Examining the Genetic Distinctiveness of the Western Subspecies of Yellow-Billed Cuckoo Coccyzus americanus occidentalis

Author: Farrell, Lindsay L.

Source: Ardea, 101(2) : 165-170

Published By: Netherlands Ornithologists' Union

URL: https://doi.org/10.5253/078.101.0211

Examining the genetic distinctiveness of the western subspecies of Yellow-billed Cuckoo *Coccyzus americanus occidentalis*

Lindsay L. Farrell[1]

Farrell L.L. 2013. Examining the genetic distinctiveness of the western subspecies of Yellow-billed Cuckoo *Coccyzus americanus occidentalis*. Ardea 101: 165–170.

The western subspecies of Yellow-billed Cuckoo *Coccyzus americanus occidentalis* has undergone severe population declines over the last two decades. Numerous petitions have warranted its listing under the Endangered Species Act; however, the question of whether the western subspecies is genetically separated from the eastern *C. a. americanus* form is controversial. Previous research found small, but significant divergence in a single mitochondrial gene based on a limited number of samples. In this study, I analyze 949 base pairs of the same mitochondrial cytochrome *b* gene from additional samples in both the western and eastern subspecies to reassess the evolutionary distinctiveness of the western taxa. Unlike the previous study, I did not find any fixed genetic differences, but only found slight, insignificant genetic differentiation between the subspecies. The separation into distinct subspecies of the eastern and western Yellow-billed Cuckoo might be more recent than can be captured by a single mitochondrial gene; alternatively high levels of gene flow might prevent complete segregation. Interestingly, I found a substantial amount of haplotype variation within the western samples, which may indicate genetic differences between western sampling locations. Future studies incorporating next generation sequencing are recommended to resolve the taxonomic status of Yellow-billed Cuckoo.

Key words: cytochrome *b*, cyt-*b*, mtDNA, Yellow-billed Cuckoo, Coccyzus

[1]Department of Biology, Lakehead University, Thunder Bay, ON, Canada; present address: Roslin Institute, The University of Edinburgh, Easter Bush, Scotland, United Kingdom
(lindsay.farrell@roslin.ed.ac.uk)

Historically, subspecies in birds were defined by differences observed in colour and pattern of plumage, and/or differences in size and proportion of various body regions, which were believed to be genetically based. Intra-specific differences in other aspects of species' biology, e.g. behaviour, were also considered in taxonomy as they may coincide with physical differences implying substantial genetic divergence. Thus, many avian subspecies were described before the advent of quantification of phenotypic traits, molecular methods and modern statistics. Often taxonomy needs to be revisited in order to determine whether the patterns described previously accurately reflect reproductive boundaries (Haig & Winker 2010), especially when conservation efforts and resources may be limited and needed elsewhere.

Once considered to be part of a single interbreeding species, Ridgeway (1887) first suggested Yellow-billed Cuckoos *Coccyzus americanus occidentalis* across the western range be treated as separate subspecies, as he considered them to be morphologically different; larger with proportionally larger and stouter bills, than the eastern *C. a. americanus* form. Slight plumage differences are also observed, with western cuckoos appearing greyer dorsally than the eastern birds, most notably on the crown, and having an orange-yellow, rather than bright yellow, lower mandible (Hughes 1999). Tape recordings support differences in vocalizations between eastern and western birds (Franzreb & Laymon 1993) and there appears to be differences in breeding phenology, with eastern birds breeding considerably earlier than their western counterparts (Franzreb & Laymon

