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RESEARCH ARTICLE

Comparative Historical Demography of Migratory and Nonmigratory Birds from the Caribbean Island of Hispaniola

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Abstract Islands offer unique opportunities for studies of evolution and historical demography. We hypothesized that wintering North American migrant bird species would show genetic evidence of population expansion over recent millennia due to the expansion of their breeding distributions following the retreat of the Laurentide ice sheet. In contrast, we presumed that non-migratory species would exhibit more stable historical demographies. We used mtDNA sequences from 649 individuals of 16 avian species on the Caribbean island of Hispaniola to test this prediction. Mismatch distributions did not differ significantly between migrants and non-migrants. However, neutrality indices indicated population expansion in the migrant species, as well as two non-migratory resident species with extensive distributions. Evidence of population expansion was less consistent in other non-migratory residents. We infer that climate prior to the Last Glacial Maximum significantly reduced effective population sizes of most migratory North American bird populations and some resident Hispaniolan bird populations. Our data further revealed that mismatch statistics were poorly correlated with and less informative than the neutrality test statistics, a consideration for future demographic studies.

Keywords Avian evolution · Mismatch distribution · Tajima's D · Fu and Li's D and F · Mitochondrial DNA · Migration

Introduction

The inference of historical demography from variation in mtDNA sequences was first pioneered for human populations (Rogers and Harpending 1992), but is now widely applied. The distribution of pairwise nucleotide differences between mtDNA sequences (i.e., individuals) provides information about historic population sizes and how they have changed over time. These demographic changes (or lack thereof) can then be related to environmental and life-history characteristics of populations. Historical demography has figured prominently in evolutionary studies of one or a few species (Merilä et al. 1997; Bos et al. 2008; Hawley et al. 2008; Lerner et al. 2009; Norgate et al. 2009; Reding et al. 2010), but few reports have simultaneously compared population histories of many species within the same region. By comparing several species from the same geographic area, one can disentangle species characteristics from regional effects to better understand factors that cause demographic change (Walker and Avise 1998).

The Greater Antilles (Fig. 1) offer an excellent opportunity to study historical demography because the islands are sufficiently large and isolated to support endemic

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Fig. 1 Map of the West Indies, with an expanded view of Hispaniola showing the boundary between Haiti (*west side* of Hispaniola) and the Dominican Republic (*east side*). The *black* area represents the Sierra de Bahoruco National Park where the samples were collected

species (e.g., those confined to an island), yet they are close enough to North America to support large numbers of wintering migrants (Ricklefs and Bermingham 2008a). Among the West Indies, Hispaniola (Fig. 1) has the highest number of bird species, including ~95 North American migrants, ~105 breeding resident species, and 31 endemic species (Latta et al. 2006). Habitats on Hispaniola are diverse and include grasslands, dry forest, moist forest, pine forest, and land converted to agriculture. Three parallel mountain ranges run east to west across the island and reach altitudes exceeding 3,000 m (Latta et al. 2006). Hispaniola also has ten offshore islands that provide habitat and breeding grounds for both resident and migrant birds (Latta et al. 2006). Throughout much of the Pliocene and portions of the Pleistocene, the island was further bisected by a deep marine channel that separated Hispaniola into two North and South paleo-islands. The mountains, deep valleys, rivers, and lakes create natural barriers to dispersal that have promoted differentiation of populations on the island (Townsend et al. 2007; Sly et al. 2010). These geographic features have interacted with Pleistocene climate change to create a shifting mosaic of habitat suitable for each of the resident species. During glacial periods, the climate of the West Indies was cooler and drier than at present (Bonatti and Gartner 1973; Curtis et al. 2001), potentially increasing the area of suitable habitat for

montane species and restricting the distributions of tropical forest species (Higuera-Gundy et al. 1999).

In this study, we considered the evolution and demographic history of 16 species of birds based on genetic data from resident and wintering populations on Hispaniola. The individual phylogeographic histories of a few species from the West Indies (such as the Bananaquit, *Coereba flaveola*, Bellemain et al. 2008, 2012), including endemics to Hispaniola (the Chat-Tanagers, *Calyptophilus*, Townsend et al. 2007), have been determined using genetic techniques, but here we ask whether general demographic patterns emerge in an assemblage of birds from the same region. If so, such patterns might inform us about environmental factors that have influenced populations across the millennia. Homogeneous demographic histories across a species assemblage would imply that general environmental impacts (such as climate or geological events) shaped them similarly (Bos et al. 2008; Qu et al. 2010). Alternatively, species-specific influences related to diet, habitat distribution, social organization, pathogens, or other factors could result in idiosyncratic demographic histories.

Demographic histories also might vary according to migratory tendencies. Hispaniolan bird species can be classified as migrant species that breed in North America and spend only a part of the year on Hispaniola, or as non-migrant permanent residents. Non-migrants include species that are endemic to the island, as well as "regional residents" that inhabit Hispaniola year-round but also occur on other islands or on the mainland. Some regional residents on Hispaniola are isolated genetically from populations of the same species on other islands, as in the case of the Bananaquit (Bellemain et al. 2008) and the Red-legged Thrush (*Turdus plumbeus,* Ricklefs and Bermingham 2008b). Some endemic populations show evidence of phylogeographic divergence that corresponds to ancient sea barriers; *Microligea* divergence coincides with the divide between the North and South paleo-islands, and *Phaenicophilus* divergence is associated with the historical isolation of the Tiburon peninsula (Sly et al. 2011). Do these geographic distributions, determined in part by migration and colonization tendencies, shape historical demographies similarly across different species? We reasoned that migrant species are most likely to exhibit population expansions due to the increase in available breeding habitat following the retreat of the Laurentide ice sheet. This was the largest glacier in North America and it covered all of Canada and the Northeast section of the US (Pielou 1991; Avise 1992; Avise and Walker 1998; Norgate et al. 2009). If retreat of this ice sheet opened new breeding habitat, migrant species should exhibit more dynamic population demographies relative to non-migratory residents and endemics. We captured wild birds in the field, extracted DNA from blood samples, and then used mtDNA sequences to critically evaluate these ideas.

Materials and Methods

Field Work and Natural History

Avian blood samples were obtained in the Aceitillar sector of the Dominican Republic's Sierra de Bahoruco National Park (Fig. 1). All fieldwork was conducted under collecting permits issued by the government of the Dominican Republic. Animal care and use protocols were approved by the Institutional Animal Care and Use Committee at the University of Missouri-St. Louis. Field methods are described in detail in Latta and Ricklefs (2010). Blood was collected by venipuncture and stored in PureGene lysis buffer. DNA was extracted using ammonium acetate purification and isopropanol precipitation (PureGene kit, Gentra Systems).

We sampled ten non-migratory species: *Coereba flaveola* (CFA), *Columbina passerina* (CPA), *Elaenia fallax* (EFA), *Loxigilla violacea* (LVI), *Microligea palustris* (MPA), *Myiarchus stolidus* (MST), *Phaenicophilus palmarum* (PPA), *Spindalis dominicensis* (SDO), *Todus subulatus* (TSU), and *Turdus plumbeus* (TPL). MPA, PPA, SDO and TSU are endemic to Hispaniola; EFA, LVI, MST and TPL are restricted to the Caribbean islands; CFA and CPA are more broadly distributed. CFA occurs throughout the Caribbean and Central and South America, and CPA is distributed throughout the Caribbean, the southern United States, Central America, and northern South America. These non-migratory species vary with regard to their food preferences: CFA is a nectarivore; CPA is primarily a granivore; MPA and TSU are insectivores; SDO is a frugivore; the other five species are omnivores. LVI, MPA, MST, TSU and TPL reside in dry forest and scrub areas. SDO is found in tropical moist forests. EFA is found primarily in pine and montane forests. CPA inhabits scrub, open pastures and fields. CFA and PPA occur in all habitat types. Thus, there is considerable variation in distribution, food preference, and typical habitats for these non-migrants.

