

# Migratory double breeding in Neotropical migrant birds

Sievert Rohwer<sup>a,1</sup>, Keith A. Hobson<sup>b</sup>, and Vanya G. Rohwer<sup>a,2</sup>

<sup>a</sup>Burke Museum and Department of Biology, University of Washington, Seattle, WA 98195; and <sup>b</sup>Environment Canada, Saskatoon, SK, Canada S7N 0H3

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Neotropical migratory songbirds typically breed in temperate regions and then travel long distances to spend the majority of the annual cycle in tropical wintering areas. Using stable-isotope methodology, we provide quantitative evidence of dual breeding ranges for 5 species of Neotropical migrants. Each is well known to have a Neotropical winter range and a breeding range in the United States and Canada. However, after their first bout of breeding in the north, many individuals migrate hundreds to thousands of kilometers south in midsummer to breed a second time during the same summer in coastal west Mexico or Baja California Sur. They then migrate further south to their final wintering areas in the Neotropics. Our discovery of dual breeding ranges in Neotropical migrants reveals a hitherto unrealized flexibility in life-history strategies for these species and underscores that demographic models and conservation plans must consider dual breeding for these migrants.

dual breeding | isotopes | itinerant breeding | west Mexico

Itinerant breeding, wherein the same individuals breed in different regions in a single season, is established or suspected for just a few species of New and Old World birds (1). Some itinerant breeders exploit unpredictable food supplies, so their breeding areas may not be in the same place from year to year, as in the red-billed quelea (*Quelea quelea*) (2, 3). However, 2 other species, European quail (*Coturnix coturnix*) and dotterel (*Charadrius morinellus*), apparently breed first in southern regions, then move north with the progression of spring to breed again (ref. 4 and Whitfield in ref. 5). We suggest that the term *migratory double breeding* might appropriately distinguish species with dual breeding ranges that are consistent from year to year. Unlike more nomadic itinerant breeders, migratory double breeders move between early and later breeding sites that are used reliably from year to year.

In July and August 2005–2007, we discovered many individuals of 5 Neotropical migrant birds breeding in coastal Sinaloa and Baja California Sur, Mexico. Coastal west Mexico receives most of its annual precipitation during a July–September monsoon (6, 7). These late summer rains stimulate the tropical deciduous forests of this region to leaf out and flower, generating an abundance of insect prey, as well as seeds from grasses and other plants. This seasonal pulse of resources is well known to attract many western Neotropical migrant birds to this region for their postbreeding molt (8).

At least 5 Neotropical migrants [yellow-billed cuckoo (*Coccyzus americanus*), Cassin's vireo (*Vireo cassinii*), yellow-breasted chat (*Icteria virens*), hooded oriole (*Icterus cucullatus*), and orchard oriole (*Icterus spurius*)] apparently move to west Mexico, not primarily to molt but to breed a second time, after having bred earlier in the north. Several field observations support this claim. First, females of these 5 species that were initiating breeding had regressed and featherless brood patches, demonstrating that they had bred earlier that same summer. Second, during hundreds of hours of netting and observation in July, we found no recently fledged juveniles of these species, suggesting that they had not bred in west Mexico earlier in the same summer. Third, many individuals of the 5 putative double breeders sampled for this study were indisputably breeding when collected in west Mexico: males had fully enlarged testes, and females were laying or incubating [sup-

porting information (SI) Table S1]. Fourth, we found many active nests for orchard orioles and hooded orioles, and males of all 5 species were singing and defending territories or guarding females. Together, these observations suggested to us that the individuals we found breeding in west Mexico had attempted to breed in the United States or Canada and then migrated to breed again in the lowland thorn forests of west Mexico.

We present 3 analyses of stable-isotope data supporting our reasoning. Although our analyses of stable isotopes constitute circumstantial evidence of dual breeding, we emphasize that such quantitative evidence is considerably stronger and more revealing than evidence based solely on physical evidence of earlier breeding in females or mirrored times of departure and arrival in different geographic regions. Unlike population comparisons, data from isotopes give evidence that specific individuals have moved to breed again in a different region, making data on the breeding condition of these individuals especially valuable. Only for the Old World red-billed quelea (2) do we have proof of dual breeding ranges based on numerous marked individuals.

To test whether these species bred earlier in the United States or Canada, we compared their stable-isotope profiles with those of a reference sample of molt migrants (31 individuals of 6 species) and of a reference sample of local resident breeders (56 individuals of 19 species) (Table S1). Molt migrants breed largely in the United States and Canada and migrate to the southwestern United States or to northwestern Mexico to molt. As expected, no molt migrant was in breeding condition when collected in west Mexico (Table S1). Resident breeders are well known to breed in west Mexico, and only species with breeding ranges that do not or scarcely extend into the United States were used as reference samples in this study; many of these resident breeders were in breeding condition when collected (Table S1).