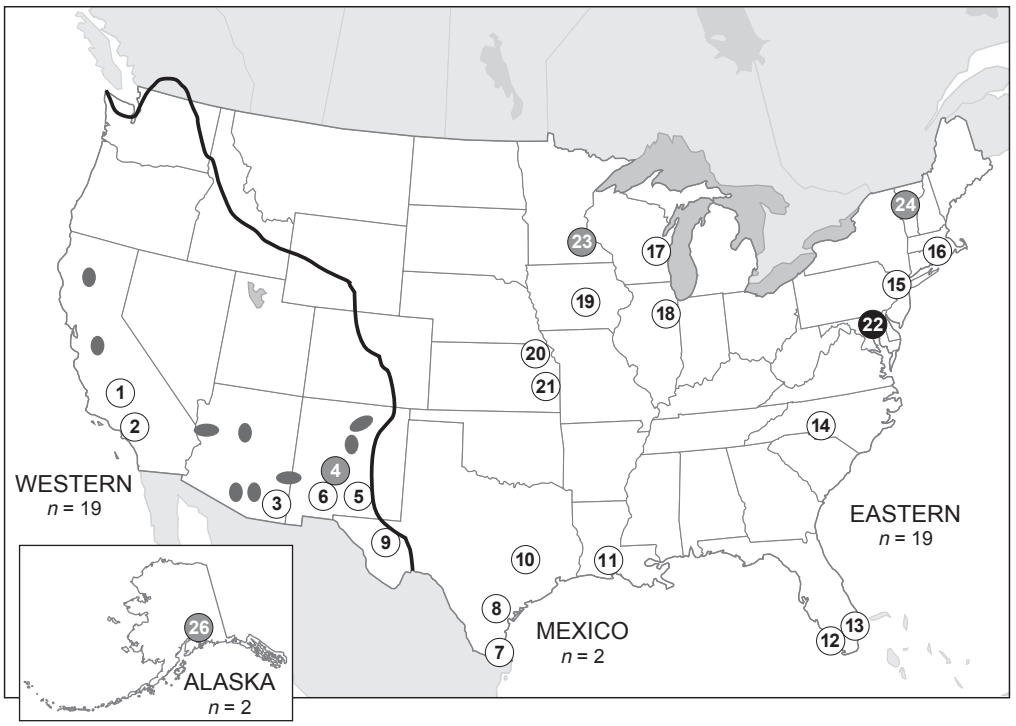

Figure 1. Map showing distribution of Yellow-billed Cuckoo *Coccyzus americanus* samples used in this study. Historical range of western population defined by black line and current populations defined by shaded ovals after Laymon & Halterman (1987). All samples west of line were designated as western population (including Alaska). Similarly, all samples east of line were designated as eastern population. Samples from Mexico AY046909 and AY046908 (not shown) treated as separate population after Banks (1988). Samples taken from Pruett *et al.* (2001) indicated by grey circles. Sample from Hughes (JMH881; 2006) indicated by solid fill. Sample from Alberta Z01.3.1 (not shown) designated as eastern population.

1993). However, several attempts to identify these subspecies based on morphological characters such as plumage, wing and bill lengths have yielded contradictory results (Banks 1988, Banks 1990, Franzreb & Laymon 1993, Hughes 1999).

Currently listed as Priority 3 of imminent immediacy, the western Yellow-billed Cuckoo is included in the annual *Review of Native Species That Are Candidates for Listing as Endangered or Threatened* in the Federal Register under the United States Fish and Wildlife Service (USFWS), but continues to be listed as 'warranted, but precluded'(Federal Register/Vol.77. No.225 November 21, 2012). Meaning, despite the fact it is listed as a Priority 3, until the subspecies is given an Endangered or Threatened listing by the USFWS, no direct conservation action may be undertaken on its behalf under the Endangered Species Act. In response to significant population declines in the remainder of the western subspecies range, Pruett *et al.* (2001) conducted the first genetic study on the Yellow-billed Cuckoo to search for evidence that the two taxa were, in fact, evolutionarily distinct. Based on limited sampling (n = 11), they found that the predominant mitochondrial DNA (mtDNA) haplotypes of the eastern and western subspecies differed by four fixed base pair changes in the cytochrome *b* gene (cyt-*b*, 978 bp). In this study, I analyze mtDNA (cyt-*b*, 949 bp –885 bp of which can be directly aligned with Pruett *et al.* (2001) sequences – including the region with the four fixed differences) from additional samples from both the eastern and western subspecies of Yellow-billed Cuckoo from a broader geographic region, to reassess the evolutionary distinctiveness of these two taxa based on the cyt-*b* gene.

METHODS

Samples were obtained from 31 Yellow-billed Cuckoo individuals, consisting of 20 tissue and 11 blood samples granted from museums and researchers in the field (Table 1) and collected over a wide geographic area of the species' range (Figure 1). In addition to the samples in the current study, sequences from Pruett *et*

Table 1. Locality information of all *Coccyzus americanus* spp. samples used in this study corresponding to continental distribution map (Figure 1) and haplotype network (Figure 2). Pruett *et al*. (2001) samples are listed last and designated with the accessions AF and AY.