We sampled six migrant species including *Dendroica caerulescens* (DCR), *Dendroica discolor* (DDI), *Dendroica palmarum* (DPA), *Dendroica tigrina* (DTI), *Mniotilta varia* (MVA), and *Seiurus aurocapilla* (SAU). All are generally insectivorous. On the breeding grounds, these six species are distributed widely in northern and eastern North America and thus have large geographic distributions compared to the endemic or regional resident species. On the Hispaniolan wintering grounds, DDI and DPA are generally found in dry scrub and pine habitats, while the other four species are found primarily in dry and moist forests. All six species are common or abundant on Hispaniola. They differ somewhat in timing of migration, but generally reside on Hispaniola from September to April (Latta et al. 2006). In all cases, the wintering grounds tend

to be much smaller (in geographic extent) than the breeding areas. This is especially true for the four *Dendroica* species that predominately winter in the West Indies (Latta et al. 2006). These species often travel in mixed company, males tend to arrive before females, and adults typically migrate at different times than the young (Zimmerman 1998).

Molecular Genetics

We analyzed the mitochondrial NADH dehydrogenase 2 subunit (ND2) gene because conserved flanking regions allow PCR primers to consistently amplify the gene in diverse avian taxa, yet the gene itself harbors considerable nucleotide variability (Sorenson 2003). We used published primers and also developed our own (Table 1) to amplify and sequence ND2 across our entire species assemblage. The published external primers are situated in conserved regions and amplify well across all species, but our internal primers are positioned in a more variable region and thus we developed specific internal primers for some species. PCR amplifications were conducted in 20 µl total volumes using ~40 ng template DNA, NEB *Taq* polymerase (1U), 0.025 U *Pfu* DNA polymerase, 1.5 mM of $MgCl_2$, 0.3 µM of each primer (H6313 and L5216; Table 1), 1.5 mM $MgCl_2$, 10 mM Tris–HCl, 50 mM KCl, 0.5 mg/ml BSA, and 0.2 mM of each dNTP. PCRs were performed in Eppendorf Mastercyclers with the following conditions: 94°C for 2 min followed by 31 cycles of 94°C for 30 s, 55°C for 30 s, and 72°C for 30 s, concluding with a 5 min extension at 72°C. Reactions were purified using a low sodium acetate precipitation and subsequently quantified using a Nanodrop8000 (Thermo Scientific) spectrophotometer and by gel electrophoresis.

Sequencing reactions were conducted in 10 µl total volumes with ~50 ng of template DNA using BigDye version 3.1 chemistry and a Prism 3730XL sequencer (Applied Biosystems, Foster City, CA, USA). Contigs were aligned and edited in Sequencher 4.7 (Gene Codes Corp., Ann Arbor, MI) from at least four overlapping bidirectional sequences, providing twofold coverage of the gene. Each individual chromatogram was checked by eye to confirm the polymorphisms identified by Sequencher 4.7. Haplotypes were deposited in GenBank (accession numbers JN568591-JN568809).

Analyses of Historical Demography

Mismatch distributions, which represent the number of pairwise differences among all pairwise haplotype comparisons within a species, were created in Arlequin 3.1 with 100 bootstrap coalescent simulations to determine confidence intervals (Excoffier et al. 2005). Unimodal mismatch distributions point to population expansion, multimodal or uniform distributions suggest stable populations at long-term equilibrium, and distributions with only one or two haplotypes indicate a severe bottleneck in the past (Slatkin and Hudson 1991; Rogers and Harpending 1992). We also used Arlequin to calculate the raggedness index (r), which quantifies the smoothness of the mismatch distributions, and the sum of the squared deviations (SSD) which compares the observed mismatch to the expected mismatch distribution under population expansion (Harpending et al. 1993). These statistics can be used to make or corroborate demographic inferences. For example, recently-expanded populations usually have smooth mismatch distributions with SSD and raggedness indices below 0.05. Because these indices are comparisons of empirical data to theoretical expectations, raggedness indices below 0.05 suggest population expansion. We also created haplotype networks for each species using TCS (Clement et al. 2000). Star-like

Table 1 Primers used in this study

Primer name	Sequence: 5'-3'	Species
H6313[a]	ACT CTT RTT TAA GGC TTT GAA GGC	All species
L5216[b]	GGC CCA TAC CCC GRA AAT G	All species
H5766[a]	RGA KGA GAA RGC YAG GAT YTT KCG	CFA, LVI
L5758[a]	GGN GGN TGA ATR GGN YTN AAY CAR AC	CFA, LVI
ND2-IR01	CCT AGG TGD GAG ATK GAK GAG AAR GC	DDI, DPA, DTI, LVI, MPA, PPA
ND2-IR02	ATD GAD GAG AAR GCY ADR ATT TTW CG	CFA, CPA, DPA, MPA, MST
ND2-IR03	GTT TGR TTD AGB CCY ATY CAB CCY CC	CFA, DDI, DPA, LVI, PPA, SAU, SDO, TPL, TSU
ND2-IF01	GGR GGV TGR ATR GGN CTN AAY CAR AC	CFA, CPA, DDI, DPA, DTI, LVI, MPA, MST, PPA, SAU, SDO, TPL, TSU
ND2-IF02	CTA GGA GGR TGR ATR GGY CTM AA	DPA, LVI, PPA, TSU

All other primers were primers developed as part of this study to sequence one or more species. H6313 and L5216 are external primers, whereas all others are internal primers

Citations for previously published primers: [a]Johnson and Sorenson 1998; [b]Sorenson et al. 1999

haplotype networks are characteristic of population expansion whereas complicated or irregular haplotype networks are more indicative of populations at demographic equilibrium.

Genetic diversity statistics, including haplotype number (h), number of polymorphic sites (s), haplotype diversity (hd), nucleotide diversity (π), and mean number of pairwise differences were estimated in Arlequin and in DnaSP (Rozas et al. 2003). Several processes, including recombination, selection, selective sweeps, and population growth or decline, can create distinctive patterns of DNA polymorphisms. Recombination is usually not an issue with mtDNA sequences, but selection can confound demographic inferences. We used Tajima's D (Tajima 1989) to compare the observed distribution of polymorphisms to the distribution expected under selective neutrality. We also calculated Fu and Li's D^* and F^* statistics (Fu and Li 1993), two additional tests of neutrality, using DnaSP. [Because these two statistics were so highly correlated (Table 4) we only refer to F^* in the text.] Significantly negative values of Tajima's D and Fu and Li's F^* statistics indicate population expansion or selection. Fu and Li's F^* statistic is more powerful at detecting background selection than Tajima's D (Fu, 1997), thus if F^* is significant but D is not, this suggests background selection. We compared neutrality statistics and mismatch distributions across species to make broad inferences about historical processes.

To verify that the coalescence of contemporary haplotypes occurred before the last glacial maxima (LGM), we calculated the time since expansion using the equation $t = \tau/2\mu$ (Schneider and Excoffier 1999), where τ is an index of time since expansion calculated from the mismatch distribution. We employed ND2 substitution rates ranging from 1 to 3% divergence per million years (μ: 5.6×10^{-6}–1.56×10^{-5}), bracketing the estimate of $\mu = 9.72 \times 10^{-6}$ (Eo and DeWoody 2010).

Results

We analyzed blood samples from 649 individuals from 16 species, averaging 40.6 individuals per species (range 20–59) (Table 2). From these blood samples, we extracted DNA and sequenced 1,041 bp of the mtDNA ND2 gene. The mean number of haplotypes per species was 13.7 (range 4–28) and the mean number of polymorphic sites per species was 16.4 (3–47) (Table 2). The overall mean nucleotide diversity was 0.0024 (range 0.0012–0.0053) and mean haplotype diversity was 0.724 (0.2–0.958). Coalescence estimates ranged from 25,814 to 154,266 years ago, thus our genetic data can reflect expansions that occurred since the LGM. We used three aspects of the mismatch distributions constructed from the DNA sequences to infer historical demographies. First, the shape of the distributions themselves can be informative. Twelve of the 16 species we assayed had smooth, unimodal mismatch distributions indicative of population expansions (Fig. 2). Second, the SSD was consistent with a model of population expansion for 15 of the 16 species (i.e., they had SSD values <0.05; Table 3). Finally, Harpending's r indicated population expansion for 8 of the 16 species (Table 3).

All 16 species displayed negative values of Tajima's D and Fu and Li F^* (i.e., all 48 values in Table 3 were negative). However, not all values of these parameters differed significantly from 0, and significant values were distributed unequally among species with respect to geographic distributions, reported in detail below. Ecological parameters such as habitat and food preference may be important, but they were not evaluated in light of the demographic statistics due to lack of power. Further studies of species with well defined habitat and food preferences would be better suited to help determine what role ecological parameters play in the demographic history of a species.