We hypothesized that resident breeders would differ in their stable H ( $\delta D$ ), C ( $\delta^{13}C$ ), and N ( $\delta^{15}N$ ) isotope composition compared with molt migrants or migratory double breeders that had recently arrived from the United States or Canada, thereby providing us with an isotopic tag of origin (SI Text). We tested for migratory double breeding in 3 ways: (i) by discriminant function analysis of isotope data to separate birds by origin, (ii) by regressions of  $\delta D$  on day of sampling to identify north–south patterns of movement, and (iii) by correlations between  $\delta D$  values in muscle and reproductive tissue to identify those individuals that had initiated breeding in Mexico after arriving there from further north.

## Results

**Discriminant Analyses.** Because molt migrants do not breed in coastal Sinaloa or Baja California Sur (8, 9), we know that recently

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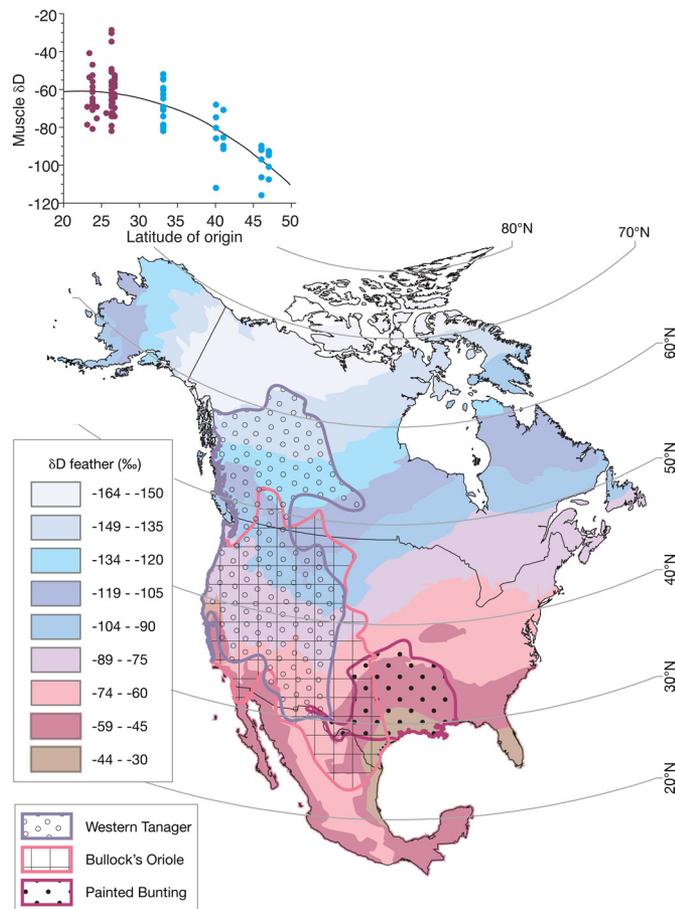
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<sup>1</sup>To whom correspondence should be addressed. E-mail: rohwer@u.washington.edu.

<sup>2</sup>Present address: Department of Biology, Queen's University, Kingston, ON, Canada K7L 3N6.

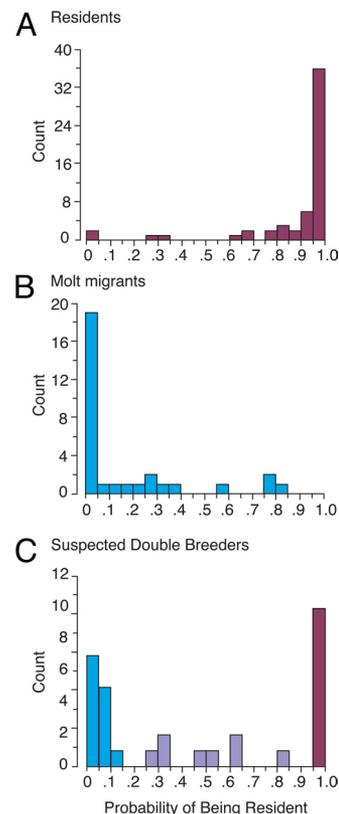
This article contains supporting information online at [www.pnas.org/cgi/content/full/0908121106/DCSupplemental](http://www.pnas.org/cgi/content/full/0908121106/DCSupplemental).