EMBL accession	Museum collection number[1]	Sample origin City/State & Country	Number on map	Corresponding haplotype
HE793167	AMNH 7235/381	Shasta County, California, USA	1	13
HE793169	MH119-207555	Kern River, California, USA	2	17
HE793168	MH119-7040	Kern River, California, USA	2	14
HE793170	MH119-207553	Kern River, California, USA	2	19
HE793178	MH119-207576	San Pedro River, Arizona, USA	3	20
HE793171	MH119-207596	San Pedro River, Arizona, USA	3	12
HE793176	MH119-20742	San Pedro River, Arizona, USA	3	7
HE793174	MH119-207535	San Pedro River, Arizona, USA	3	17
HE793172	MH1212-13712	San Pedro River, Arizona, USA	3	12
HE793173	MH1212-13715	San Pedro River, Arizona, USA	3	17
HE793175	MH1212-13719	San Pedro River, Arizona, USA	3	17
HE793177	MH119-207594	San Pedro River, Arizona, USA	3	14
HE793180	NK11992/MSB18032	Albuquerque, New Mexico, USA	4	24
AY046906	NK43108	Albuquerque, New Mexico, USA	4	16
AY046907	NK35431	Albuquerque, New Mexico, USA	4	18
AY046905	NK3993	Albuquerque, New Mexico, USA	4	14
HE793181	NK103366/MSB23134	Bitterlake NWR, New Mexico, USA	5	6
HE793179	NK116168/MSB23827	Socorro, New Mexico, USA	6	11
HE793184	LSUMZ B-23431	Jeff Davis County, Texas, USA	9	27
HE793182	NK11964/MSB18642	Atacosta NWR, Texas, USA	7	25
HE793185	LSUMZ B-37016	Edinberg, Hidalgo County, Texas, USA	8	17
HE793183	LSUMZ B-21785	Travis County, Texas, USA	10	21
HE793186	UWBM 615/2002-130	Cameron, Louisiana, USA	11	5
HE793188	UFNUMB 44090	Naples, Florida, USA	12	17
HE793187	UFNUMB 44087	Ft. Lauderdale, Florida, USA	13	17
HE793189	UWBM 78029/2510	Mecklenburg County, North Carolina, USA	14	17
HE793190	AMNH 7466/Coll619	Suffolk County, New York, USA	15	22
HE793191	UWBM 68159/2000-091	Massachusetts, Barnstable, USA	16	17
HE793192	FMNH 441576	Brown County, Wisconsin, USA	17	22
HE793193	FMNH 363714	DuPage County, Illinois, USA	18	26
HE793194	FMNH 429371	Marion County, Iowa, USA	19	23
HE793195	MBR 2690/89937	Douglas County, Kansas, USA	20	17
HE793196	MBR 27/88591	Jefferson County, Kansas, USA	21	17
HE793197	RAM Z01.3.1	Edmonton, Alberta, CAN	-	3
-	JMH881	Maryland, USA	22	4
AF249271	UAM 10131	Minnesota, USA	23	8
AF249270	UAM 10130	Minnesota, USA	23	9
AY046910	UAM 13376	Rutland, Vermont, USA	24	10
AY046909	CNAV 23931	Veracruz, Mexico	-	1
AY046908	UAM 10354	Veracruz, Mexico	-	2
AF249269	UAM 7059	Ketchikan, Alaska, USA	26	14
AF249268	UAM 6953	Juneau, Alaska, USA	26	15

[1] Specimen numbers refer to source collections: AMNH, American Museum of Natural History, New York; ANSP, Academy of Natural Sciences of Philadelphia; CNAV, Coleccion Nacional de Aves, Mexico; FMNH, Field Museum of Natural History, Chicago; JMH, Janice M. Hughes, Lakehead University; LSUMZ, Louisiana State University Museum of Natural History, Baton Rouge; MBR, University of Kansas Natural History Museum, Lawrence; MH, Murrelet Halterman, University of Nevada, Reno; NK, University of New Mexico, Albuquerque; NWR, National Wildlife Refuge; RAM, Royal Alberta Museum, Edmonton; UAM, University of Alaska Museum, Fairbanks; UF, Florida Museum of Natural History, Gainesville; UWBM, University of Washington Burke Museum of Natural History, Seattle.

al. (2001) were obtained from the European Molecular Biology Laboratory (EMBL) European Nucleotide Archive (www.ebi.ac.uk/ena) and included in all analyses. All samples in this study were grouped *a priori* as eastern or western based on geographic locality and/or morphological differences between the subspecies as recorded and assigned by museum collections, in order to search for the four fixed base pair differences previ-

ously shown to be divergent between the eastern and western forms and compare population pairwise F_{ST} values. Accordingly, in this dataset 21 samples were designated as western and 21 samples as eastern (Figure 1).