Non-Migrants

The number of individuals sampled for each of the four endemic species ranged from 29 to 58, the number of haplotypes ranged from 4 to 13, and the haplotype diversity ranged from 0.20 to 0.81 (Table 2). None of the SSD values associated with the mismatch distributions were significant (Fig. 2) and only SDO had a raggedness index below 0.05 (Table 3). Three of the 4 endemic species (PPA, SDO and TSU) had significantly negative Tajima's D values (−0.66 to −2.32) but only one of these (SDO) additionally had a significantly negative F^* value (−0.64 to −2.42), indicating population expansion (Table 3).

Samples of the six regional resident species ranged from 20 to 59, the number of haplotypes from 8 to 28, and the haplotype diversity ranged from 0.63 to 0.96 (Table 2). The CFA and CPA mismatch distributions closely resembled the null model of population expansion, whereas EFA, LVI, MST, and TPL did not (Fig. 2). The significant SSD values for LVI and MST also do not support population expansion. In addition, the haplotype networks for CFA and CPA were more star-like in their appearance (Fig. 3). CFA, CPA, LVI, and TPL had raggedness indices below 0.05, but none were significant (Table 3). Of six Tajima's D values for regional resident species (range −0.86 to −2.21), only those for CFA (−2.21) and CPA (−2.14) were significantly negative (Table 3). These two species also had significantly negative values of Fu and Li's F^* (−2.66 and −2.43).

Table 2 The species used in this study, their range status, sample size (*n*), and genetic diversity

Species (abbr) common name	Type	Food	Habitat*	*n*	h	s
Coereba flaveola (CFA) Bananaquit	R	N	All types; most numerous in mesic scrub and forest, particularly moist broadleaf forest	58	16	17
Columbina passerina (CPA) Common Ground Dove	R	G	Scrub; pastures, fields, clearings and second growth (pine at higher elevations)	35	11	15
Elaenia fallax (EFA) Greater Antillean Elaenia	R	O	Primarily pine, sometimes more open forest and wet broadleaf forest	20	8	7
Loxigilla violacea (LVI) Greater Antillean Bullfinch	R	O	Drier habitats, dense growth, broadleaf	59	28	36
Myiarchus stolidus (MST) Stolid Flycatcher	R	O	Dry forest and desert scrub, pine and moist broadleaf forest, and shade coffee	31	11	9
Turdus plumbeus (TPL) Red-legged Thrush	R	O	Xeric and second growth woodland to humid forest	32	10	13
Microligea palustris (MPA) Green-tailed Ground-tanager	E	I	Lowland dry scrub, desert thorn, and dry broadleaf; moist broadleaf at higher elevations	58	12	8
Phaenicophilus palmarum (PPA) Black-crowned Palm-tanager	E	O	All types; from desert scrub to pine forest	56	12	13
Spindalis dominicensis (SDO) Hispaniolan Spindalis	E	F	Montane forest, evergreen forest, broadleaf, pine	32	13	15
Todus subulatus (TSU) Broad-billed Tody	E	I	Dry limestone forest, shade coffee, lowland and montane second growth	29	4	3
Dendroica caerulescens (DCR) Black-throated Blue Warbler	M	I	Forest; shade coffee, humid montane broadleaf forest, pine	20	9	9
Dendroica discolor (DDI) Prairie Warbler	M	I	Open woodland; dry thorn scrub forest, and margins of mangroves	57	18	20
Dendroica palmarum (DPA) Palm Warbler	M	I	Open woodland; drier open areas	58	21	24
Dendroica tigrina (DTI) Cape May Warbler	M	I	Forest; thorn scrub, dry forest, shade coffee, transitional broadleaf forest, pine	43	15	15
Mniotilta varia (MVA) Black-and-white Warbler	M	I	Forest; dry forest, moist broadleaf forest, shade coffee, pine	20	8	12
Seiurus aurocapilla (SAU) Ovenbird	M	I	Forest; dry forest, moist broadleaf forest, shade coffee	42	23	47

Genetic diversity is represented by the number of haplotypes (h) and the number of polymorphic sites (s). R, regional residents (species that breed on the island); E, endemics (endemic to Hispaniola); and M, migrants (do not breed on the island). In the primary food column; N, nectarivore; G, granivore; I, Insectivore; F, frugivore; and O, omnivore

* Latta et al. 2003, 2006, and http://avibase.bsc-eoc.org/

Migrants

The six migratory species were each represented by 20–58 individuals, 8–23 haplotypes, and haplotype diversities between 0.59 and 0.90 (Table 2). Haplotype networks were star-like, characteristic of population expansion (Fig. 3). One exception was SAU, which had a more complex haplotype network that may be indicative of admixed populations. DTI had a significant SSD value inconsistent with population expansion (Fig. 2). DDI, DPA and DTI had raggedness indices below 0.05, but none were significant (Table 3). Tajima's *D* values for all six migrant species were significantly negative (−1.77 to −2.41), and all but one species (MVA) showed significantly negative Fu and Li's *F** (−1.81 to −3.06).

Multi-species comparisons

In general, *D* and *F** decrease (become more negative) from endemics to regional residents and from regional residents to migratory species. However, separate comparisons by one-way ANOVA (SAS GLM procedure) of the neutrality test statistics revealed no significant difference across the three distribution types (*D*: $F = 2.11$,

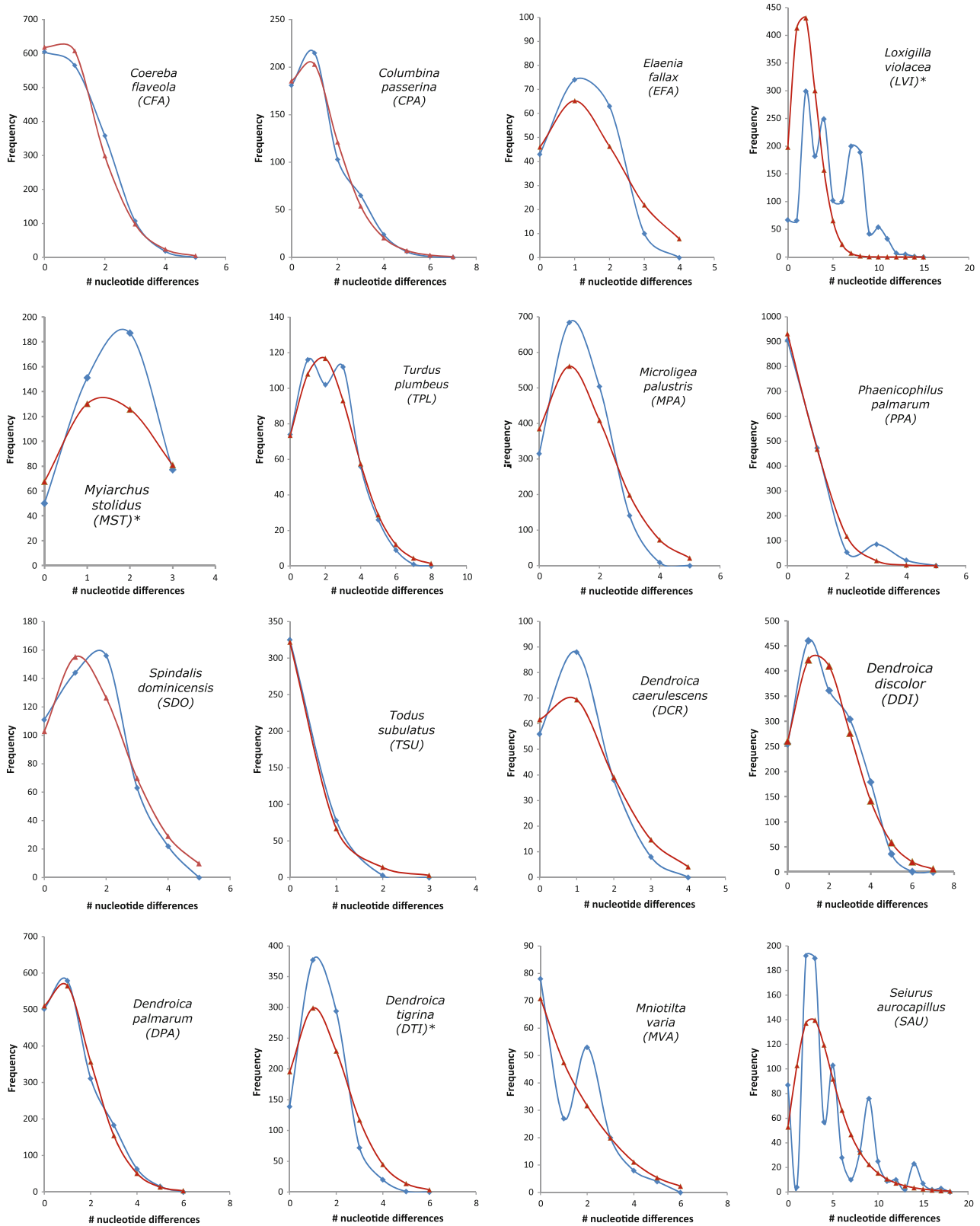


Fig. 2 Mismatch distributions of all 16 species. *Blue* lines with diamonds represent the observed data. *Red* lines with triangles represent the null model of population expansion. The first six mismatch distributions are regional residents, the next four are endemics, and the last six are migrant species. The *x*-axis represents the number of nucleotide differences and the *y*-axis represents the frequency of each class. *Significant mismatch distribution (Color figure online)