**Fig. 1.** Feather  $\delta D$  clines for North America based on the regression between feather  $\delta D$  in ref. 35 and the growing-season mean precipitation  $\delta D$  of ref. 36. *Inset:* Relationship between muscle  $\delta D$  and latitude of origin for the birds in the 2 reference samples used for the discriminant analysis: west Mexican resident breeders (maroon) and molt migrants from the United States and Canada (blue). We show the ranges of 3 exemplar molt migrants on the continental map for  $\delta D$  to illustrate how the latitude at which molt migrants breed affects the  $\delta D$  values they will show when collected in west Mexico. The southern-most range is for painted buntings from the midwest, which are molt migrants; painted buntings that breed in the eastern United States are not shown because they molt on their breeding range.

arrived molt migrants (individuals not molting or in early stages of the molt) should differ from resident breeders in  $\delta D$  values, owing to well-established continental patterns in foodwebs (Fig. 1), and in  $\delta^{15}N$  and  $\delta^{13}C$  values, owing to broad influences of climate on plants, including photosynthetic pathways (e.g., ref. 10). For our first analysis, we used a discriminant function to distinguish resident breeders and molt migrants according to 3 isotopes measured in both muscle and claw. True migratory double breeders (i.e., those that had bred previously in the same year in the United States or Canada) should classify with the reference sample of molt migrants, whereas potential double breeders that breed in west Mexico without having moved there from the north should classify with known Mexican residents. As predicted, discriminant analysis readily separated most individuals of our reference samples (molt migrants and resident breeders), with only 4 individuals in each reference group misclassified (Fig. 2 *A* and *B*). Most of this discrimination was due to the strong north–south geographic clines in precipitation  $\delta D$  in North America, but differences in  $\delta^{15}N$  between residents and molt migrants also contributed importantly to discriminating the reference samples (*SI Text*).

Group assignment probabilities for 31 individuals of 3 species



**Fig. 2.** Assignment probabilities from the discriminant analysis. (*A*) Probability of classifying as resident for the reference sample of resident breeders ( $n = 57$ ). (*B*) Probability of classifying as resident for the reference sample of molt migrants ( $n = 37$ ). (*C*) Assignment probabilities for 31 suspected double breeders (yellow-billed cuckoo,  $n = 10$ ; Cassin's vireo,  $n = 8$ ; hooded oriole,  $n = 13$ ).

suspected of double breeding (yellow-billed cuckoo, Cassin's vireo, and hooded oriole) are shown in Fig. 2*C*. Nine of the 13 hooded orioles, 3 of the 8 Cassin's vireos, and 1 of the 10 yellow-billed cuckoos were assigned to the molt migrant reference group with probabilities of .85 or higher, and 2 more Cassin's vireos and 1 more hooded oriole classified with the molt migrants with probabilities between .65 and .75 (Fig. 2*C*). The reproductive condition or behavior of these 13 individuals suggests that most could have moved to western Mexico for a second round of breeding after an earlier breeding attempt in the United States or Canada (Table 1). The single yellow-billed cuckoo was a female collected August 19, 2006 that had just laid a 3-egg clutch [3 collapsed follicles (11)]. Two of the 3 Cassin's vireos were singing persistently from territories when collected, and at least 1 had enlarged testes (testes destroyed when shot for the second); the third was a postbreeding male that had started to molt, and this species is not a molt migrant (12). Five of the 9 hooded orioles were males with breeding testes that had not initiated molt; 2 were incubating females from late August; and 2 were males with regressing testes that had initiated molt (Table 1). Although hooded orioles are thought to be molt migrants (8), the 2 molting males had likely moved to Mexico to breed because their testes were still moderately enlarged (Table 1). All of these 13 individuals were collected at the end of July or in August, giving them plenty of time to have bred earlier north of Mexico.

In contrast, 8 yellow-billed cuckoos, 1 Cassin's vireo, and 2 hooded orioles classified with resident breeders with probabilities of .87 or higher (Fig. 2*C*), suggesting either that they had bred earlier in Mexico or that they carried an isotopic signature from an earlier breeding in the United States that could not be distinguished from the signature of our reference sample of resident breeders. Our

**Table 1. Breeding details for the 23 migratory double breeders identified by the discriminant analysis and by the 2 patterns of tissue contrasts**

