Molecular systematics of swifts of the genus *Chaetura* (Aves: Apodiformes: Apodidae)

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Abstract

Phylogenetic relationships among swifts of the morphologically conservative genus *Chaetura* were studied using mitochondrial and nuclear DNA sequences. Taxon sampling included all species and 21 of 30 taxa (species and subspecies) within *Chaetura*. Our results indicate that *Chaetura* is monophyletic and support the division of the genus into the two subgenera previously identified using plumage characters. However, our genetic data, when considered in combination with phenotypic data, appear to be at odds with the current classification of some species of *Chaetura*. We recommend that *C. viridipennis*, currently generally treated as specifically distinct from *C. chapmani*, be returned to its former status as *C. chapmani viridipennis*, and that *C. andrei*, now generally regarded as synonymous with *C. vauxi aphanes*, again be recognized as a valid species. Widespread Neotropical species *C. spinicaudus* is paraphyletic with respect to more range-restricted species *C. fumosa*, *C. egregia*, and *C. martinica*. Geographically structured genetic variation within some other species of *Chaetura*, especially notable in *C. cinereiventris*, suggests that future study may lead to recognition of additional species in this genus. Biogeographic analysis indicated that *Chaetura* originated in South America and identified several dispersal events to Middle and North America following the formation of the Isthmus of Panama.

Keywords

Chaetura; swifts; Apodidae; phylogenetics; species limits; cryptic species

Introduction

The systematics of many Neotropical organisms remain woefully understudied. Their evolutionary relationships are typically poorly understood, and species diversity is often severely underestimated, even in such relatively well-studied groups as birds (e.g., Isler et al. 1998). Numbers of species are most commonly underestimated in groups that are conservative in characters traditionally used in lower-level systematics; in birds, these characters have generally been plumage and morphometrics. Morphology of swifts (Aves: Apodidae), which are among the most aerial of birds, is notoriously conservative, and the widespread New-World genus *Chaetura* is one of the most uniform of swift genera. As Wetmore (1957) noted, "the genus is one in which close superficial similarities are the rule, so that to separate the species it is necessary to scan closely for details that would be disregarded in a group of greater diversity." The taxonomic problems resulting from morphological conservatism within this genus have been exacerbated by a lack of museum specimens, a lack of understanding of geographic variation in plumage, and the high mobility of many species (Marín 1997), in addition to a dearth of behavioral data on most taxa.

Chaetura was once considered to encompass a wide range of species, including many now placed in the Old World genera *Hirundapus*, *Mearnsia*, *Neafrapus*, *Rhaphidura*, *Telecanthura*, and *Zoonavena* (Peters 1940, Lack 1956). Although previous authors (e.g., Lack 1957, Meise 1964) considered the American species to form a monophyletic group within *Chaetura*, it was only later that Brooke (1970), using plumage, size, breeding behavior, and geographical distribution, restricted the genus to its current configuration. *Chaetura* is now considered to consist of a group of 9-11 species endemic to the New World (e.g., Chantler 1999, Dickinson and Remsen 2013; del Hoyo and Collar 2014; Gill and Donsker 2015; Table 1).

Not surprisingly, given the lack of morphological variation, species limits in *Chaetura* have been and continue to be unsettled. Marín (1997, 2000), in studies based on morphological characters, recommended that *C. chapmani viridipennis*, *C. andrei meridionalis*, *C. spinicaudus fumosa*, and *C. cinereiventris egregia*, which except for *egregia* (e. g., Wolters 1976, Parker and Remsen 1987) were almost universally treated as subspecies, be elevated to species status, and that *C. andrei andrei* be merged into *C. vauxi*. Most subsequent general references have followed these recommendations (e.g., Dickinson and Remsen 2013, del Hoyo and Collar 2014, Gill and Donsker 2015, Remsen et al. 2015). However, a more recent recommendation, that *C. brachyura ocypetes* be elevated to species status based on morphological and vocal differences from other forms of *brachyura* (Ridgely and Greenfield 2001, but see Schulenberg et al. 2007), has not been generally accepted. Moreover, two taxa treated as

species throughout much of the 20th century, *C. vauxi richmondi* and *C. vauxi gaumeri* (following Ridgway 1911, Cory 1918, and Peters 1940), are now generally treated as subspecies (following Griscom 1932, Sutton 1941).

The case of *C. andrei* is a particularly interesting and illustrative example of taxonomic confusion within *Chaetura*. Nominate *andrei*, described by Berlepsch and Hartert (1902), is definitively known from only five specimens collected in eastern Venezuela in the 1890s (Marín 1997), although Cherrie (1916) noted that it was "not uncommon" where he collected it in Caicara. Specimens of *Chaetura a. meridionalis* were classified under the species *C. pelagica* until Hellmayr (1907) described *meridionalis* as a southern population of *andrei.* Marín (1997) argued that the degrees of plumage and size differences between *andrei* and *meridionalis* were similar to those between other congeners recognized as species (e.g., *vauxi* and *pelagica*) and, therefore, recommended elevating *meridionalis* to species status pending further analyses. Furthermore, he concluded that nominate *andrei* was inseparable in size and color from worn individuals of *C. vauxi aphanes*, which occurs in the northern cordillera of Venezuela, and he recommended that these be merged and that *andrei* (the name has priority over *aphanes*) henceforth be considered a subspecies of *vauxi*.

Although morphological variation in the genus is slight, *Chaetura* has sometimes been divided into two subgenera based on differences in contrast in plumage between back and rump (Brooke 1970): *Acanthylis*, which includes currently recognized species *spinicaudus*, *fumosa*, *martinica*, *egregia*, and *cinereiventris*; and *Chaetura*, which includes currently recognized species *pelagica*, *vauxi*, *chapmani*, *viridipennis*, *brachyura*, and *meridionalis*, as well as *andrei*. Marín (2000) recognized these subgenera as the gray-rumped (=*Acanthylis*) and the brown-rumped (=*Chaetura*) groups, and further divided the gray-rumped group into gray-rumped and pale-rumped subgroups (Table 1). The gray-rumped subgroup consisted of the single species *cinereiventris,* and the pale-rumped subgroup contained four allopatrically or parapatrically distributed species (*martinica*, *spinicaudus*, *fumosa*, and *egregia*), which he referred to as the "*martinica* species complex."

In this study we used molecular data to: (1) assess the monophyly of *Chaetura* as currently circumscribed (Brooke 1970); (2) determine whether subgenera *Acanthylis* and *Chaetura* (Brooke 1970), and the pale- and gray-rumped subgroups (Marín 2000) of *Acanthylis*, are monophyletic; (3) conduct a preliminary assessment of the monophyly of each species of *Chaetura*; and (4) evaluate the genetic status of proposed (and in several cases generally accepted) species *fumosa*, *egregia*, *richmondi, viridipennis, meridionalis,* and *ocypetes* relative to their putative or former conspecifics *spinicaudus*, *cinereiventris*, *vauxi, chapmani, andrei,* and *brachyura*, respectively.