Table 3 Genetic diversity and neutrality statistics for each species

Species common name	Type	π	hd (SD)	SSD	r	Tajima's D	Fu and Li's D^*	Fu and Li's F^*
Bananaquit (CFA)	R	0.0024	0.634 (0.07)	0.0021	***0.042***	***−2.21****	***−2.40****	***−2.66****
Common Ground Dove (CPA)	R	0.0012	0.696 (0.08)	0.0018	***0.049***	***−2.14****	***−2.25****	***−2.43****
Greater Antillean Elaenia (EFA)	R	0.0020	0.774 (0.08)	0.0140	0.111	−1.27	−1.36	−1.45
Greater Antillean Bullfinch (LVI)	R	0.0053	0.958 (0.01)	0.1032*	***0.036***	−1.27	−1.90	−2.09
Stolid Flycatcher (MST)	R	0.0020	0.893 (0.03)	0.0196*	0.132	−0.86	−1.10	−1.32
Red-legged Thrush (TPL)	R	0.0034	0.851 (0.04)	0.0028	***0.026***	−1.08	−0.56	−0.76
Green-tailed Ground-tanager (MPA)	E	0.0013	0.809 (0.04)	0.0133*	0.116*	−0.66	−0.53	−0.64
Black-crowned Palm-tanager (PPA)	E	0.0023	0.412 (0.08)	0.0040	0.155	***−2.32****	−1.99	−2.22
Hispaniolan Spindalis (SDO)	E	0.0014	0.776 (0.07)	0.0047	***0.049***	***−2.01****	***−2.23****	***−2.42****
Broad-billed Tody (TSU)	E	0.0014	0.200 (0.10)	0.0015	0.404	***−1.73****	−0.75	−0.67
Black-throated Blue Warbler (DCR)	M	0.0019	0.705 (0.11)	0.0117	0.124	***−2.10****	***−2.06****	***−2.24****
Prairie Warbler (DDI)	M	0.0018	0.840 (0.04)	0.0027	***0.036***	***−1.77****	***−2.77****	***−2.92****
Palm Warbler (DPA)	M	0.0026	0.696 (0.07)	0.0012	***0.041***	***−2.41****	***−2.78****	***−3.06****
Cape May Warbler (DTI)	M	0.0023	0.846 (0.04)	0.0199*	***0.049***	***−1.87****	***−2.46****	***−2.65****
Black-and-white Warbler (MVA)	M	0.0031	0.590 (0.13)	0.0259	0.126	***−2.22****	−1.65	−1.81
Ovenbird (SAU)	M	0.0043	0.899 (0.04)	0.0360	0.099**	***−2.07****	−2.50	***−2.77****

π = nucleotide diversity; hd = haplotype diversity; SSD = sum of squared deviations; r = raggedness index. Italicized and bolded values indicate population expansion

R = regional residents (species that breed on the island), E = endemics (endemic to Hispaniola), and M = migrants (do not breed on the island). Fu and Li's D^* and F^* statistics are more powerful in detecting background selection, therefore when D is significant and D^* and F^* are not, it is typically characteristic of expansion (Fu 1997)

* $P < 0.05$

** $P < 0.01$

$P = 0.16$; D^*: F = 3.30, $P = 0.07$; F^*: F = 3.29, $P = 0.07$). However, migrants ($n = 6$) had significantly lower values compared to non-migrants ($n = 10$) with respect to D^* and F^* (D^*: F = 6.70, $P = 0.02$; $F^* = 6.42$, $P = 0.024$), and D approached significance (F = 3.99, $P = 0.066$).

Significantly negative neutrality parameters (D, D^*, and F^*) indicate population expansion or selection. About half the species in our sample showed strong evidence for population expansion, having significantly negative values of both D and F^* (Table 3). Five of the six migrant species (DCR, DDI, DPA, DTI, and SAU) showed significantly negative F^*, whereas only three of ten non-migrants (CFA, CPA, and SDO) did. Note that the three species with significant Tajima's D statistics but nonsignificant Fu and Li's F^* statistics had the lowest haplotype diversity in the sample (PPA, hd = 0.41; TSU, hd = 0.20; MVA, hd = 0.59), which could indicate a lack of statistical power for these species.

Discussion

Rogers and Harpending (1992) showed that historical changes in population size can affect the genetic diversity of a population, and that information about population history can be extracted from DNA sequences. Many subsequent studies have illustrated the ability of contemporary DNA sequences to provide insight into the dynamics of historical populations (Merilä et al. 1997; Bos et al. 2008; Hawley et al. 2008; Lerner et al. 2009; Norgate et al. 2009; Reding et al. 2010; Qu et al. 2010). We evaluated molecular signatures of historical demography using mismatch distributions and demographic statistics. Our inferences concerning avian historical demography are broad in the sense that they consider multiple species simultaneously. Thus far, most studies on historical demography have focused on one or more populations of a particular species. Comparison of demographic histories of a sample of species can indicate the impact of general environmental factors on populations within a region and permit a statistical appraisal of the influence of species characteristics on demography within a common environmental context. For example, Qu et al. (2010) compared the evolutionary histories of five avian species in different regions of the Qinghai-Tibetan plateau. Three species from the platform of the plateau (Tibetan Snow Finch, *Montifringilla adamsi*; Blanford's Snow Finch, *Pyrgilauda blanfordi*; Horned Lark, *Eremophila alpestris*) showed signs of population expansion, while two species (Twite,