Species	Number	Date	Breeding data (measurements in mm)
Discriminant analysis			
YBCU	82371	Aug. 19	Female starting incubation (3 collapsed follicles), no molt
CAVI	81189	Aug. 4	Singing, territorial male, no molt; gonads shot
CAVI	82683	Aug. 28	Singing, territorial male, testis 6.5 × 4.5, no molt
CAVI	82690	Aug. 30	Singing, postbreeding male, molting, testis 2.5 × 1.5
HOOR	81191	Aug. 6	Postbreeding male, starting molt, testis 4 × 4
HOOR	81253	July 29	Postbreeding male, starting molt, testis 7 × 4.5
HOOR	82353	Aug. 30	Incubating female, no molt (edematous brood patch)
HOOR	82373	Aug. 20	Incubating female, no molt (edematous brood patch)
HOOR	82386	Aug. 28	Breeding male, starting primary molt, testis 13 × 7
HOOR	82461	Aug. 6	Breeding male, no molt, testis 11 × 7
HOOR	82462	Aug. 6	Breeding male, no molt, testis 8.5 × 6
HOOR	82507	Aug. 6	Breeding male, no molt, testis 10 × 7
HOOR	82623	Aug. 6	Breeding male, no molt, testis 7 × 5
Depleted $\delta$ D muscle compared with reproductive tissue			
YBCU	84072	July 17	Breeding male, calling when shot, testis 10 × 6, no molt
YBCU	84088	July 24	Breeding male, calling when shot, testis 11 × 5.5, no molt
OROR	84046	July 7	Breeding male, testis 12 × 9, seminal vesicles 6 × 5, no molt
OROR	83975	July 7	Breeding male, testis 11.5 × 8, seminal vesicles 5 × 4, no molt
OROR	84049	July 7	Breeding male, testis 10 × 7.5, seminal vesicles 5 × 4.5, no molt
OROR	84076	July 19	Laying female, no molt
Enriched $\delta$ D muscle compared with reproductive tissue			
YBCU	83966	July 24	Laying female, no molt
YBCH	83981	July 13	Female about to lay, oviduct enlarging, ovum 6, old brood patch, no molt
YBCH	84020	July 13	Breeding male, testis 9 × 6, no molt
OROR	83973	July 7	Laying female, no molt

YBCU, yellow-billed cuckoo; CAVI, Cassin's vireo; HOOR, hooded oriole; OROR, orchard oriole; YBCH, yellow-breasted chat.

discriminant analysis failed to classify any yellow-breasted chat or orchard oriole with the molt migrant reference group. In retrospect, this makes sense because deuterium ( $\delta$ D) contributed strongly to distinguishing molt migrants and resident breeders (*SI Text*), yet the deuterium isoscape for west Mexico matches that found in southwestern and south central regions of the United States, where these 2 species regularly breed (Fig. 1).

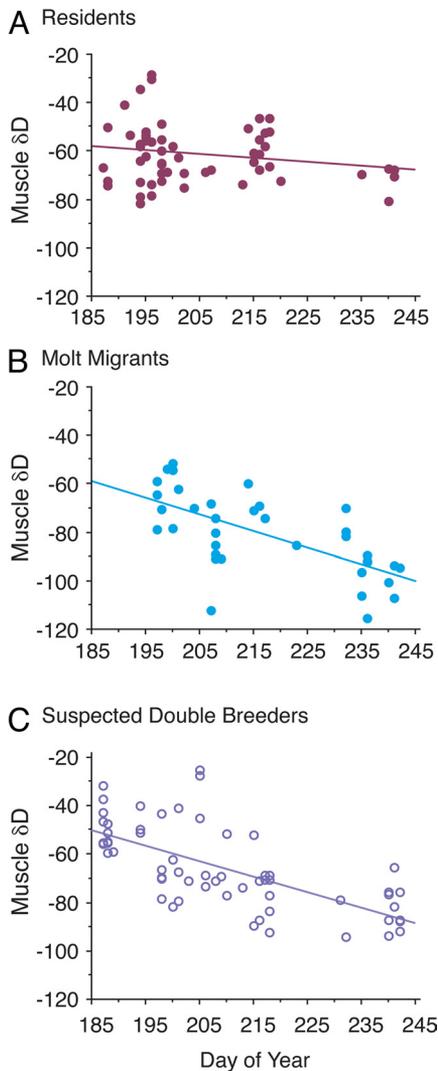
**Deuterium and Date of Sampling.** Our second analysis sought evidence of double breeding by contrasting the predicted relationships between muscle  $\delta$ D on date of sampling for resident breeders with that for molt migrants and suspected double breeders. We attempted to sample molt migrants and suspected double breeders shortly after they had arrived in west Mexico. If we succeeded in sampling these individuals soon after their arrival in west Mexico, the  $\delta$ D values for both groups should be negatively associated with date of collection; this follows because individuals that bred earlier in southern regions of the United States (areas more enriched with deuterium) should arrive earlier in west Mexico, whereas individuals that bred later in more northern regions of the United States or Canada (areas more depleted in deuterium) should arrive later in west Mexico. In contrast, resident breeders from west Mexico collected during the same time period should show no correlation between  $\delta$ D values and day of collection because we did not expect the local isoscape to change during our July–August sampling period.