Sequencing and data analysis

Genomic DNA from tissue stored in either 100% ethanol or HCL Tris Lysis buffer and blood preserved on FTA cards were isolated using the standard phenol chloroform extraction method. Blank extraction controls were included to check for cross contamination. An 949 bp fragment of the cytochrome *b* (cyt-*b*) gene, 885 bp of which can be directly aligned with Pruett *et al.* (2001) sequences, was amplified for all individuals using the primers L14490 (Sorenson *et al.* 1999) and H16065 (Sheldon *et al.* 2005). In addition, two internal primer pairs (CYTB1: 5'-CAAGGACCTAGTAGGATTCACT-3', and CYTB2: 5'-CACAAATCATCAATTCATCAA-3') were used to sequence cyt-*b* in both directions, ensuring complete gene sequence and correct base identification. PCRs were conducted in 40 μl reaction volumes, with final concentrations of 200 μM dNTPs, 0.2 mM of each primer, 2.0 μM $MgCl_2$, 1 x PCR buffer minus Mg, and 1 U Platinum Taq DNA polymerase (Invitrogen) with remaining volume completed with ddH_2O and DNA template. PCR amplification was performed using a thermal cycler with the following cycling parameters: 94°C for 2 min; 45 cycles of 94°C for 60s, annealing at 50–55°C for 60s, 72°C for 2.3 min. Sequences were generated using the ABI PRISM 3100 Genetic Analyzer (Applied Biosystems). All sequences were aligned and compared to EMBL searches of *C. americanus* cyt-*b* sequences (Appendix 1) to verify sequence identity using Se-Al v. 2.0a11 (Rambaut 2002). Arlequin v.3.1 (Excoffier *et al.* 2005) was used to perform an analysis of molecular variance (AMOVA) and compare population pairwise F_{ST} values. The relationship between all mtDNA haplotypes was examined by constructing a haplotype network with the comparable 885 bp aligned sequences from this study and Pruett *et al.* (2001) using TCS v.1.18 (Clement *et al.* 2000).

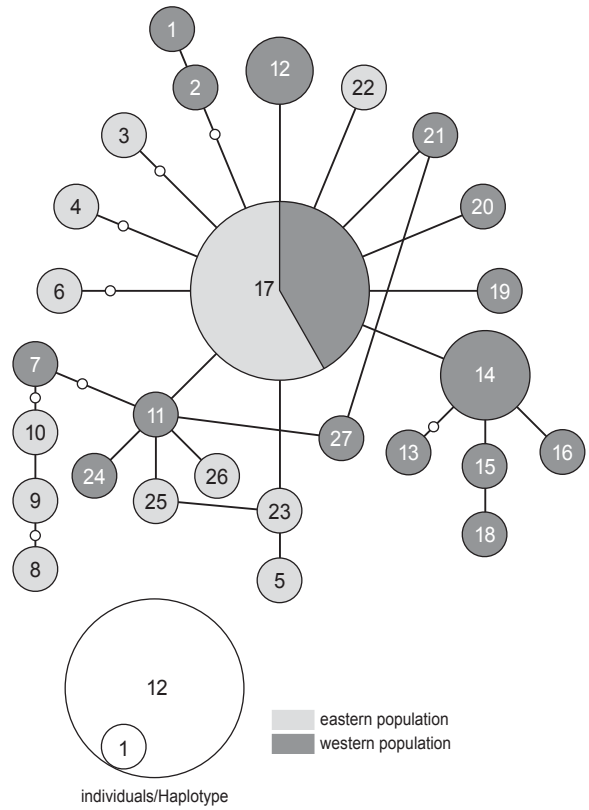

Figure 2. A minimum spanning network inferred from maximum parsimony. The haplotype network is derived from 885 bp of the mtDNA cytochrome *b* gene. Pruett *et al.* (2001) data is incorporated and designated by haplotype groupings: [1, 2], [8, 9, 10], and [14,15,16,18]. The relative sizes of the circles represent the number of individuals contained within each haplotype and pie slices represent the fraction of each population contained within each haplotype. Open circles represent missing haplotypes.