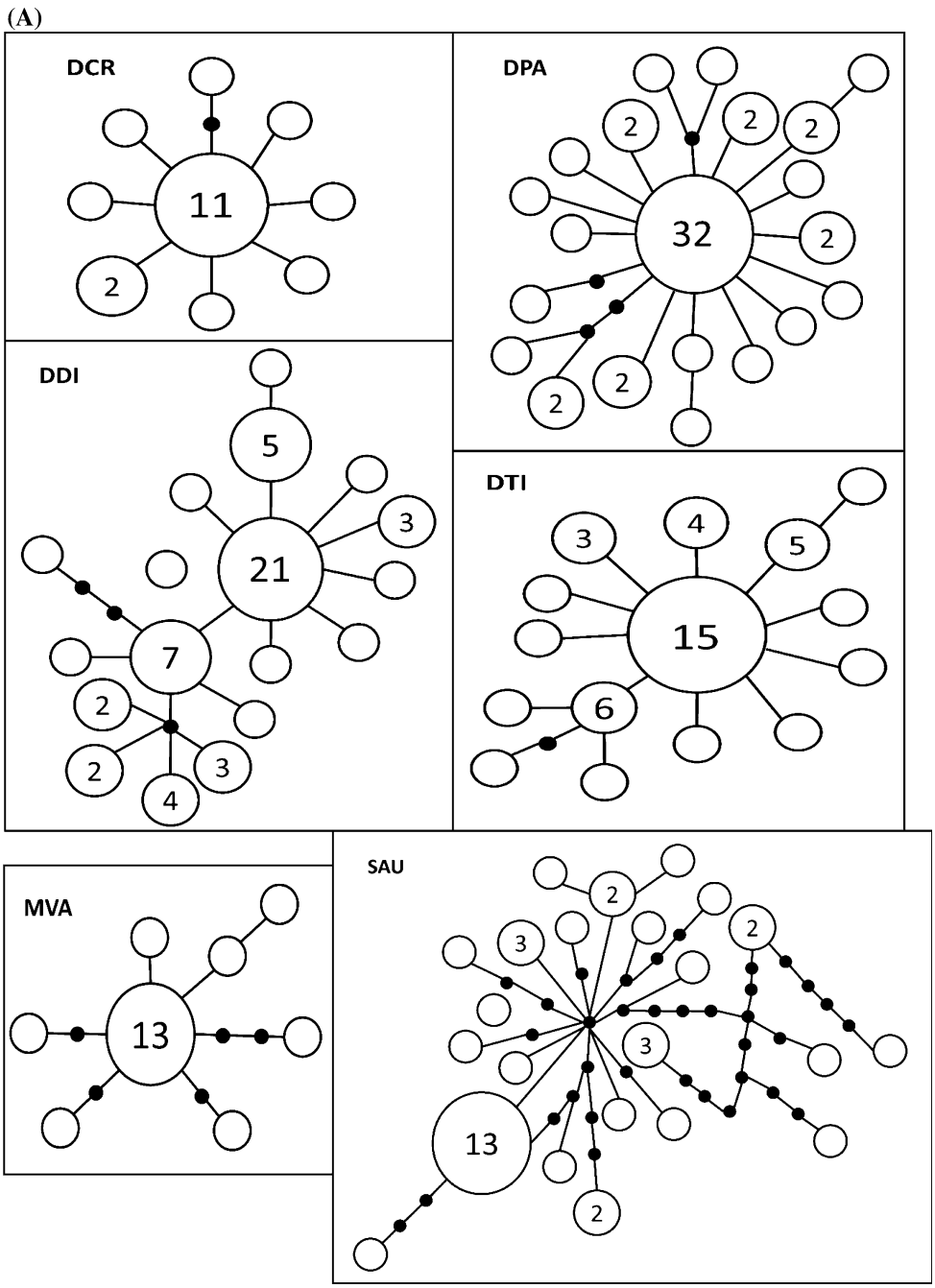


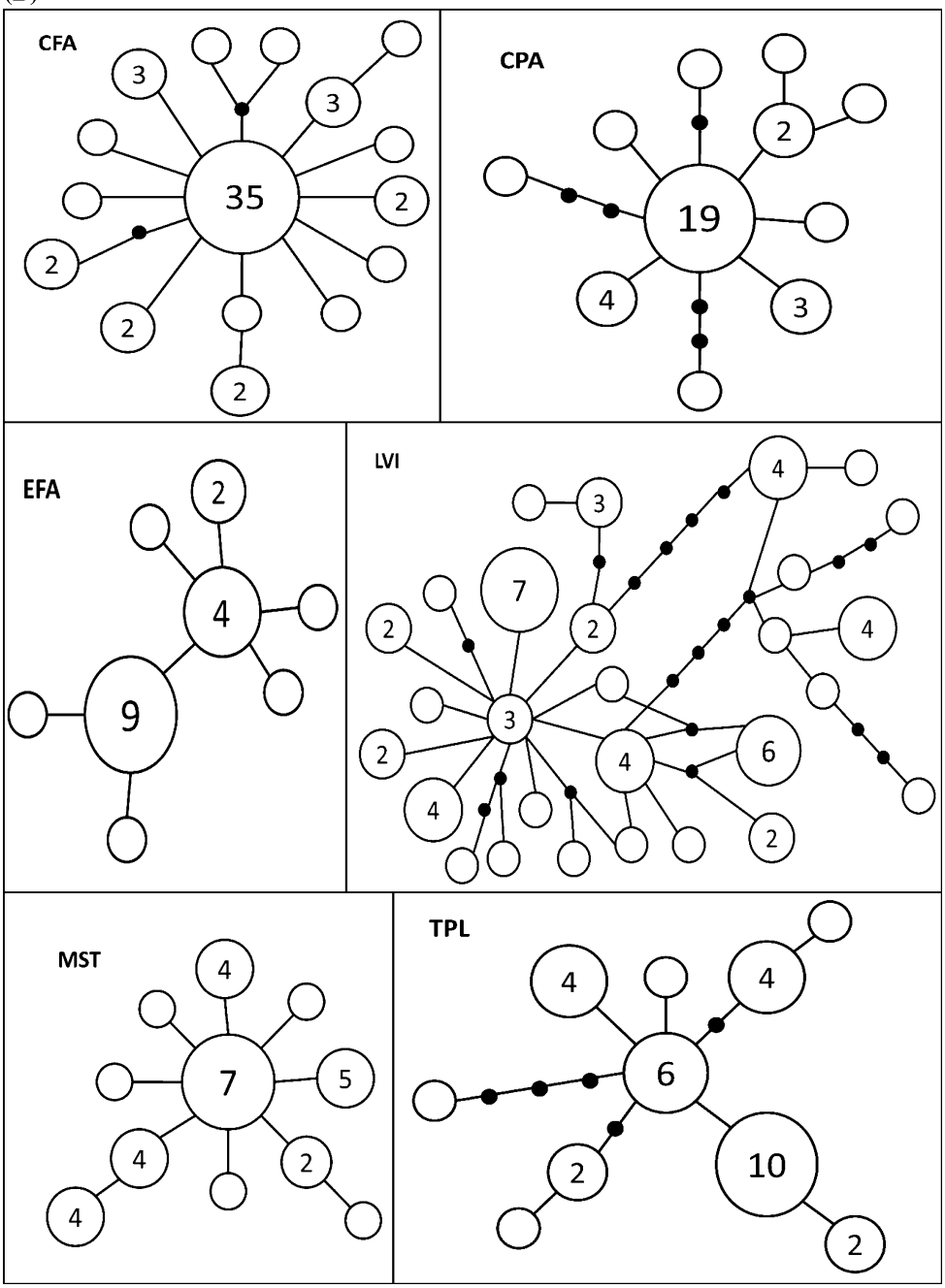
Fig. 3 Haplotype networks for (**a**) Migrants; (**b**) Regional residents; and (**c**) Endemics. Star-like haplotypes generally are indicative of expanding populations while complex networks are characteristic of stable populations. The smallest (*red*) circles are *nodes* that represent a polymorphic site. The number of individuals with a given haplotype is shown by the numbers inside the circles; circles with no numbers inside represents a haplotype found in a single individual

Carduelis flavirostris; Black Redstart, *Phoenicurus ochruros*) distributed at the edge of the plateau had historically stable populations. They suggested that substantial parts of the platform species' distributions were ice-covered and its post-glacial retreat would have allowed for population expansion; species at the edge of the plateau were less affected by glaciers. Our data sets, on migratory and resident species on a Caribbean island, provide similar insights into the evolutionary dynamics of New World birds.

Non-Migrants

Of the four species endemic to Hispaniola, three do not show evidence of recent expansion, suggesting that effective population sizes in the past were not reduced compared to the present. This suggests that suitable habitat has been stable for these species, at least more so than with regional resident or migratory species. MPA represents a monotypic genus endemic to Hispaniola. The genus *Phaenicophilus*,