Methods

Our sampling was guided by the taxonomy of Dickinson and Remsen (2013), except that for sampling purposes (1) *C. andrei* was considered a species rather than a subspecies of *C. vauxi*, and (2) we recognized two additional subspecies of *C. spinicaudus*: *aethalea* and *latirostris*. We sampled two individuals of every species and subspecies for which tissue samples were available, except for *C. c. chapmani*, *C. cinereiventris sclateri*, *C. egregia*, and *C. meridionalis*, for which three individuals were sampled, and *C. brachyura cinereocauda*, *C. brachyura ocypetes*, *C. cinereiventris guianensis*, and *C. viridipennis*, for which only single individuals were available (Table 2). Fresh tissue samples of *C. martinica*, *C. andrei*, and key subspecies *C. vauxi aphanes* and *C. c. cinereiventris* were unavailable; *C. martinica*, *C. andrei*, and *C. c. cinereiventris* were sampled from toepads of museum study skins, and *C. vauxi aphanes* was sampled from dried tissue from skeletons collected by CTC. Our sampling included all species and 21 of the 30 taxa within *Chaetura*; in all, 42 individuals of *Chaetura* were sampled. Difficult identifications were checked by re-examination, and in some cases re-identification, of voucher specimens. To maximize the potential to document genetic variation within taxa, individuals of the same species or subspecies were chosen from localities as distant from each other as possible, although in many cases the distance was necessarily minimal (Table 2). We also sampled three species of swifts not currently considered to be part of *Chaetura*. Two of these species (*Neafrapus cassini* and *Hirundapus caudacutus*) were once placed in *Chaetura* and are now considered part of the Chaeturini; thus, they provide a simple test of monophyly of the genus (tissues of *Mearnsia*, *Rhaphidura*, *Telecanthura*, and *Zoonavena* were not available). The third species was the more distantly related *Apus apus*, which belongs to a different tribe of Apodidae (Apodini) and which was designated the outgroup in all phylogenetic analyses.

DNA was extracted from tissue samples using Qiagen DNeasy blood and tissue DNA extraction kits. For toepads and skeletal samples, DNA was extracted in a physically isolated ancient DNA laboratory following strict protocols to minimize and detect contamination. All surfaces and equipment were regularly treated with a 50% solution of bleach and/or UV irradiation, and sterile, disposable blades were used for cutting tissue and skeletal samples. Extraction blanks and negative controls were employed to detect potential contamination. DNA extractions were conducted via a phenol/chloroform procedure with subsequent centrifugal dialysis (Fleischer et al. 2000). DNA extractions and PCR setup were conducted in the ancient DNA laboratory prior to moving to the separate contemporary DNA lab.

We sequenced three DNA fragments for each fresh tissue sample: the mitochondrial gene nicotinamide adenine dinucleotide dehydrogenase subunit 2 (ND2), intron 15 of the Z-linked aconitase gene (ACO15), and intron 3 of the Z-linked muscle-specific kinase gene (MUSK). PCRs were performed on a Biorad DNA Engine Tetrad 2 thermocycler in 25 uL reactions. Thermocycling conditions were as follows: 95°C for 4 min; 40 cycles of 94°C for 45 sec, 52°C for 45 sec, 72°C for 90 sec; and 72°C for 10 min. The annealing temperature was increased to 54-60°C for amplification of ACO15 for several individuals.

For fresh tissue samples, ND2 was amplified in two pieces, using paired primers L5216 and H5766 (both Sorenson et al. 1999) for the first piece and primers L5758 (Sorenson et al. 1999) and H6313 (Johnson and Sorenson 1998) for the second piece. Primer pair L5758/H6313 did not amplify for sample UAM 17562, so we paired L5758 with H6113 (Zwiers et al. 2008) instead, resulting in a shorter sequence for this individual. Primers used for ACO15 were ACO Ai15fbb and ACO Ai15ra (Fernandes et al. 2013), and primers for MUSK were MUSK-I3F and MUSK-I3R (Kimball et al. 2009). For samples from museum specimens, ND2 was amplified in smaller pieces using a wide variety of primers, most designed specifically for this study (Table 3). Primer pairs typically used for more recent specimens were L5216/H5538sw, L5390sw/H5766, L5758sw/H6113sw, and L6076sw/H6313; older specimens required amplification of smaller pieces of DNA. Internal primers were also designed for the nuclear introns, but we were unable to obtain nuclear sequences for samples taken from museum specimens. PCR products were cleaned for cycle sequencing using ExoSAP-IT (Affymetrix). Samples were sequenced in both directions using an ABI PRISM 3130 automated sequencer and assembled, edited, and aligned using Sequencher 4.9 (GeneCodes Corp., Ann Arbor, Michigan, USA). All sequences have been submitted to GenBank (accession numbers xxx-xxx).

Numbers of variable and parsimony-informative characters were calculated using PAUP*4.0 (Swofford 2003), and single-gene, nuclear-only, and concatenated phylogenetic trees were estimated using maximum likelihood (ML) as implemented in RAxML 8.0.0 (Stamatakis 2014; http://embnet.vital-it.ch/raxml-bb/) and IQ-tree 1.3.8 (Nguyen et al. 2014), and Bayesian approaches as implemented in MrBayes 3.1.2 (Ronquist and Huelsenbeck 2003). For the ML analyses, separate phylogenetic trees were inferred for mitochondrial sequences, nuclear sequences, and for the combined dataset. Combined ML and Bayesian analyses were partitioned by gene. RAxML analyses were performed with the GTR + Γ model of sequence evolution and included 100 bootstrap replicates in addition to the search for the most likely tree. For IQ-tree analyses, we used a partitioning scheme and best-fit models of DNA sequence evolution identified by partitionFinder 2.0 (Lanfear et al., 2016) treating each codon

position and each nuclear intron as data subsets. In our search we considered JC, HKY, and GTR models, each with and without estimated base frequencies, the proportion of invariant sites, and Γ distributed rate heterogeneity. We selected models with AICc, and used the greedy partitioning search scheme. PartitionFinder identified four partitions for downstream phylogenetic inference: each ND2 codon position (1st: HKY+I+Γ+X, 2nd: HKY+I+X, 3rd: GTR+I+X) and the combined MUSK5 and ACO15 introns (GTR+G+X). For each gene and the nuclear-only and concatenated alignments, we implemented 100 iteration ML tree searches and 1000 ultrafast bootstrap approximations.

Concatenated Bayesian analyses were run for 10,000,000 generations, using the previously inferred model of sequence evolution. Tree searching was conducted using four independent Metropolis coupled Markov chains, with adjustment of chain heating conditions (temp 5 0.1–0.05) for improved chain swap acceptance rates, and sampling every 100 generations; trees from the first 25% of generations were discarded as burn-in. Evaluation of stationarity and chain convergence was conducted by plotting posterior probabilities from the run in the program Tracer v1.6 (Rambaut et al. 2014). Trees were uploaded into Geneious 6.1.1 (Kearse et al. 2012) to determine a consensus tree rooted at *Apus apus*.