RESULTS

An 949 bp fragment of the cytochrome *b* (cyt-*b*) – 885 bp of which can be directly aligned with Pruett *et al.* (2001) sequences, including the four fixed base pair sites previously identified to differentiate between eastern and western forms – was sequenced for 31 individuals in this data set. Aligned sequences had a base composition of 28.8% A, 35.0% C, 12.6% G, and 23.6% T. No insertions or deletions, frameshift mutations, or unexpected stop codons were detected. Combining this additional sampling with that of Pruett *et al.* (2001), I found no evidence of any fixed genetic differences between groups as identified as eastern and western subspecies in this study. I found 27 haplotypes, with one common haplotype and a star-like radiation of mostly singletons around it (Figure 2). There was some degree of geographic sub-structuring in the western haplotypes, but no significant east–west division of the subspecies. The most common haplotype (17) was found both in the east and the west; the western haplotypes placed within the eastern birds (7, 11 and 24) were sampled in Arizona and New Mexico (Figure 2). The haplotype diversity in eastern sample was 0.0035

(0.0021) and the haplotype diversity in the western sample was 0.9532 (0.0358); standard errors in parentheses. In addition, an AMOVA estimated that 92% of the variation is explained by variation within populations. The AMOVA revealed weak, but not significant genetic structure among geographical regions (F_{ST} = 0.07). Pairwise F_{ST} values among regions are significant between Mexico (haplotype 1 and 2) and all other regions (F_{ST} values between 0.35 and 0.75) while all other F_{ST} values were non-significant (Table 2).

Table 2. Pairwise F_{ST} for Yellow-billed Cuckoo populations used in this study. Significant differences are indicated by asterisk.

	Western	Alaska	Mexico	Eastern
Western	0.00000			
Alaska	0.04544	0.00000		
Mexico	0.43033*	0.75000*	0.00000	
Eastern	0.01317	0.08196	0.34944*	0.00000

DISCUSSION

A previous study examining the distinctiveness of the eastern and western subspecies of Yellow-billed Cuckoo found significant genetic differences between these two taxa based on the mitochondrial cytochrome *b* gene (Pruett *et al.* 2001). To assess the role of limited sampling in the previous study and the evolutionary distinctiveness of the western taxa, I sequenced additional samples from a broader geographic region at the same cytochrome *b* locus. Surprisingly, I was not able to clearly distinguish between these two taxa using a number of statistical analyses. Instead, I documented a substantial amount of haplotype variation within the western samples, which is indicative of local interbreeding and sub-structuring among these fragmented populations. Whether these western populations are interbreeding with other northwest populations not sampled in this study is not known, and further study on the genetic exchange in these declining western populations is recommended. In addition, the mtDNA haplotypes identified in Pruett *et al.* (2001) as unique to the eastern subspecies, grouped with haplotypes from the western group in the current study, and visa versa. Of additional note are the significant pairwise F_{ST} values between the population in Mexico and the other regions, as these two samples from Veracruz (EMBL accession: AY46908 & AY46909; Table 2) contained two unique fixed polymorphisms not shared among other regions. Banks (1988) identified a number of morphological differences in birds from northeastern Mexico and suggested that populations from this region represented a southeastern extension along the Gulf of Mexico from eastern Texas that are isolated from other birds. It would appear that from this new molecular analysis, this population of Yellow-billed Cuckoos are distinctly different not only in morphology, but genetically divergent as well, however, I report this finding with caution and recommend additional sequencing of individuals is needed in order to confirm a distinctly different Mexican population of Yellow-billed Cuckoo.

The main explanation the author can provide for the differences between this and the previous study is firstly a limited sample size (n = 11, Pruett *et al.* 2001; n = 31, this study), which in these declining populations will continue to be an issue. If Pruett *et al.* (2001) had sampled from a wider population and obtained a higher number of samples, the correlation between the four fixed base pair changes and the east–west division of the subspecies would have broken down and no longer been supported. Secondly, the phenotypic differentiation between the subspecies, such as differences in morphology, plumage and behavioural traits, might have been achieved over relatively recent periods of time, and lineage sorting of the mtDNA might simply not be completed. The star-like radiation of the haplotype network indicates incomplete lineage divergence, so it may be that the western and eastern populations of Yellow-billed Cuckoo became isolated fairly recently and a single mitochondrial gene cannot yet capture this divergence. In a recent monograph on avian subspecies, it is recommended the best approach to assessing subspecies status is to include multiple lines of evidence, genetic and morphological, and a single mtDNA study alone should not suffice (Haig & Winker 2010, Pruett & Winker 2010). The use of next generation sequencing technology is rapidly becoming the optimal tool for identifying molecular markers on a large scale at a reasonable cost to discover genes that control morphological, behavioural and physiological phenotypes, even in non-model organisms with no *a priori* genomic information. With the numerous behavioural and morphological differences observed between the two populations of Yellow-billed Cuckoo, the status of the western subspecies would greatly benefit from the use of next-generation sequencing.