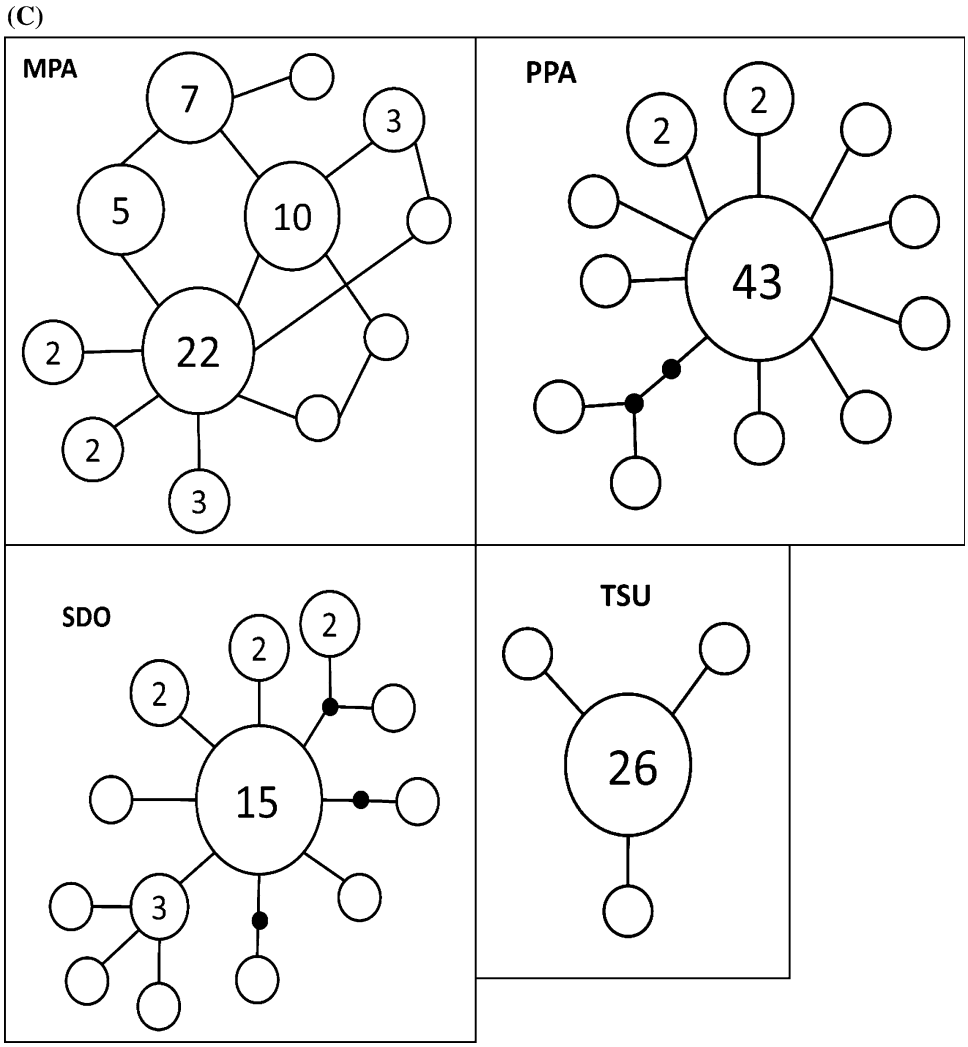
Fig. 3 continued



which contains PPA, is also endemic to Hispaniola. Todies (TSU) comprise an older lineage with endemic species on Hispaniola, Cuba, Jamaica, and Puerto Rico (Overton and Rhoads 2004; Mayr and Knopf 2007) and thus may have undergone multiple cycles of expansion and contraction that are not reflected in contemporary mtDNA sequences. Only one endemic species (SDO, *Spindalis dominicensis*) exhibited evidence of recent population expansion.

Among the regional residents, two populations (Bananaquit (CFA) and Common Ground Dove (CPA)) present evidence of recent population growth, whereas the other four do not. The Bananaquit and the Common Ground Dove are widely distributed throughout the West Indies and in Central and South America; their geographic ranges are not restricted to the Caribbean and thus recent population expansion seems plausible for these two species because they have not been confined to the islands. However, the range expansion of CFA in the Greater Antilles occurred prior to the coalescence times of the island populations, which are reciprocally monophyletic (Bellemain et al. 2008), and therefore is related to the genetic evidence of local population expansion. The four species with historically stable populations are

Fig. 3 continued



restricted to the Greater Antilles: Greater Antillean Elaenia (EFA; Hispaniola and Jamaica), Greater Antillean Bullfinch (LVI; Hispaniola, Jamaica, Bahamas), Stolid Flycatcher (MST; Hispaniola and Jamaica), and the Red-legged Thrush (TPL; Greater Antilles, Bahamas). Mitochondrial DNA evidence suggests that the Hispaniolan populations of EFA, LVI, and TPL are monophyletic, but that Hispaniolan and Jamaican populations of MST are recently derived (unpubl. data; Ricklefs and Bermingham 2008b). Thus, while it is tempting to relate recent population expansion on Hispaniola to recent expansion through the Greater Antilles, only two of the Hispaniolan resident species are not deeply monophyletic populations (CPA and MST). Among the 8 monophyletic resident populations on Hispaniola, only two (CFA and SDO) show evidence of recent expansion.

Migrants

Five of the 6 migrant species show evidence supporting recent population expansion (Table 3), which differs significantly from 3 of 10 island residents ($G = 4.13$, df $= 1$, $P = 0.042$). Eighteen of the 24 test statistics among these 6 migrant species provide strong evidence for expansion. This supports our initial hypothesis that North American residents had restricted ranges during glacial maxima and that their populations have expanded since the Last Glacial Maximum, as shown for other migratory species based on North American samples (Mila et al. 2000, 2006, 2007; McKay 2009). Collectively, these results give strong support to the idea that migratory species have undergone significant population growth in their recent evolutionary history.

Multi-species Approach

Our strategy was not to consider intraspecific variation across multiple genomic loci, but across multiple biological species. This approach is particularly well-suited to broad taxonomic surveys because (a) conserved regions provide binding sites for "universal" PCR primers; (b) the

Table 4 Pearson's correlation coefficients between SSD, *r*, *D*, *D**, and *F**

	SSD	r	D	D*	F*
SSD	–	−0.168 (*0.534*)	0.257 (*0.336*)	−0.003 (*0.993*)	−0.013 (*0.963*)
r	–	–	0.042 (*0.879*)	0.496 (*0.051*)	0.548 (*0.028*)
D	–	–	–	0.722 (*0.002*)	0.708 (*0.002*)
D*	–	–	–	–	0.996 (*<0.001*)
F*	–	–	–	–	–

P values are italicized and in parentheses next to the corresponding correlation coefficient ($n = 16$)

clonal nature of mtDNA provides immediate discrimination of sequence haplotypes (i.e., cloning is not required); and (c) its fourfold smaller effective population size means mtDNA genomes respond more rapidly to genetic drift than do nuclear genomes (Zink and Barrowclough 2008). Nevertheless, there are tradeoffs in a broad molecular approach that does not focus in detail on a single population or lineage, but considers an entire species assemblage. Most obviously, the addition of nuclear genes would increase the number of nucleotides sequenced, expand the number of polymorphic sites surveyed, and could decrease the variance associated with our summary statistics (although genes with very different substitution rates might also mute demographic signatures). Furthermore, additional loci would help rule out selective sweeps that might otherwise be mistaken as signatures of historical population bottlenecks followed by demographic expansion. To evaluate the possibility of natural selection on the mtDNA ND2 gene, we performed McDonald-Kreitman (1991) tests in DnaSP (Rozas et al. 2003) on populations consistent with expansion (i.e., CFA, CPA, DCR, DDI, DPA, DTI, SDO and SAU). McDonald-Kreitman tests were conducted using orthologous sequences from *Gallus gallus* and from a congeneric species, but revealed no significant deviations from selective neutrality (data not shown). These results are similar to those of Zink et al. (2006) on genetic variation in the ND2 gene in the Eurasian nuthatch (*Sitta europaea*).

Across the 16 species we surveyed, there was broad congruence among the demographic statistics *D*, *D**, and *F**. Table 3 shows that all 48 values were negative, 54% (26 of 48) significantly so. The distribution of significant values varied among our geographic categories, presumably because of both statistical and biological factors.

Statistically, there were strong correlations among *D*, *D**, and *F** (particularly between *D** and *F**) but only weak correlations involving SSD and *r* (Table 4). SSD and *r* values are derived from the mismatch distributions. Mismatch distributions, SSD, and *r* typically have large confidence intervals because they are based on the polymorphic sites contained among a few haplotypes and thus these tests are very conservative (Felsenstein 1992), meaning that it is less likely that the null model of population expansion will be rejected (Schneider and Excoffier 1999). This is especially apparent in the SSD values (Table 3) where 15 of the 16 species have values <0.05 but most are statistically insignificant. Mismatch distribution data is also more complex to interpret because different features of the wave (e.g., slope, magnitude, and wavelength) can communicate varying characteristics of the population (Rogers and Harpending 1992). Different trajectories of population growth and even population bottlenecks can produce the same wave characteristics. These factors and the discordances among our mismatch distributions, raggedness indices, and neutrality statistics suggest to us that mismatch distributions are considerably less informative (and perhaps less objective) than *D*, *D**, and *F**. Mismatch statistics may have some value in a comparative context (i.e. across multiple populations or species), but the neutrality statistics (*D*, *D**, and *F**) are more easily and objectively interpreted.