We also implemented a time-calibrated coalescent Bayesian analysis in *BEAST 2.2 (Bouckaert et al., 2014), treating each named taxon (species or subspecies) as a tip. We used the models selected by partitionFinder, except that we substituted the simpler HKY+Γ+X models for HKY+I+Γ+X and GTR+G+X to minimize convergence issues and parameter interactions associated with overparameterization. Preliminary runs implementing a relaxed lognormal clock resulted in 95% confidence intervals of the clock rate standard deviation including zero, justifying use of a strict clock (Ho and Duchêne, 2014). We set the clock rate for ND2 at 2.1% per million years (pairwise; Weir and Schluter, 2008), selected a Yule tree prior, and a “linear with constant root” population size. We ran two independent MCMC chains of 100 million, sampling every 10,000 generations. We discarded the first 25% of samples as burnin, and ensured that parameter estimates had converged and reached effective sample sizes over 200 in Tracer 1.5 (Rambaut and Drummond, 2007). We summarized the remaining 3000 posterior species trees and gene trees as a maximum clade credibility trees in LogCombiner (Bouckaert et al., 2014).

We conducted a second species tree analysis using ASTRAL-III 5.5.6 (Zhang et al. 2017) to check whether incomplete lineage sorting or missing data for some genes introduced biases into our *BEAST coalescent or concatenated analyses. We used the ML gene trees inferred from RAxML as input trees.

To infer the biogeographic origin and history of *Chaetura*, we implemented the DEC, DIVALIKE, and BAYAREALIKE models, each with and without the +J jump-dispersal parameter, in BioGeoBEARS

(Matzke, 2014). Biogeographic reconstructions used the *BEAST maximum clade credibility species tree. We defined the following regions as areas: 1 - Old World (outgroups), 2 - Nearctic, 3 - Middle America north of the Panama Canal Zone, 4 - South America including eastern Panama and Trinidad and Tobago, and 5 - Caribbean. Two species (*C. cinereiventris* and *C. brachyura*) have limited distributions in the Caribbean but were coded as mainland only because we lacked tissue of the relevant subspecies. We used AICc to select the best-fit model for interpretation. We also estimated the history of seasonal migratory behavior evolution with 1-rate Maximum likelihood using the ACE function in the R package phytools (Revell 2012), again using the maximum clade credibility species tree. Data on seasonal migratory behavior (Table 1) were taken from Collins (1968), Sick (1993), Chesser (1994), and Chantler (2000).

Consideration of species limits was based primarily on the Biological Species Concept (BSC), the species definition most commonly used by ornithological references (e.g., Dickinson and Remsen 2013). However, we also used the bGMYC (Reid and Carstens, 2012) model to provide a purely molecular perspective on species limits in *Chaetura*, through identification of lineages more divergent than expected under panmixia. We used 100 trees randomly subsampled from the ND2 posterior distributions and a 0.5 threshold to identify potential species; we selected this threshold as a best estimate and a compromise between identifying false species and not identifying true species.

Our genetic results prompted us to investigate phenotypic differences between *C. andrei* and *C. vauxi aphanes*, which were previously thought to belong to separate species but had been merged based on a perceived lack of phenotypic difference (Marín 1997). Study skins of these two taxa located at the American Museum of Natural History, where most or all skins of *C. andrei* are housed, were examined as part of our study. Plumage of the available skins of *C. andrei*, including the type, was directly compared to that of available specimens of *C. vauxi aphanes*, and rectrices of the two taxa were measured using digital calipers. Diameter of the central rectrices was measured at the base, where they emerge from the skin.

Results

Complete ND2 sequences (1041 bp) were obtained for 41 of 45 individuals, and partial sequences (659-869 bp) for single samples of *cinereiventris cinereiventris* and *vauxi vauxi* and both samples of *andrei*. Complete or near-complete nuclear sequences (721 aligned bp for ACO15, 612 for MUSK) were obtained for the 38 individuals for which tissue samples were available, except for ACO15 for one sample of *egregia*. The number of variable characters among the ingroup was 298, 260 of which

were parsimony-informative. Distribution of parsimony-informative characters among genes was as follows: 214 in ND2, 24 in ACO15, and 22 in MUSK. First, second, and third codon positions of ND2 differed substantially in number of parsimony-informative characters, as expected for protein-coding genes: 43 (20.0%) at first codon positions, 17 (7.9%) at second positions, and 154 (72.0%) at third positions.

All concatenated trees based on likelihood and Bayesian analyses were virtually identical (Fig. 1), and all indicated that *Chaetura* is monophyletic. Support for this result was strong but not overwhelming (90% RAxML bootstrap, 97% IQ-tree bootstrap, 0.96 MrBayes posterior probability), likely because of weaker support in the mitochondrial data for this deep relationship. Of the outgroups, *Neafrapus cassini* was most closely related to *Chaetura.* The subgenera *Acanthylis* and *Chaetura* were strongly supported as monophyletic in all combined trees (100% bootstraps, 1.0 pp). Relationships within subgenus *Chaetura* were strongly supported and well-resolved: the two northernmost species, *pelagica* and *vauxi*, formed a clade sister to a clade including *chapmani*, *viridipennis*, *andrei*, *meridionalis*, and *brachyura*. Resolution within subgenus *Acanthylis* was poor: the four pale-rumped species (*spinicaudus, fumosa*, *martinica,* and *egregia*; Marín 2000) formed a subclade, rather weakly supported (48% and 59% bootstraps, 0.90 pp), and were extremely closely related, whereas the gray-rumped subgroup (*cinereiventris*; see below) could not be shown to be monophyletic.

Trees based solely on mitochondrial and nuclear data differed only slightly from the combined trees. The mitochondrial IQ-tree was identical to the combined data tree, but in the RAxML phylogeny, *Neafrapus* was a weakly supported sister to subgenus *Chaetura* (61% bootstrap). In the nuclear trees, *chapmani* and *viridipennis* were sister to *pelagica* and *vauxi* rather than to *andrei*, *meridionalis*, and *brachyura*. This result was well supported (98% and 73% bootstraps) and was the only instance of well-supported conflict between the nuclear and mitochondrial data.