ACKNOWLEDGEMENTS

During this work, LLF was a Masters student in the department of Biology at Lakehead University. Special thanks to Janice M. Hughes for the use of *Coccyzus americanus* (JMH881) & *Coccyzus melacoryphus* (JMH20063) sequences, and valuable editorial input during the thesis version of this manuscript. Thank you to the anonymous reviewers who provided the many helpful comments and revisions that greatly improved the manuscript. Thank you Dr. Carney Matheson and Ryan Lamers for laboratory and statistical support at the Paleo-lab at Lakehead University, Canada. Thank you to all the universities and museums who generously granted tissue loans: Nate Rice (Academy of Natural Sciences of Philadelphia), Sharon Birks (University of Washington Burke Museum of Natural History), Murrelet Halterman (University of Nevada, Reno), Donna Dittmann (Louisiana State University Museum of Natural History), Mark Robbins (University of Kansas Natural History Museum), Terry Yates & Cheryl Parmenter (University of New Mexico), Jocelyn Hudon (Provincial Museum of Alberta), Shannon Hackett and David Willard (Field Museum of Natural History). This work was supported by a Natural Science and Engineering Research Council of Canada grant to JMH and LLF.

REFERENCES

Banks R.C. 1988. Geographic variation in the Yellow-billed Cuckoo. Condor 90: 473–477.

Banks R.C. 1990. Geographic variation in the Yellow-billed Cuckoo: Corrections and comments. Condor 92: 538.

Ballard J.O. & Whitlock C.W. 2004. The incomplete natural history of mitochondria. Mol. Ecol. 13: 729–744.

Clement M., Posada D. & Crandall K.A, 2000. TCS: a computer program to estimate gene genealogies. Mol. Ecol. 9: 1657–1660.

Excoffiner L., Laval G. & Schneider S. 2005. Arlequin ver 3.1: an integrated software package for population genetics data analysis. Evol. Bioinform. Online 1: 47–50.

Franzreb K.E. & Laymon S.A. 1993. A reassessment of the taxonomic status of the Yellow-billed Cuckoo. Western Birds 24: 17–28.

Haig S.M. & Winker K. 2010. Avian subspecies: Summary and prospects. Ornithol. Monogr. 67: 172–175.

Hughes J.M. 1999. Yellow-billed Cuckoo (*Coccyzus americanus*). In: Poole A and Gill F (eds) The Birds of North America, No. 418. The Birds of North America, Inc., Philadelphia, Pennsylvania.

Johnson M.J. 2009. Understanding the habitat needs of the declining western yellow-billed cuckoo: U.S. Geological Survey Fact Sheet 2009–3091.

Laymon S.A. & Halterman M.D. 1987. Can the Western Subspecies of the Yellow-Billed Cuckoo be Saved from Extinction? Western Birds 18: 19–25.

Pruett C.L., Gibson D.D. & Winker K. 2001. Molecular "cuckoo clock" suggests listing of the western Yellow-billed Cuckoos may be warranted. Wilson Bull. 113: 228–231.

Pruett C.L. & Winker K. 2010. Alaska Song Sparrows (*Melospiza melodia*) demonstrate that genetic marker and method of analysis matter in subspecies assessments. Ornithol. Monogr. 67: 162–171.

Rambaut A. 2002. Sequence Alignment Editor v2.0a11. University of Oxford, United Kingdom.

Ridgway R. 1887. A Manual of North American Birds. Lippincott Publishers, Philadelphia, Pennsylvania.

Sheldon F.H., Whittingham L.A., Moyle R.G., Slikas B. & Winkler D.W. 2005. Phylogeny of swallows (Aves: Hirundinidae) estimated from nuclear and mitochondrial DNA sequences. Mol. Phylogenet. Evol. 35: 254–270.