As predicted, molt migrants and suspected double breeders showed strong negative regressions of muscle  $\delta$ D on day of collection, whereas Mexican residents showed no relationship (Fig. 3 A–C). Furthermore, each of our suspected double breeders with samples of 10 or more individuals from west Mexico (yellow-billed cuckoo, orchard oriole, hooded oriole, and yellow-breasted chat) showed significant negative regressions of muscle  $\delta$ D on day of collection (Fig. S1 A–D). These negative regressions further support migratory double breeding for hooded oriole and yellow-billed cuckoo and suggest double breeding for orchard oriole and yellow-breasted chat.

**Comparing  $\delta$ D in Muscle and Reproductive Tissue.** Our third analysis sought to identify double breeders by regressing muscle  $\delta$ D on  $\delta$ D for reproductive tissue (enlarged testes or oviducts; Fig. S2), which we began sampling only in 2007 (Table S1). The logic behind this approach is that migratory birds regress reproductive tissues but augment breast muscles before they migrate (13, 14). Thus, if a bird bred in the north and then migrated to west Mexico to breed again, it should have regressed its reproductive organs and enlarged its breast muscles in the United States or Canada, before it migrated to Mexico (see *SI Text* for exceptions that would not follow this pattern). Once in Mexico this bird would then rebuild its reproductive organs for the second breeding attempt. Thus, there should be no correlation between the  $\delta$ D signatures in reproductive and muscle tissue for our presumed migratory double breeders collected in west Mexico: their reproductive tissue should carry a local  $\delta$ D signature, but their breast muscle should carry a  $\delta$ D signature from their northern breeding site. In contrast, the  $\delta$ D signatures from these 2 tissues should be highly correlated in resident breeders because both their reproductive and muscle tissues were built in west Mexico. Comparing  $\delta$ D values in different tissues from the same individuals has the additional advantage of controlling for much of the interindividual variation in  $\delta$ D values (well illustrated in Figs. 1 and 3 and Fig. S2) that is caused by diet and other factors.

As predicted, the correlation between  $\delta$ D in reproductive and muscle tissue was near zero for suspected double breeders ( $n = 16$ ,  $r^2 = 0.03$ ; Fig. 4B), suggesting that most of these individuals had built their muscle and reproductive tissue in different places and were true migratory double breeders. In contrast, this same correlation was high for west Mexican residents ( $n = 16$ ,  $r^2 = 0.86$ ; Fig. 4A), presumably because their muscle and reproductive tissues were built in the same location. The strength of this correlation for residents, together with the lack of relationship for suspected double breeders, supports the utility of this approach to better defining movement histories.

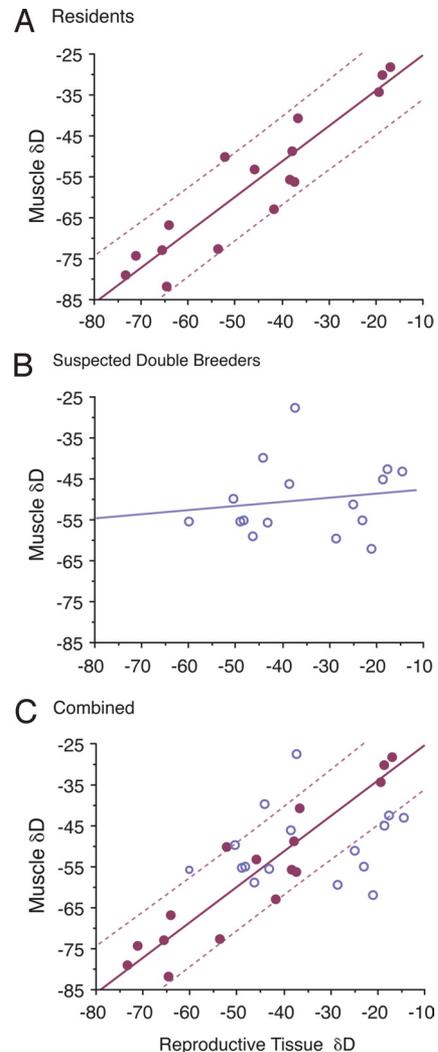
We can now combine these regressions for residents (Fig. 4A) and suspected double breeders (Fig. 4B) to identify individuals that are especially likely to be double breeders. To do this we used the