The phylogenies based on coalescent *BEAST (Fig. 2) and ASTRAL-III (not shown) were similar to those produced using concatenated data. The *BEAST maximum clade credibility tree from the analyses (Fig. 2) was identical to the most likely tree (Fig. 1) except that the polytomy of *martinica*, *egregia+spinicaudus aethalea*, and *fumosa+spinicaudus spinicaudus* was resolved such that *martinica* and *egregia+spinicaudus aethalea* were sister taxa, albeit with poor posterior probability support. In the ASTRAL-III tree, the relationships of *chapmani-viridipennis* were unresolved within subgenus *Chaetura* (grouping with neither *vauxi-pelagica* nor *brachyura-andrei-meridionalis*), as might be expected for an area in which nuclear and mitochondrial data conflict. Curiously, the deepest division within subgenus *Acanthylis*, which was between *martinica* and all other taxa in the ASTRAL analysis but between most of

cinereiventris and all other taxa in the concatenated analyses. The latter ASTRAL result is puzzling, given that no analyses of strictly mitochondrial data (only mtDNA was available for *martinica*) produced a remotely similar result, and given the lack of divergence between *martinica, egregia*, and *spinicaudus*, and the large divergence both within *cinereventris* and between *cinereiventris* and other taxa in subgenus *Acanthylis* (see below). However, this result, like all phylogenetic relationships among the pale-rumped subgroup, lacked strong statistical support.

Most species of *Chaetura* were monophyletic, but *cinereiventris*, *spinicaudus*, and *chapmani* were not demonstrably monophyletic (Fig. 1). Subspecies *cinereiventris phaeopygos* was a very weakly supported sister (43% and 62% bootstraps, 0.84 pp) to the pale-rumped subgroup, thus essentially forming a trichotomy with the pale-rumped subgroup and *cinereiventris sclateri/guianensis/cinereiventris*. The pale-rumped subgroup formed what was essentially a four-fold polytomy, consisting of (1) *martinica*, (2) *spinicaudus aetherodroma*, (3) sister taxa *fumosa* and *spinicaudus spinicaudus*, and (4) rather weakly supported sister taxa *egregia* and *spinicaudus aethalea*. The single individual of *viridipennis* was sister to one *chapmani* individual, making *chapmani* paraphyletic with respect to *viridipennis*, but all individuals of *chapmani* and *viridipennis* were very closely related and support for relationships among individuals was poor.

Mean mitochondrial divergence between sister species was low, ranging from 0.1% (between *chapmani* and *viridipennis*) to 2.9% (between *pelagica* and *vauxi*); almost all sister species were less than 2% divergent. Maximum divergence between species was 11.5% (between *vauxi* and *cinereiventris*). Mean nuclear divergence between sister species was also very low, ranging from 0.1% (between *chapmani* and *viridipennis*) to 1.1% (between *pelagica* and *vauxi*). Maximum nuclear divergence between species was 2.2% (between *vauxi* and *cinereiventris*).

Recently or formerly accepted species *fumosa*, *egregia*, and *vauxi richmondi* were each monophyletic and distinct from their former or current conspecifics, *egregia* and *richmondi* differing by ca. 0.5% in mtDNA (Table 4). Currently recognized species *fumosa* also differed from its sister taxon *spinicaudus spinicaudus* by 0.5%, but differed from *spinicaudus aethalea* and *spinicaudus aetherodroma* by 1.0% and 1.2%, respectively. Species *spinicaudus* as currently recognized (i.e., without *fumosa*) was paraphyletic with respect to *fumosa*. The single individual of *brachyura ocypetes* was sister to all other individuals of *brachyura* and differed from them by 0.6% in mtDNA. Currently lumped species *andrei* and its proposed consubspecific *vauxi aphanes* were not closely related and differed by 7.0% in mtDNA; *andrei* was more closely related to its former conspecific *meridionalis* (mean mitochondrial divergence of 3.2%), but these were not sister taxa, instead forming a clade with *brachyura*, which was sister to

meridionalis. The largest intraspecific mitochondrial divergence, by far, was that between *cinereiventris phaeopygos* and its conspecifics *cinereiventris cinereiventris*, *cinereiventris sclateri*, and *cinereiventris guianensis*, which was 6.2%. Divergence between these taxa in the nuclear introns was also relatively high (0.7%).

All BioGeoBEARS analyses (Fig. 2 shows the best-fit model, DEC+J) indicated that *Chaetura* and both subgenera originated in South America. More recent dispersal was proposed to North America through independent events in *pelagica* and *v. vauxi*, to the Caribbean (*martinica*), and to Middle America through independent events in *vauxi richmondi* and the Middle American representatives of *Acanthylis* (*cinereiventris phaeopygos*, *spinicaudus aetherodroma*, and *fumosa*). Two other subspecies (*cinereiventris* and *brachyura*) include presumed peripheral isolates in the southern Lesser Antilles, and so probably represent additional dispersals to the Caribbean, but we lacked tissue of these subspecies. The time-caibrated *BEAST tree indicated that *Chaetura* separated from *Neafrapus cassini* roughly 13 mya and that subgenera *Acanthylis* and *Chaetura* separated ca. 10.5 mya. Diversification within *Acanthylis* and *Chaetura* occurred relatively recently, within the past 3.5 my for *Acanthylis* and within the past 5 my for *Chaetura*. Most speciation within the genus *Chaetura* ocurred in the Pleistocene.

Migration occurs only in subgenus *Chaetura*; analyses indicated that long-distance migration likely originated independently in South American austral migrant *meridionalis* and in both north temperate migrants. Although these two migrants, *vauxi* and *pelagica*, are sister species, the position in our trees of *v. vauxi*, the migrant subspecies, indicated that its migratory behavior and colonization of the north temperate zone probably evolved independently of *pelagica*. However, the evolution of migration in the ancestor to *vauxi-pelagica*, followed by a loss of migration in *vauxi* subspecies other than *v. vauxi*, is an alternate reconstruction. Intratropical migrant *viridipennis* also appears to have developed seasonal migratory behavior independently.

The bGYMC analyses (Supplemental Figure 1) produced a very conservative taxonomy under the 0.5 threshold, lumping some broadly sympatric taxa as single species. For example, the entire pale-rumped subgroup (i.e., *martinica*, *egregia*, *spinicaudus*, and *fumosa*) and *cinereiventris phaeopygos* were considered a single species under the 0.5 threshhold. Only six species were recognized under this criterion, in contrast to the 9-11 species recognized in current references. Only at the very liberal 0.95 threshold did the bGMYC analysis identify a number of species similar to that in current taxonomy, although the identity of species recognized differed considerably.

Four study skins of *C. andrei* (AMNH 477325, the type; AMNH 477326-477327, the two skins sampled for our genetic study; and AMNH 477328) were directly compared to seven study skins of *C.*

vauxi aphanes (AMNH 150208-150209, AMNH 150211, AMNH 648819, and AMNH 786081-786083). A fifth study skin of *andrei* (AMNH 177146) could not be located. Plumages of the two taxa are quite distinct: the lower breast and belly of *andrei* are noticeably darker than those of *vauxi aphanes*, and the undertail coverts of *andrei* are paler than or concolorous with the belly, whereas the undertail coverts of *vauxi aphanes* are darker than the belly. Moreover, the light area of the throat tends to be smaller and better delineated in *andrei* than in *vauxi aphanes*, and the upperparts of *andrei* are lighter brown (olive brown) than those of *vauxi aphanes*, which are blackish-brown. Even worn-plumaged *vauxi aphanes* were readily distinguishable from *andrei*.