Sorenson M.D., Ast J.C., Dimcheff D.E., Yuri T. & Mindell D.P. 1999. Primers for a PCR-based approach to mitochondrial genome sequencing in birds and other vertebrates. Mol. Phylogenet. Evol. 12: 105–114.

SAMENVATTING

De westelijke ondersoort van de Geelsnavelkoekoek *Coccyzus americanus occidentalis* is in de laatste decennia sterk in aantal afgenomen. De ondersoort is opgenomen in de wet voor bedreigde soorten in de Verenigde Staten, maar zijn bescherming is nog niet geïmplementeerd. Een van de redenen voor het gebrek aan bescherming is een controverse over het feit of er genetisch verschil bestaat tussen de westelijke ondersoort en de ondersoort *americanus* die in het oosten van de Verenigde Staten voorkomt. In een eerder onderzoek was namelijk slechts een klein verschil in het cytochroom *b* uit het mitochondriaal DNA gevonden. Bovendien was die analyse op een beperkt aantal monsters gebaseerd. De auteur bemonsterde in een vervolgonderzoek aanvullend materiaal uit zowel het westelijke als het oostelijke deel van het verspreidingsgebied. Zij analyseerde 949 basisparen in het cytochroom *b*. In tegenstelling tot het eerdere onderzoek vond de auteur slechts een zwakke, statistisch niet significante genetische differentiatie. Het kan zijn dat genetische verschillen tussen de oostelijke en westelijke populaties niet kunnen worden aangetoond omdat de afscheiding nog zo recent is dat de evolutionaire lijnen nog niet gescheiden zijn in het cytochroom *b*. Een alternatieve verklaring zou kunnen zijn dat er vrij veel genetische uitwisseling is tussen de populaties. Een onverwachte ontdekking was het voorkomen van vrij veel genetische variatie binnen de westelijke ondersoort, wat kan duiden op differentiatieprocessen binnen deze populatie. De auteur stelt voor dat bij vervolgonderzoek aan de taxonomische status van de Geelsnavelkoekoek gebruik wordt gemaakt van nieuwe genetische methoden waarbij een veel groter deel van het genoom kan worden bekeken. (YIV)

Corresponding editor: Yvonne I. Verkuil
Received 26 April 2013; accepted 25 October 2013

Appendix 1. Other sequences used in this study. Pruett *et al*. (2001) samples included in analysis indicated by an asterisk (*). All *Coccyzus melacoryphus* were used for alignment of cytochrome *b* purposes only. Additional *C. americanus* (JMH881) was included in analysis in the eastern nominate population. Specimen numbers refer to source collections: AF/AY, EMBL; ANSP, Academy of Natural Sciences, Philadelphia; CNAV, Coleccion Nacional de Aves, Mexico; JMH, Janice Hughes, Lakehead University; NK, University of New Mexico, Albuquerque; ROM, Royal Ontario Museum; UAM, University of Alaska Museum, Fairbanks.

Taxon	EMBL accession	Museum accession	Sample origin City/State & Country	Source
Coccyzus americanus	AY046906*	NK43108	Albuquerque, New Mexico, USA	EMBL
Coccyzus americanus	AY046907*	NK35431	Albuquerque, New Mexico, USA	EMBL
Coccyzus americanus	AY046905*	NK3993	Albuquerque, New Mexico, USA	EMBL
Coccyzus americanus	AF249271*	UAM 10131	Minnesota, USA	EMBL
Coccyzus americanus	AF249270*	UAM 10130	Minnesota, USA	EMBL
Coccyzus americanus	AY046910*	UAM 13376	Rutland, Vermont, USA	EMBL
Coccyzus americanus	AY046909*	CNAV 23931	Veracruz, Mexico	EMBL
Coccyzus americanus	AY046908*	UAM 10354	Veracruz, Mexico	EMBL
Coccyzus americanus	AF249269*	UAM 7059	Ketchikan, Alaska, USA	EMBL
Coccyzus americanus	AF249268*	UAM 6953	Juneau, Alaska, USA	EMBL
Coccyzus melacoryphus	-	JMH20036	Paraguay, South America	JMH
Coccyzus melacoryphus	-	JMH5958	Ecuador, South America	JMH
Coccyzus melacoryphus	-	ANSP 4661	Ecuador, South America	JMH
Coccyzus americanus	-	JMH881	Maryland, USA	ROM