**Fig. 3.** Regression of muscle  $\delta D$  on day collected in west Mexico for (A) residents ( $n = 56$ ;  $P = 0.15$ ;  $r^2 = 0.04$ ), (B) molt migrants ( $n = 35$ ;  $P < 0.0001$ ;  $r^2 = 0.44$ ), and (C) suspected double breeders ( $n = 60$ ;  $P < 0.0001$ ;  $r^2 = 0.46$ ). Day 185 is July 4. The lack of relationship for residents, together with the strong and similarly negative regressions for molt migrants and for suspected double breeders, suggest that latitude of origin determines  $\delta D$  values in the latter 2 groups. Because we know molt migrants bred earlier in the north, we can infer that the suspected double breeders did the same because their regression statistics (C) match those for molt migrants (B) and not those for resident breeders from west Mexico (A).

regressions for resident breeders as our expected relationship for birds that are not migratory double breeders and set confidence intervals for individuals of 85% around this regression line (matching the assignment cutoffs used in the discriminant analysis; Fig. 4A). We then superimposed the plot for suspected double breeders (Fig. 4B) on the plot for residents and recognized individuals falling outside the 85% confidence intervals around this line as likely migratory double breeders (Fig. 4C). Whether suspected double breeders fall above or below the confidence intervals around this line suggests their region of origin in the United States or Canada.

Six suspected double breeders (4 orchard orioles and 2 yellow-billed cuckoos) fell below the 85% confidence intervals for the resident regression (Fig. 4C). Their muscle  $\delta D$  was depleted relative to the expected value predicted from their reproductive tissue, suggesting that their muscle  $\delta D$  matched those expected from the northern United States or Canada. Four suspected double breeders (1 orchard oriole, 1 yellow-billed cuckoo, and 2 yellow-breasted



**Fig. 4.** Regression of muscle  $\delta D$  on  $\delta D$  for reproductive tissue for (A) residents and (B) suspected double breeders. The close association in residents ( $n = 16$ ;  $r^2 = 0.86$ ;  $P < 0.0001$ ) follows because muscle and reproductive tissues were both generated locally. The lack of association in suspected double breeders ( $n = 16$ ;  $r^2 = 0.03$ ;  $P = 0.55$ ) is expected if their reproductive tissue was generated in west Mexico but their muscle tissue was generated during an earlier breeding attempt in the United States or Canada. (C) Combination of graphs in A and B, identifying migratory double breeders that fall outside of the 85% confidence limits for individuals in the resident regression (A).

chats) carried muscle  $\delta D$  that was enriched relative to that expected from their reproductive  $\delta D$  (Fig. 4C), suggesting that these individuals had come from the southern United States. Remarkably, the discriminant analysis classified every one of these 10 suspected double breeders with Mexican residents at probabilities of 0.96 or higher. Only with information from newly recrudescenced reproductive tissue were we able to determine that these birds were not residents.

The gonadal data for these 10 specimens leave no doubt that all were breeding when collected in west Mexico (Table 1). Six specimens had  $\delta D$  depleted in their breast muscle compared with their reproductive tissue: 2 were yellow-billed cuckoos that were calling when shot and had fully enlarged testes; 3 were male orchard orioles with fully enlarged testes and seminal vesicles; the sixth was a female orchard oriole that was laying. Four other specimens had  $\delta D$  enriched in their breast muscle compared with their reproductive tissue: 1 was a laying female yellow-billed cuckoo, collected on July 24; 1 was a female yellow-breasted chat that was about to lay

and that had bred earlier that season (indicated by her old brood patch); 1 was a male chat with fully enlarged testes; the last was a laying female orchard oriole. All of these 10 specimens were collected in July, which is late enough for them either to have bred earlier that season north of Mexico.

A third group of 6 suspected double breeders fell within the 85% confidence limits of the regression of muscle  $\delta D$  on reproductive tissue  $\delta D$  for residents (Fig. 4C), making it impossible for this analysis to classify them as migratory double breeders. Note, however, that even these 6 individuals could have bred earlier in the United States in regions where the isoscape for muscle  $\delta D$  matches that for west Mexico. The fact that the regression of muscle  $\delta D$  on day of collection was as negative for our sample of suspected double breeders as it was for our sample of molt migrants (Fig. 3B and C) suggests that the similarity in west Mexican and midwestern isoscapes is why these individuals fell near the line for west Mexican resident breeders. All were in breeding condition (Table S1).