As previously published, specimens of *andrei* have shorter tails and tail spines than those of *vauxi aphanes* (spines extending 2-3 mm beyond the vane in *andrei* versus 5-8 mm in *vauxi aphanes*), but length varies seasonally due to wear, and all skins of *andrei* have worn tails from the same time of year (February-March), making the validity of this difference difficult to assess (Marín 1997). However, diameter of rectrices, particularly at the base where they emerge from the skin, is presumably not subject to wear, and significant non-overlapping differences were found in the diameter of the central rectrices: mean values per individual ranged from 0.655 – 0.760 mm in *andrei* (mean 0.722, n = 4) versus 0.800 – 0.855 mm (mean 0.829, n = 7) in *vauxi aphanes* ($p = 0.005$).

Discussion

***Relationships within* Chaetura.**—Our results indicate that *Chaetura* is monophyletic, consistent with the restricted definition of the genus (Brooke 1970). The deep structure of our phylogenetic tree is consistent with previous views based on morphological variation in *Chaetura*: thus, monophyly of subgenera *Acanthylis* and *Chaetura* is supported by all trees, although reciprocal monophyly of Marín's (2000) pale-rumped and gray-rumped subgroups was inconclusive.

Recent classifications of species of *Chaetura* (e.g., Brooke 1970, Marín 2000) have consistently arranged species in accordance with the subgeneric arrangement, but species were previously grouped somewhat differently. In particular, *brachyura* and *andrei* (and *meridionalis*) were not grouped with va*uxi*, *pelagica*, and *chapmani*. The tree in Meise (1964), for example, indicated that *brachyura* and *andrei* form a group sister to the rest of the species, and that the other groups consisted of *spinicaudus*, *cinereiventris*, and *martinica* on the one hand and va*uxi*, *pelagica*, and *chapmani* on the other. Peters (1940) placed *brachyura* and *andrei* (along with *Telecanthura melanopygia* of western Africa) at the end of the linear sequence of his large genus *Chaetura*, far removed from the other New World species.

Although va*uxi*, *pelagica*, and *chapmani* (including *viridipennis*) have often been considered closely related (American Ornithologists' Union [1983] and Marín [1997] suggested that they form a superspecies), and although va*uxi*, *pelagica*, and *chapmani* are sister species in our nuclear analyses, *chapmani* is sister to the *andrei/meridionalis/brachyura* clade in our mitochondrial and combined trees (cf. Biancalana et al. 2017). This circumstance, in which the mitochondrial data contradict both nuclear and traditional phenotypic data, may be the result of stochasticity in lineage sorting affecting the mtDNA. Similarly strong and contradictory results have been obtained for buntings of the genus *Passerina* (Carling and Brumfield 2008), among others.

In all analyses, *cinereiventris, egregia,* and *martinica* did not form a monophyletic group, and therefore do not appear to form a superspecies, as Chantler (1999) had suggested. Rather, *egregia* and *martinica*, along with *fumosa*, were more closely related to *spinicaudus* than to *cinereiventris*. Marín (2000) reached a similar conclusion on the basis of differences between *cinereiventris* and the other taxa in body coloration and loral plumage, and therefore placed *cinereiventris* alone in his gray-rumped subgroup.

The placements of *andrei* and *meridionalis* in our trees are also at odds with current notions regarding their relationships. In particular, *andrei* was not closely related to *vauxi aphanes*. Our genetic data indicated that these two taxa were only distantly related within the brown-rumped group: *andrei* as sister to *meridionalis/brachyura* and *vauxi aphanes* as part of the *vauxi/pelagica* clade. Several phenotypic differences were also found to separate the two taxa. In addition to differences in throat patch size/contrast and coloration of upperparts, *andrei* has a distinctly darker lower breast and belly and paler undertail coverts than any examined *vauxi aphanes*, in which the lower breast and belly were lighter than the undertail coverts, and the shafts of its central rectrices were diagnostically smaller in diameter than those of *vauxi aphanes*. Chantler (2000, p. 194) noted similar differences in underparts: *andrei* shows "greater throat contrast, with underparts being sooty-brown from upper breast to vent, and diagnostically in that undertail-coverts are paler (grey-brown) than belly as opposed to darker than belly in Vaux's."

Likewise, *meridionalis* was not closely related to *pelagica*. Marín (1997) found only minor phenotypic differences between these taxa and noted that they could be treated as populations of the same species, although he advocated maintaining them as separate species pending genetic or vocal data. Our genetic data indicated that these species, like *andrei* and *vauxi aphanes*, were rather distantly related within the brown-rumped group despite their superficial similarity: *pelagica* was sister to *vauxi*, whereas *meridionalis* was sister to *brachyura*.

Biogeography and Migration.—Reconstruction of a South American origin for *Chaetura* is not surprising, given the predominantly South American distribution of most species. From there, several species appear to have dispersed to Middle and North America within the past 3 my. Although the more typical pattern is for North American groups to have successfully colonized South America (Smith and Klicka 2010), other counterexamples (e.g., tyrant flycatchers) exist. That the dispersal of species of *Chaetura* to Middle and North America followed the formation of the Isthmus of Panama ca. 3-4 mya (Smith and Klicka 2010) suggests that the two occurrences may have been related, even in a group of birds as mobile as *Chaetura*.

Although many aspects of the behavior and ecology (e.g., nest site and nest construction; Chantler 2000) of swifts of the genus *Chaetura* are remarkably uniform, species in the subgenera *Chaetura* and *Acanthylis* differ markedly in the extent to which they migrate. Long-distance temperate-tropical migration has arisen only in subgenus *Chaetura*, which includes Nearctic-Neotropical migrants *vauxi* and *pelagica* as well as South American austral migrant species *meridionalis* (Sick 1993). This subgenus also contains the intratropical migrant *viridipennis* (Collins 1968).

In contrast, no species in the subgenus *Acanthylis* are thought to be seasonally migratory. To some extent this reflects differences in species' distributions in *Acanthylis* and *Chaetura*. Most species in subgenus *Acanthylis* occur only in the tropical zone; the only exception to this is *cinereiventris*, the nominate subspecies of which inhabits tropical and subtropical southeastern Brazil, northeastern Argentina, and eastern Paraguay. In contrast, three species of subgenus *Chaetura* breed at least partially in the temperate zone, extending as far north as southeastern Alaska (*vauxi*) and southern Canada (*pelagica*) and as far south as northern Argentina (*meridionalis*) and Chile (wintering *pelagica*).