Ramos-Onsins and Rozas (2002) found Class I statistics (based on segregating site frequency; *D*, *D**, and *F**) are more powerful than Class III statistics (based on pairwise sequence differences or mismatch distribution; SSD and *r*) at detecting population expansion under different scenarios (varying sample size, number of segregating sites, time, and degree of expansion). Thus, inferences solely based on mismatch distributions are overly conservative, more nuanced, and less powerful than neutrality test statistics. Therefore mismatch distributions and statistics should only be used in conjunction with neutrality statistics.

Beyond these statistical factors, the population histories of the species in each category appear to have strongly influenced their evolutionary demographies. The endemic species, and many populations of the regional residents, have been restricted to Hispaniola for periods that exceed the coalescence time of their mtDNA genes. A variety of factors conspire to restrict historical population sizes and/or dampen the long-term growth of island populations. For instance, island populations are subject to repeated episodes of colonization and extinction. Furthermore, maximum population size has been restricted by island size and/or dispersal ability. Also, in non-glaciated mountainous regions, species may have shifted up and down in elevation and therefore could have avoided strong bottlenecks and subsequent expansions (Hewitt 1996). This is possible for individuals on Hispaniola because of the high mountain ranges that cover much of the island. Thus, relative to migratory species, it is less likely that as a group, resident

populations have undergone recent concerted increases in size.

In contrast, it seems more likely that migrant species which today breed in continental North America may have dramatically increased their range (and associated population sizes) since the LGM roughly 15,000 years ago (Mila et al. 2000; Mila et al. 2006, 2007b; Hughes and Hughes 2007; McKay 2009). Thus, migrant species have had more opportunity for sustained population growth. Regional residents appear to have experienced growth rates intermediate to endemics or migrants as their distributions (range limits) are intermediate in scope.

It is also important to note that there have been many glacial climate cycles in the Quaternary. Thus, all the North American migrant populations have probably gone through population contractions (bottlenecks) and then experienced subsequent expansions. As glaciers receded, long distance dispersers would have quickly filled these areas and thus experienced exponential growth by being on the leading edge of expansion (Hewitt 1996, 2000). This scenario leaves the trailing edge of dispersers with less available unoccupied habitat to support population expansion. In the case of Neotropical birds, trailing edge dispersers include endemics and some regional residents, whereas leading edge dispersers include migratory and other species capable of sustained flight. Because the leading edge dispersers already occupy the habitat and serve as competitors, trailing edge dispersers may not experience exponential growth. The incidence and rate of population growth is reflected in the contemporary mtDNA mismatch distributions and demographic statistics. Whereas many species may have experienced cycles of population expansion and contraction during the Pleistocene, our molecular data support the notion that leading edge migratory species generally experienced more pronounced population growth than non-migratory species.

One point not yet discussed is that our data indicate the genetic diversity of the migrant populations is similar to the genetic diversity in island populations. One might expect greater genetic diversity in continental populations of migrants because of their much more expansive breeding habitat and corresponding larger population sizes. However, our mtDNA data reveal no such trend. This might be due to slower growth of effective population size (N_e) relative to census size in migratory (i.e., rapidly expanding) populations because long-term N_e is disproportionately affected by generations with small N_e (Lande and Barrowclough 1987). Thus, there is an expected lag time before N_e reflects demographic expansions. Microsatellite data may be informative in this comparative context, as population genetic diversity in birds is associated with the extent of available habitat and, by inference, N_e (Eo et al. 2011). This may be mediated in part by increases in heterozygosity associated with population expansion, followed by increases in genome-wide mutation rates (Amos et al. 2008).

Conclusions

Mitochondrial DNA sequence variation can provide considerable insight into a species' evolutionary history. By simultaneously considering the evolutionary history of a broad species assemblage, we can begin to better understand what common evolutionary forces have shaped these lineages. Our data have revealed that populations with different contemporary distributions (e.g., endemics versus migrants) also experienced different demographic histories. Mismatch data were largely independent from neutrality statistics, which revealed that migratory populations have been similarly affected by climate change in that most species show genetic signatures of population growth that coincides with glacial retreat after the LGM. This is in contrast to island endemics that, in comparison, have maintained more stable population sizes over many millennia. Our results are based on a single population sample from each species; future efforts should consider multiple populations from each species to produce species-level inferences. This multi-species approach to historical demography has the potential to reveal broad evolutionary patterns comparable to the distributional histories evident in comparative phylogeographic studies (Avise 2000).

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References

Amos, W., Flint, J., & Xu, X. (2008). Heterozygosity increases microsatellite mutation rate, linking it to demographic history. *BMC Genetics, 9*, 72.

Avise, J. C. (1992). Molecular population structure and the biogeographic history of a regional fauna: A case history with lessons for conservation biology. *Oikos, 63*, 62–76.

Avise, J. C. (2000). *Phylogeography*. Cambridge, MA: Harvard University Press.

Avise, J. C., & Walker, D. (1998). Pleistocene phylogeographic effects on avian populations and the speciation process. *Proceedings of the Royal Society of London Series B-Biological Sciences, 265*, 457–463.

Bellemain, E., Bermingham, E., & Ricklefs, R. E. (2008). The dynamic evolutionary history of the bananaquit (*Coereba flaveola*) in the Caribbean revealed by a multigene analysis. *BMC Evolutionary Biology, 8*, 240.

Bellemain, E., Gaggiotti, O., Fahey, A. & Ricklefs, R.E. (2012). Demographic history and genetic diversity in the West Indian *Coereba flaveola* populations. *Genetica,* in revision.

Bonatti, E., & Gartner, S., Jr. (1973). Caribbean climate during Pleistocene ice ages. *Nature, 244*, 563–565.

Bos, D. H., Gopurenko, D., Williams, R. N., & DeWoody, J. A. (2008). Inferring population history and demography using microsatellites, mitochondrial DNA, and major histocompatibility complex (MHC) genes. *Evolution, 62*, 1458–1468.

Clement, M., Posada, D., & Crandall, K. (2000). TCS: A computer program to estimate gene genealogies. *Molecular Ecology, 9*, 1657–1660.

Curtis, J. H., Brenner, M., & Hodell, D. A. (2001). Climate change in the circum-Caribbean (Late Pleistocene to Present) and implications for regional biogeography. In C. A. Woods & F. E. Sergile (Eds.), *Biogeography of the West Indies. Patterns and Perspectives* (pp. 35–54). Boca Raton, FL: CRC Press.

Eo, S. H., & DeWoody, J. A. (2010). Evolutionary rates of mitochondrial genomes correspond to diversification rates and to contemporary species richness in birds and reptiles. *Proceeding of the Royal Society B-Biological Sciences, 277*, 3587–3592.

Eo, S. H., Doyle, J. M., & DeWoody, J. A. (2011). Genetic diversity in birds is associated with body mass and habitat type. *Journal of Zoology (London), 283*, 220–226.

Excoffier, L., Laval, L. G., & Schneider, S. (2005). Arlequin ver. 3.0: An integrated software package for population genetics data analysis. *Evolutionary Bioinformatics, 1*, 47–50.

Felsenstein, J. (1992). Estimating effective population size from samples of sequences: Inefficiency of pairwise and segregating sites as compared to phylogenetic estimates. *Genetical Research, 59*, 139–147.

Fu, Y. (1997). Statistical tests of neutrality of mutations against population growth, hitchhiking and background selection. *Genetics, 147*, 915–925.

Fu, Y., & Li, W. H. (1993). Statistical tests of neutrality of mutations. *Genetics, 133*, 693–709.

Harpending, H. C., Sherry, A. R., Rogers, A. R., & Stoneking, M. (1993). Genetic structure of ancient human populations. *Current Anthropology, 34*, 483–496.

Hawley, D. M., Briggs, J., Dhondt, A. A., & Lovette, I. J. (2008). Reconciling molecular signatures across markers: mitochondrial DNA confirms founder effect in invasive North American house finches (*Carpodacus mexicanus*). *Conservation Genetics, 9*, 637–643.

Hewitt, G. M. (1996). Some genetic consequences of ice ages, and their role in divergence and speciation. *Biological Journal of the Linnean Society, 58*, 247–276.

Hewitt, G. M. (2000). The genetic legacy of the Quaternary ice ages. *Nature, 405*, 907–913.