## Discussion

Our discovery of distinct breeding areas separated by hundreds to thousands of kilometers heralds a new chapter in the use of stable isotopes in documenting migratory connectivity (15, 16) and has important conservation and research implications. Although dual breeding in nocturnal migrants has been inferred for the northward migration in species that winter in Africa and breed in Eurasia (ref. 4 and Whitfield in ref. 5), the double breeding we document here is for the Neotropical–Nearctic migration system and occurs on the southward migration. These isotopic data defy a long-held assumption that migrants breed, with almost no exception, in only one place between just 2 bouts of migration. Instead migratory double breeders have added an additional migration and breeding season to their annual cycle, with attendant periods of physiologic transition. This discovery challenges the notion that physiologic mechanisms prevent migrants from incorporating additional life-history stages in their annual cycle, either because of constraints on the time required to move from one physiologic state to another (e.g., migration to breeding), or because of incompatibilities in the endocrine control mechanisms supporting different life-history stages (17, 18). Instead, migratory double breeding shows that entirely new life-history stages can be added to the annual cycle and challenges the notion that physiologic transition times are as important in constraining life history diversification as previously hypothesized.

Because the half-life of deuterium in muscle is just 8 days or so for the range of body masses we examined, we collected our molt migrant reference sample and our potential double breeders shortly after their arrival in west Mexico to minimize the risk of missing a northern signature in both groups. To this end, we excluded molt migrants that were advanced in flight feather molt, and we limited most of our sampling of potential double breeders to July (Table S1), fearing that August specimens might represent birds that had moved to breed in west Mexico in July and that could have been there long enough to have lost their northern signature (half-life hypothesis). Alternatively, however, migrants that breed reasonably far north in the United States or Canada (where  $\delta D$  values are more negative than in west Mexico) breed later than southern conspecifics; thus the opportunity for northern migrants to move to west Mexico to breed again comes later in the summer (opportunity hypothesis). The strong negative regression between day of collection and deuterium levels in muscle suggests that the opportunity hypothesis applies more strongly to our data (Fig. 3 and Fig. S1). There should be no relationship between day of collection and muscle deuterium for residents, and none was found, but the negative regressions for potential double breeders suggest that they first bred further north in North America (Fig. 3 and Fig. S1). These negative relationships suggest that we erred in not collecting more potential double breeders in August, when nests were still being initiated. At least some of these later breeders had likely just arrived

in west Mexico from more northern areas and could easily have been distinguished from west Mexican resident breeders by the discriminant analysis.

Contrasts in the development of reproductive tissues between our reference sample of molt migrants and our suspected double breeders help exclude the possibility that double breeders were molt migrants that had not yet started their molt. Most of the suspected double breeders included in our isotope analyses were in full breeding condition with enlarged testes or oviducts (Table S1). Additionally, many of the males were singing and defending territories or guarding females, several of the females were laying or incubating, and we found many active nests of orchard and hooded orioles. Finally, orchard orioles and yellow-billed cuckoos molt on their wintering grounds, well to the south of the region where they breed again in northwest Mexico, whereas yellow-breasted chats and Cassin's vireos are both thought to molt on their breeding grounds; only hooded orioles are thought to be molt migrants (8), and we found them to be molting in west Mexico after breeding there in late summer. In contrast, our reference sample of molt migrants had regressed reproductive structures, and we observed no evidence of breeding in these species during 3 years of fieldwork (Table S1). Many additional females of our 5 species of suspected double breeders had dry brood patches, suggesting that they had bred earlier that same season, presumably in the north.

Two of our tests suggest that many the individuals of the 5 potential double breeders we examined had bred earlier in the north. However, these numbers tell us nothing about the percentage of individuals in northern breeding populations that attempt a second round of breeding in Mexico. Hooded orioles are extraordinarily common as late summer breeders in Baja California Sur, and orchard orioles, yellow-breasted chats, and yellow-billed cuckoos breed abundantly in the coastal lowlands of extreme southern Sonora and much of coastal Sinaloa. Thus, migratory double breeding may be common in at least some North American breeding populations of these species.

