smithsonian Miscellaneous Collections 150:1–483.

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