Species limits.—Genetic data, although insufficient to determine species limits of allotaxa under the Biological Species Concept, can provide additional perspective in cases of limited morphological and behavioral data. Previous conclusions about species limits in *Chaetura* were based largely on plumage and morphometrics (e.g., Marín 1997, 2000) and have occasionally incorporated vocal data (e.g., Ridgely and Greenfield 2001). Little is known about the influence of these characters on reproductive isolation in *Chaetura* swifts, increasing the potential value of genetic data in providing another view of differentiation within the genus.

Not surprisingly, levels of genetic divergence separating proposed or recently recognized species from their conspecifics show a great deal of variation (Table 4). At the lower end of the spectrum are *viridipennis* and *chapmani*, which differ by 0.1% in both mitochondrial and nuclear DNA. This level of mtDNA divergence is much lower than that between other putative species pairs and is exceeded in

some cases by differentiation within a subspecies (e.g., within *cinereiventris phaeopygos*). The level of phenotypic differentiation between *viridipennis* and *chapmani* has also been questioned. Marín (1997) based his split of *viridipennis* from *chapmani* on differences in wing length similar to those between *vauxi* and *pelagica*. However, differences in wing length are expected between migratory and sedentary forms of a species, and putative differences in plumage have been questioned. The greenish gloss or iridescence of the type of *viridipennis*, compared to the bluish or purple iridescence of nominate *chapmani* (Corey 1918, Naumburg 1930), has been shown to be related to degree of feather wear (Collins 1968) and is, therefore, not a valid difference even between subspecies. These factors have resulted in some references (e.g., del Hoyo and Collar 2014) retaining *viridipennis* as a subspecies of *chapmani*. Our genetic data are consistent with the view that differences between these taxa are less than those between other taxa of *Chaetura* currently considered separate species.

Levels of divergence within the pale-rumped group of *Acanthylis* were also quite low, ranging from a minimum of 0.3% between *C. spinicaudus aethalea* and *C. egregia*, to a maximum of 1.2% between *C. spinicaudus aetherodroma* and *C. fumosa*. These taxa have long been recognized as valid species under the BSC. That the bGMYC analysis failed to identify these as species simply suggests that this method (and presumably other methods of molecular species delimitation) will fare poorly when speciation has been recent, as appears to be the case in this group of swifts. Ironically, in this cryptic, morphologically conservative group, morphology nevertheless appears to be a better indicator of species limits than does mtDNA.

In contrast, *andrei* is extremely distinct genetically, differing by 3.2% in mtDNA from its former conspecific *meridionalis* and by 7.0% from *vauxi aphanes*, the taxon with which it is currently lumped. Such mitochondrial divergences are well within the range displayed between valid species within *Chaetura*, which in some cases are below 1.0%. Moreover, neither *andrei* + *meridionalis* nor *andrei* + *vauxi aphanes* formed the monophyletic groups typical of, although not required of, species. *Chaetura meridionalis* is likewise highly divergent from *pelagica* (6.5% mtDNA, 1.2% nuclear DNA), a species with which it has been considered to be possibly conspecific (Marín 1997).

Levels of differentiation between *egregia* and its former conspecific *cinereiventris* are also high (Table 4), but this appears to reflect an error in the previous classification of these taxa, because *egregia* is much more closely related to the *martinica-fumosa-spinicaudus* subgroup than to *cinereiventris* (as indicated by Marín 2000). Indeed, all taxa within the pale-rumped subgroup are rather weakly differentiated, ranging from 0.5-1.2% divergence in mtDNA. Levels of divergence for two taxa currently

maintained as subspecies in most or all references, *brachyura ocypetes* and *vauxi richmondi*, when compared to their nominate forms, are at the lower end of this range at 0.6%.

Intraspecific genetic diversity was unexpectedly high in *cinereiventris* and *spinicaudus*, neither of which is monophyletic. *Chaetura fumosa*, now generally considered specifically distinct from *spinicaudus*, differed from the three sampled subspecies of *spinicaudus* by 0.5-1.2% in mtDNA; *fumosa* formed a clade with *spinicaudus spinicaudus* to the exclusion of *spinicaudus aethalea* and *spinicaudus aetherodroma*, which differed from each other and from nominate *spinicaudus* by ca. 1.0-1.2% in mtDNA. Subspecies *spinicaudus aethalea*, currently lumped by most references with nominate *spinicaudus*, did not form a clade with the latter and differed from it appreciably (0.9% divergent in mtDNA, 0.3% in nuclear). A fourth subspecies, previously recognized but also currently lumped with nominate *spinicaudus*, *spinicaudus latirostris*, was not sampled.

Genetic variation within *Chaetura cinereiventris* was considerable: the single individual of nominate *cinereiventris*, which formed a clade with *guianensis* and *sclateri*, differed from them by 2.9%, and these subspecies differed from *phaeopygos*, the sole Central American subspecies, by >6.0% in mtDNA and by >0.7% in nuclear DNA. Such high intraspecific differentiation exceeded most within-subgenus interspecific divergences between species of *Chaetura.* We are aware of no suggestion that *cinereiventris phaeopygos* differs greatly in phenotype from its conspecifics; however, Marín (2000) did indicate that *cinereiventris* may include more than one species and planned an as yet unpublished review of the *cinereiventris* group. Chantler (2000) noted that the underparts of subspecies *occidentalis*, *sclateri*, and *phaeopygos* are noticeably darker than those of *cinereiventris*, *lawrencei*, and *guianensis*, but these groupings do not match the genetic data; likewise, the suggestion that nominate *cinereiventris* is specifically distinct from all other subspecies (del Hoyo and Collar 2014) does not reflect the deepest genetic division within this species. The genetic data for *cinereiventris* and for *spinicaudus*, combined with their highly fragmented distributions (see maps in Chantler 2000), suggest that unrecognized species may be involved, and that further study of morphological and behavioral variation within both species is warranted.

Taxonomic Conclusions and Recommendations

Based on the combined genetic and phenotypic data, we recommend that *C. viridipennis* be returned to its former status as a subspecies of *C. chapmani*. Although it may be subspecifically distinct from *chapmani*, *viridipennis* differs only slightly from that form in both genotype and phenotype, and

does so to a much lesser degree than other *Chaetura* taxa generally considered specifically distinct. In contrast, we recommend that *C. andrei* be recognized as a taxon distinct from *C. vauxi aphanes* and that it be considered specifically distinct from its former conspecific *C. meridionalis*. This would also return *aphanes* to its former status as a subspecies of *C. vauxi*. Other conclusions (e.g., regarding *brachyura ocypetes* and *vauxi richmondi*) are precluded by the limits of our sampling and the ambiguity of the combined phenotypic and genotypic data. We would recommend intensified study of these taxa and such variable species as *C. spinicaudus* and especially *C. cinereiventris*, which show levels of intraspecific genetic differentiation unusually high for species of this genus; taxa such as *C. cinereiventris phaeopygos* of Central America may be specifically distinct.