Molt migrants, as well as migratory double breeders, depend on habitat preservation in coastal Sinaloa and southern Sonora. The recent conversion of vast regions of coastal thorn forest to industrial agriculture in this region could be responsible for notable population declines in some species of both molt migrants and double breeders. Painted buntings (*Passerina ciris*) from the Midwestern breeding population move to west Mexico to molt (19) and have declined rapidly since the 1970s (20). Similarly, the yellow-billed cuckoo, which seems to be a migratory double breeder, is declining in eastern North America (20) and has disappeared from most of its breeding range in the western United States and Canada, even though substantial areas of riparian breeding habitat still exist in some areas where historically it was a common breeder (21, 22). Yellow-billed cuckoos are abundant late-summer breeders in the deciduous thorn scrub of coastal west Mexico that has not been converted to agriculture (23). If, as we propose, yellow-billed cuckoos are migratory double breeders, the population viability for cuckoos breeding in the United States and Canada may require that some fraction of those birds breed again in west Mexico. This second breeding may have been especially critical to the viability of western yellow-billed cuckoo populations because dry conditions west of the Rocky Mountains arrest primary productivity in mid-summer, precisely when food becomes abundant in west Mexico.

Migratory double breeding poses many interesting questions about plasticity in avian life histories. How will spacing and mating systems, clutch sizes, and patterns of parental care and sexual conflict vary between early breeding sessions in the United States and Canada and later sessions in west Mexico? How do offspring from the same parents that are produced in regions separated by thousands of kilometers of longitude orient appropriately for migration? And how do adults that breed after a bout of fall migration to west Mexico schedule their molt appropriately? Although the direction and distance of intercontinental migrations

are known to be inherited in some passerines that migrate long distances at night (24–26), the direction of migration and its scheduling relative to the annual molt must be flexible in migratory double breeders. Orchard orioles and eastern yellow-billed cuckoos are unusual among Neotropical migrants that breed in eastern North America for molting not on their breeding grounds but on their winter range (8). If, as we suggest, some fraction of these eastern-breeding populations breeds a second time in west Mexico before going on to winter in Central and South America, then some adults molt after a single bout of breeding and migration, whereas others molt after 2 bouts of breeding and migration. Finally, as nocturnal migrants, how do the offspring of cuckoos and orioles hatched in eastern North America orient southwest in their migration, whereas offspring hatched from the same parents in west Mexico orient southeast in their migration toward a presumably common winter range?

## Materials and Methods

The value of using naturally occurring stable isotope ratios to trace the origin of migratory wildlife is well established (27). The underlying principle is that tissues of organisms come into isotopic equilibrium with local foodwebs and that these foodwebs vary geographically in their isotopic signatures. These differences in isotopic patterns, or *isoscapes*, derive from biogeochemical processes related to climate, topography, and weather patterns, from differences in photosynthetic pathways, and from variation in geologic substrates (28). For organisms that move between isoscapes, information on previous origins can be inferred, provided the elemental turnover rate in tissue retains enough of the original signature of origin to be traced. Elemental turnover rates for C and N in avian muscle, liver, and blood have been determined (29–31) and in general follow allometric relationships related to body mass (32). For a bird of 40 g, typical of the species examined here, the half-life of C and N in muscle tissue is on the order of 10 days. Source isotope signatures are typically considered to be traceable within 2 half-lives or 20 days. For metabolically inactive tissues like feathers and claws, source isotope signatures are essentially locked in and represent the location and food where they were grown.

Less information is available for the turnover rates of H in avian tissues, but we anticipate that in general those values established for C and N apply. Use of this isotope is further complicated by the fact that H bonded to N and O in tissues is capable of exchange with ambient body water. For example, controlled laboratory studies on captive birds have established that approximately 20% of H is exchangeable with drinking water (33). For metabolically inactive tissues like feathers and claws, our H isotope measurements correspond to the nonexchangeable fraction of H (34). Stable H isotope values in muscle tissue may take up a local signal faster than for C and N, owing to exchange with drinking water, but because the bulk (i.e., 80%) of that tissue does not exchange water, a half-life of 8 days is not unreasonable. To our knowledge, no elemental turnover information is available for reproductive tissues, but unless solely stored reserves are used to generate these tissues, they are almost certainly formed from elements derived from the breeding grounds and so can be considered local.

Our samples in Sinaloa and Baja California Sur were collected in July and August of 2005–2007 (Table S1). Molt migrants were collected from July 16 (the earliest date we encountered them) to August 30. To ensure that the molt migrants included in our analyses had recently arrived in west Mexico and would give us northern isotopic signatures, we used individuals that either had not initiated molt or were in early stages of the primary molt. Residents were collected between July 6 and August 29. Potential double breeders were collected from July 6 to August 30; only 12 of the 57 potential double breeders were collected in the latter part of August. If these birds had moved to west Mexico to breed some weeks earlier, they may have been sampled late enough to have acquired a local isotopic signature, biasing results against the discovery of migratory double breeding.

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