Higuera-Gundy, A., Brenner, M., Hodell, D. A., Curtis, J. H., & Leyden, B. W. (1999). A 10,300 ^{14}C yr record of climate and vegetation change from Haiti. *Quaternary Research, 52*, 159–170.

Hughes, A. L., & Hughes, M. K. (2007). Coding sequence polymorphism in avian mitochondrial genomes reflects population histories. *Molecular Ecology, 16*, 1369–1376.

Johnson, K. P., & Sorenson, M. D. (1998). Comparing molecular evolution in two mitochondrial protein coding genes (cytochrome *b* and ND2) in the dabbling ducks (Tribe: Anatini). *Molecular Phylogenetics and Evolution, 10*, 82–94.

Lande, R., & Barrowclough, G. F. (1987). Effective population size, genetic variation, and their use in population management. In M. E. Soulé (Ed.), *Viable populations for conservation*. New York, NY: Cambridge University Press.

Latta, S. C., & Ricklefs, R. E. (2010). Prevalence patterns of avian Haemosporida on Hispaniola. *Journal of Avian Biology, 41*, 25–33.

Latta, S. C., Rimmer, C. C., & McFarland, K. P. (2003). Winter bird communities in four habitats along an elevational gradient on Hispaniola. *Condor, 105*, 179–197.

Latta, S., Rimmer, C., Keith, A., Wiley, J., Raffaele, H., McFarland, K., et al. (2006). *Birds of the Dominican Republic and Haiti*. Princeton, NJ: Princeton University Press.

Lerner, H. R. L., Johnson, J. A., Lindsay, A. R., Kiff, L. F., & Mindell, D. P. (2009). It's not too late for the harpy eagle (*Harpia harpyja*): high levels of genetic diversity and differentiation can fuel conservation programs. *PLoS One, 4*, 10.

Mayr, G., & Knopf, C. W. (2007). A tody (Alcediniformes: Todidae) from the early Oligocene of Germany. *Auk, 124*, 1294–1304.

McDonald, J. H., & Kreitman, M. (1991). Adaptive protein evolution at the Adh locus in *Drosophila*. *Nature, 351*, 654–656.

McKay, B. D. (2009). Evolutionary history suggests rapid differentiation in the yellow-throated warbler *Dendroica dominica*. *Journal of Avian Biology, 40*, 181–190.

Merilä, J., Björklund, M., & Baker, A. J. (1997). Historical demography and present day population structure of the greenfinch, *Carduelis chloris*- an analysis of mtDNA control-region sequences. *Evolution, 51*, 946–956.

Mila, B., Girman, D. J., Kimura, M., & Smith, T. B. (2000). Genetic evidence for the effect of a postglacial population expansion on the phylogeography of a North American songbird. *Proceedings of the Royal Society of London Series B-Biological Sciences, 267*, 1033–1040.

Mila, B., Smith, T. B., & Wayne, R. K. (2006). Postglacial population expansion drives the evolution of long-distance migration in a songbird. *Evolution, 60*, 2403–2409.

Mila, B., Smith, T. B., & Wayne, R. K. (2007a). Speciation and rapid phenotypic differentiation in the yellow-rumped warbler *Dendroica coronata* complex. *Molecular Ecology, 16*, 159–173.

Mila, B., McCormack, J. E., Castaneda, G., Wayne, R. K., & Smith, T. B. (2007b). Recent postglacial range expansion drives the rapid diversification of a songbird lineage in the genus *Junco*. *Proceeding of the Royal Society B-Biological Sciences, 274*, 2653–2660.

Norgate, M., Chamings, J., Pavlova, A., Bull, J. K., Murray, N. D., & Sunnucks, P. (2009). Mitochondrial DNA indicates late Pleistocene divergence of populations of *Heteronympha merope*, an emerging model in environmental change biology. *PLoS One, 4*, 11.

Overton, L. C., & Rhoads, D. D. (2004). Molecular phylogenetic relationships based on mitochondrial and nuclear gene sequences for the Todies (Todus, Todidae) of the Caribbean. *Molecular Phylogenetics and Evolution, 32*, 524–538.

Pielou, E. C. (1991). *After the ice age*. Chicago, IL: University of Chicago Press.

Qu, Y., Lei, F., Zhang, R., & Lu, X. (2010). Comparative phylogeography of five avian species: Implications for Pleistocene evolutionary history in the Qinghai-Tibetan plateau. *Molecular Ecology, 19*, 338–351.

Ramos-Onsins, S. E., & Rozas, J. (2002). Statistical properties of new neutrality tests against population growth. *Molecular Biology and Evolution, 19*, 2092–2100.

Reding, D. M., Freed, L. A., Cann, R. L., & Fleischer, R. C. (2010). Spatial and temporal patterns of genetic diversity in an endangered Hawaiian honeycreeper, the Hawaii Akepa (*Loxops coccineus coccineus*). *Conservation Genetics, 11*, 225–240.

Ricklefs, R., & Bermingham, E. (2008a). The West Indies as a laboratory of biogeography and evolution. *Philosophy Transactions of the Royal Society B-Biological Sciences, 363*, 2393–2413.

Ricklefs, R., & Bermingham, E. (2008b). Likely human introduction of the red-legged thrush (*Turdus plumbeus)* to Dominica, West Indies. *Auk, 125*, 299–303.

Rogers, A. R., & Harpending, H. C. (1992). Population growth makes waves in the distribution of pairwise genetic differences. *Molecular Biology and Evolution, 9*, 552–569.

Rozas, J., Sánchez-DelBarrio, J. C., Messeguer, X., & Rozas, R. (2003). DnaSP, DNA polymorphism analyses by the coalescent and other methods. *Bioinformatics, 19*, 2496–2497.

Schneider, S., & Excoffier, L. (1999). Estimation of past demographic parameters from the distribution of pairwise differences when the mutation rates vary among sites: application to human mitochondrial DNA. *Genetics, 152*, 1079–1089.

Slatkin, M., & Hudson, R. R. (1991). Pairwise comparisons of mitochondrial DNA sequences in stable and exponentially growing populations. *Genetics, 129*, 555–562.

Sly, N. D., Townsend, A. K., Rimmer, C. C., Townsend, J. M., Latta, S., & Lovette, I. J. (2010). Phylogeography and conservation genetics of the Hispaniolan endemic Palm-Tanagers (Aves: *Phaenicophilus*). *Conservation Genetics, 11*, 2121–2129.

Sly, N. D., Townsend, A. K., Rimmer, C. C., Townsend, J. M., Latta, S. C. & Lovette, I. J. (2011). Ancient islands and modern invasions: Disparate phylogeographic histories among Hispaniola's endemic birds. *Molecular Ecology, 20*, 5012–5024.

Sorenson, M. D. (2003). Avian mtDNA primers. http://people.bu.edu/msoren/primers.html.

Sorenson, M. D., Ast, J. C., Dimcheff, D. E., Yuri, T., & Mindell, D. P. (1999). Primers for PCR-based approach to mitochondrial genome sequencing in birds and other vertebrates. *Molecular Phylogenetics and Evolution, 12*, 105–114.

Tajima, F. (1989). Statistical method for testing the neutral mutation hypothesis by DNA polymorphism. *Genetics, 123*, 585–595.

Townsend, A. K., Rimmer, C. C., Latta, S. C., & Lovette, I. J. (2007). Ancient differentiation in the single-island avian radiation of endemic Hispaniolan Chat-Tanagers (Aves : *Calyptophilus*). *Molecular Ecology, 16*, 3634–3642.

Walker, D., & Avise, J. C. (1998). Principles of phylogeography as illustrated by freshwater and terrestrial turtles in the southeastern United States. *Annual Review of Ecology, Evolution, and Systematics, 29*, 23–58.

Zimmerman, J. L. (1998). Migration of Birds. *Circular 16*. Fish and Wildlife Service, U.S. Department of the Interior.

Zink, R. M., & Barrowclough, G. F. (2008). Mitochondrial DNA under siege in avian phylogeography. *Molecular Ecology, 17*, 2107–2121.

Zink, R. M., Drovetski, S. V., & Rohwer, S. (2006). Selective neutrality of mitochondrial ND2 sequences, phylogeography and species limits in *Sitta europaea*. *Molecular Phylogenetics and Evolution, 40*, 679–686.