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Table 1. Species and characteristics of swifts of the genus *Chaetura*. Listed are the 11 species in Dickinson and Remsen (2013) and additional species *C. andrei*, which they considered synonymous with *C. vauxi aphanes*. Number of subspecies follows Dickinson and Remsen (2013), except that for sampling purposes, we recognized two additional subspecies of *C. spinicaudus*: *aethalea* and *latirostris*. Classification of phenotype follows Marín (1997, 2002); gray-rumped species belong to subgenus *Acanthylis* brown-rumped species to subgenus *Chaetura* (Brooke 1970). For use of *spinicaudus* rather than *spinicauda*, see David and Gosselin (2002).

Species	**English Name**	**Phenotype**	**Distribution**	**No. Subspp. Sampled/ No. Subspp.**
fumosa	Costa Rican Swift	Gray-rumped (Pale)	Costa Rica and Panama	monotypic
spinicaudus	Band-rumped Swift	Gray-rumped (Pale)	S. Middle America and n. South America	3/4
martinica	Lesser Antillean Swift	Gray-rumped (Pale)	Lesser Antilles	monotypic
egregia	Pale-rumped Swift	Gray-rumped (Pale)	C-w. South America	monotypic
cinereiventris	Gray-rumped Swift	Gray-rumped (Gray)	S. Lesser Antilles, Middle America, and n-c. South America	4/7
vauxi	Vaux's Swift	Brown-rumped	W. North America, Middle America, n. South America; NA subspecies winters in Middle America	3/7
pelagica	Chimney Swift	Brown-rumped	E. North America; winters in w. South America	monotypic
chapmani	Chapman's Swift	Brown-rumped	S. Middle America and n. South America	monotypic
viridipennis	Amazonian Swift	Brown-rumped	C. South America, winters in n. South America	monotypic
andrei	Ashy-tailed Swift	Brown-rumped	Venezuela	monotypic
meridionalis	Sick's Swift	Brown-rumped	C. South America, winters in n. South America	monotypic
brachyura	Short-tailed Swift	Brown-rumped	S. Lesser Antilles, s. Middle America, n. and c. South America	3/4

Table 2. Tissue/voucher numbers and collecting localities for sequenced individuals of *Chaetura* species and outgroups. No tissues were available for *C. martinica*, *C. c. cinereiventris, C. vauxi aphanes*, and *C. andrei*; numbers in brackets refer to museum specimens from which samples were taken. See Acknowledgments for explanation of museum abbreviations.

Species/Subspecies ID	Tissue number	Locality
C. fumosa 1	FMNH 393015	Costa Rica: Prov. Puntarenas, Rincon, Peninsula de Osa
C. fumosa 2	FMNH 393016	Costa Rica: Prov. Puntarenas, 17 km WSW Chacarita, Alto Mongos, Fila Cal
C. spinicaudus spinicaudus 1	USNM B05190	Guyana: Prov. Essequibo, Waruma River, E bank, ca. 15 river km S Kako River (05° 30’ N, 60° 47’ W)
C. spinicaudus spinicaudus 2	USNM B22118	Guyana: Prov. Upper Takutu - Upper Essequibo, Upper Rewa River (02° 58’ 17’’ N, 58° 35’ 37’’ W)
C. spinicaudus aetherodroma 1	LSUMZ B-11772	Ecuador: Prov. Esmeraldas, El Placer (00° 52' N, 78° 33' W)
C. spinicaudus aetherodroma 2	LSUMZ B-26388	Panama: Prov. Panamá, W. end Serrania de San Blas, 21 km by road NE Chepo
C. spinicaudus aethalea 1	LSUMZ B-35309	Brazil: Pará, 126 km NW Alta Floresta, S bank Rio São Benedito (9°06' 44" S, 56° 56' 32" W)
C. spinicaudus aethalea 2	LSUMZ B-35310	Brazil: Pará, 126 km NW Alta Floresta, S bank Rio São Benedito (9° 06' 44" S, 56° 56' 32" W)
C. martinica 1	[USNM 487572]	Dominica: Central Forest Reserve
C. martinica 2	[USNM 487575]	Dominica: near McFarlin
C. cinereiventris phaeopygos 1	LSUMZ B-27307	Costa Rica: Prov. Limón, 1.5 km S of Bristol Baltimore
C. cinereiventris phaeopygos 2	LSUMZ B-27310	Costa Rica: Prov. Alajuela, 25 km N of Pital, near Boca Tapada
C. cinereiventris guianensis	USNM B05267	Guyana: Prov. Essequibo, Waruma River, E bank, ca. 15 river km S Kako River (05° 30’ N, 60° 47’ W)
C. cinereiventris sclateri 1	MSB:Bird 41910	Peru: Depto. Amazonas, ca. 1.75 km N Gozen
C. cinereiventris sclateri 2	FMNH 320472	Peru: Depto. Cuzco, Tono
C. cinereiventris sclateri 3	FMNH 320475	Peru: Depto. Cuzco, Tono
C. cinereiventris cinereiventris	[MVZ 167217]	Paraguay: Depto. Itapúa, Hotel El Tirol, ca. 4 km NE Capitán Miranda
C. egregia 1	MSB:Bird 37281	Peru: Depto. Madre de Dios, Alerta
C. egregia 2	FMNH 320471	Peru: Depto. Cuzco, Tono
C. egregia 3	LSUMZ B-9194	Bolivia: Depto. Pando, Nicolás Suarez, 12 km by road S of Cobija, 8 km W on road to Mucden
C. vauxi vauxi 1	AMNH DOT15579	USA: Washington, Kings County, Seattle
C. vauxi vauxi 2	UAM 17562	USA: Alaska, Haines Borough, Haines

C. vauxi richmondi 1	FMNH 393010	Costa Rica: Prov. Guanacaste, 17 km SSW Santa Cruz, Cerro Vista al Mar
C. vauxi richmondi 2	FMNH 393011	Costa Rica: Prov. Guanacaste, 17 km SSW Santa Cruz, Cerro Vista al Mar
C. vauxi aphanes 1	[USNM 656481]	Venezuela: Aragua, Portachuelo Pass
C. vauxi aphanes 2	[USNM 656482]	Venezuela: Aragua, Portachuelo Pass
C. pelagica 1	FMNH 368202	USA: Illinois, Cook County, Chicago, McCormick Place
C. pelagica 2	USNM B08929	USA: Virginia, Loudoun County, Dulles International Airport
C. chapmani 1	USNM B05266	Guyana: Prov. Essequibo, Waruma River, E bank, ca. 15 River Km S Kako River (05° 30’ N, 60° 47’ W)
C. chapmani 2	USNM B14165	Guyana: near Linden (06° 01’ N, 58° 12’ W)
C. chapmani 3	LSUMZ B-73389	Brazil: Amazonas, Munic. Manaus, Km 15 Road ZF-2, ca. 65 km N Manaus
C. viridipennis	FMNH 389717	Brazil: Rondonia, Cachoeira Nazaré, W bank Rio Ji-Paraná
C. andrei 1	[AMNH 477326]	Venezuela: Altagracia, Orinoco
C. andrei 2	[AMNH 477327]	Venezuela: Altagracia, Orinoco
C. meridionalis 1	KUMNH 142	Paraguay: Depto. Concepción, Parque Nacional San Luis
C. meridionalis 2	KUMNH 418	Paraguay: Depto. Concepción, Parque Nacional San Luis
C. meridionalis 3	KUMNH 3717	Paraguay: Depto. Itapúa, Parque Nacional San Rafael, San Pedro Mi (26° 31'S, 55° 48'W)
C. brachyura brachyura 1	USNM B13134	Guyana: Wiwitau Mountain, East Rupinuni Savannah (02° 52’ N, 59° 16’ W)
C. brachyura brachyura 2	USNM B14209	Guyana: Linden Highway, St. Cuthbert's Mission Road (06° 18’ N, 58° 13’ W)
C. brachyura brachyura 3	MSB:Bird 43027	Peru: Depto. San Martín, ca. 3.5 km E Incaico
C. brachyura cinereocauda	LSUMZ B-20238	Brazil: Amazonas, Munic. Novo Airao, Arquipelago das Anavilhanas (60° 45' W, 02° 45' S)
C. brachyura ocypetes	LSUMZ B-67035	Peru: Depto. Tumbes, Parque Nacional Cerros de Amotape, El Platano (04° 07' 46" S, 80° 37' 13" W)
Neafrapus cassini	AMNH DOT 10650	Central African Republic: Sangha-Mbaere Prefecture, 1 km N Bayanga
Hirundapus caudacutus	USNM B30253	Russia: Chitinskaya Oblast', Krasnochikoiskiy Rayon, 98 km S, 97 km E Krasnyi Chikoi, at upper Chikoi valley
Apus apus	USNM B07814	England: Co. Suffolk, Mildenhall Air Force Base

Table 3. Primers used for amplifying ND2 from museum specimens of species of *Chaetura*. Primers with suffix "sw" and L5965 were designed specifically for this study. L5215 was taken from Hackett (1996); L5216, H5766, and H6313 from Sorenson et al. (1999); and L5219 from Zwiers et al. (2008). H5977sw and H6113sw were modified from Zwiers et al. (2008).

Primer Name	Sequence 5' – 3'
L5215	TATCGGGCCCATACCCCGAAAAT
L5216	GGCCCATACCCCGRAAATG
L5219	CCCATACCCCGAAAATGAGWSG
L5374sw	AGCCATCAYYCCHCTCATCGC
L5375sw	AGCCATCATCCCACTCATCGCA
L5388sw	CATCGCAAAACACCACCACC
H5388sw	GCRGCYTCGATGGCYCGTGG
L5390sw	TCATCGCAAAACAYCACCAYCCA
H5390sw	TTGCRGCYTCGATGGCTCGT
H5397sw	AARTAYTTGRTTGCRGCYTCGAT
L5419	GAAGCTGCAACAAAATACTT
L5530sw	CCACCCCATCTCATGTGCCCT
H5538sw	AGTCCGAGTTTTATTGCAATKGCTGT
H5564sw	TGGGAATCAGAAGTGGAATGGGACT
L5565sw	TYGCRATRAARCTCGGRCTWG
L5697sw	GCCCACTCACTAAACCCAGCCC
H5706sw	GGCGGCTGAGGAAATTGCTATGGT
L5758sw	GGCTGAATAGGGCTTAACCAAAC
H5766	RGAKGAGAARGCYAGGATYTTKCG
L5853sw	ACAACCCCAAACTAACCCTACTAACCT
H5863sw	ACGGTGATGGTTATTAGGCAGT
H5881sw	TTGTGTTTAGGGTGAGGAACACGG
L5965	AARMCCCNAYACTAAAYGC
H5977sw	GWCCRGCTARGGAYAGCAGRGTDA
L6076sw	CARGAAMTAACYTCARCAGC
L6099sw	GCCACAATCATCACTCCTCTCCC
L6104sw	CNCTCCTCTCCCTHCTA
L6106sw	CNCTCCTCTCCCTHCTAGG
H6113sw	TAGTAYGYAGGCGGAGRTARAAG
L6251sw	CTCTCCACCCTACTCCTCCC
H6257sw	AGTGGCAAGGATTATGGGGG
H6313	ACTCTTRTTTAAGGCTTTGAAGGC

Table 4. Data on genetic differentiation from their current or former conspecifics for proposed, recently recognized, or formerly recognized species of *Chaetura*. The last column indicates whether the taxa in the first and second columns form a monophyletic group.

taxon	separate species from?	mtDNA divergence	nuclear divergence	monophyletic with supposed conspecifics?
viridipennis	*chapmani*	0.1%	0.1%	yes
fumosa	*spinicaudus spinicaudus*	0.5%	0.1%	yes
	spinicaudus aetherodroma	1.2%	0.0%	no
	spinicaudus aethalea	1.0%	0.4%	no
ocypetes	*brachyura* *brachyura/cinereocauda*	0.6%	0.1%	yes
richmondi	*vauxi vauxi/aphanes*	0.6%	0.5%*	yes
egregia	*cinereiventris phaeopygos*	3.6%	0.4%	no
	cinereiventris guianensis	5.0%	0.5%	no
	cinereiventris sclateri	5.4%	0.5%	no
	cinereiventris cinereiventris	4.9%	n/a	no
andrei	*meridionalis*	3.2%	n/a	no
	vauxi aphanes	7.0%	n/a	no
meridionalis	*pelagica*	6.5%	1.2%	no

*nuclear divergence between *vauxi vauxi* and *vauxi richmondi* only; no nuclear data were available for *vauxi aphanes*

Figure Legends

Figure 1. MRE bootstrap tree from RAxML analysis of the combined nuclear and mitochondrial data for *Chaetura* species and outgroups (outgroups *Apus apus* and *Hirundapus caudacuta* not shown due to space constraints). Values above the branches are (1) ML bootstrap support values from the RAxML analysis, (2) ML bootstrap support values from the IQ-tree analysis, and (3) Bayesian posterior probabilities from the MrBayes analysis. See text for details of analyses. Asterisks indicate bootstrap support values of 100 and posterior probabilities of 1.0.

Figure 2. Ancestral range reconstruction (left) and evolution of seasonal migratory behavior (right) in *Chaetura* swifts based on the time-calibrated *BEAST maximum clade credibility tree. Ancestral range reconstructions are the most likely state for each node inferred under the DEC+J model from BioGeoBEARS; see the text for information on the coding of areas. BioGeoBEARS indicated a South American origin with independent colonizations of North and Middle America and the Caribbean. Seasonal migratory behavior reconstructions are the most likely state under a 1-rate model and indicate that each evolution of seasonal migration in *Chaetura* was independent. Time scale in millions of years before present (MYA).