

Non-breeding season events influence sexual selection in a long-distance migratory bird

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The study of sexual selection has traditionally focused on events and behaviours immediately surrounding copulation. In this study, we examine whether carry-over effects from the non-breeding season can influence the process of sexual selection in a long-distance migratory bird, the American redstart (*Setophaga ruticilla*). Previous work on American redstarts demonstrated that overwintering in a high-quality habitat influences spring departure dates from the wintering grounds, advances arrival dates on the breeding grounds and increases apparent reproductive success. We show that the mixed-mating strategy of American redstarts compounds the benefits of overwintering in high-quality winter habitats. Males arriving to breed in Canada from high-quality winter habitats arrive earlier than males from poor-quality habitats, resulting in a lower probability of paternity loss, a higher probability of achieving polygyny and ultimately higher realized reproductive success. Such results suggest that the process of sexual selection may be influenced by events interacting throughout the annual cycle.

Keywords: non-breeding; sexual selection; American redstart; carry-over; extra-pair paternity; polygyny

1. INTRODUCTION

For long-distance migratory birds, individual life history, ecology and behaviour are shaped by events and selective pressures acting throughout the annual cycle (Greenberg & Marra 2005). The challenge in understanding seasonal interactions lies in the inherent difficulty of tracking individuals and tracing the impacts of carry-over effects (i.e. events occurring during one season that *carry over* to influence an individual or population's performance in subsequent seasons; Marra *et al.* 1998) between the phases of the annual cycle, often between continents. Until recently, making connections between breeding and wintering populations and detecting potential carry-over effects has remained elusive. However, advances in the utilization of naturally occurring biochemical markers, such as stable isotopes, in animal tissues have allowed us to begin making connections throughout the annual cycle (Webster *et al.* 2002; Rubenstein & Hobson 2004). Studies using stable-hydrogen isotopes have revealed connections between wintering and breeding populations (Webster *et al.* 2002), use of migratory stopover sites (e.g. Yohannes *et al.* 2007) and patterns of migration (e.g. Kelly *et al.* 2002). This technique has also revealed previously unknown carry-over effects. Studds *et al.* (2008) recently demonstrated that natal dispersal is influenced by conditions during the non-breeding season, whereby individuals overwintering in high-quality habitats disperse south of their natal origin, while individuals in low-quality winter habitats migrate much further and ultimately breed north of their natal origin. Indeed, carry-over effects from the non-breeding

period have now been shown to impact many aspects of individual life history in a variety of species.

In European barn swallows (*Hirundo rustica*), favourable winter conditions in Africa (inferred by the normalized difference vegetation index, NDVI) advance population-level arrival onto the breeding grounds in Italy (Saino *et al.* 2004a). Furthermore, NDVI in winter is positively correlated with population-level breeding success and the length of tail streamers, a sexually selected trait that is moulted on the wintering grounds (Saino *et al.* 2004b). At the individual level, territory acquisition in high-quality winter habitats can have significant fitness consequences. American redstarts (*Setophaga ruticilla*) holding winter territories in high-quality habitats have higher annual return rates (Marra & Holmes 2001), are in better condition (Marra & Holberton 1998; Studds & Marra 2005) and ultimately depart the wintering grounds earlier than individuals holding territories in low-quality habitat (Marra *et al.* 1998; Studds & Marra 2005). Moreover, these consequences can carry over to subsequent seasons. Stable-carbon isotope analysis has revealed that the quality of an individual's winter habitat can influence condition during migration (black-throated blue warblers (*Dendroica caerulescens*); Bearhop *et al.* 2004) and the timing of arrival on breeding areas (American redstarts; Marra *et al.* 1998; Norris *et al.* 2004). Norris *et al.* (2004) demonstrated that carry-over effect of winter habitat on arrival date ultimately predicted apparent reproductive success, with early arriving birds fledging more offspring. However, the carry-over effects may also influence whether males achieve polygyny as well as within-pair (WP) and extra-pair (EP) paternity—processes that are important drivers of sexual selection in many songbirds (Webster *et al.* 2007). Thus, examining the carry-over effects using only apparent reproductive success

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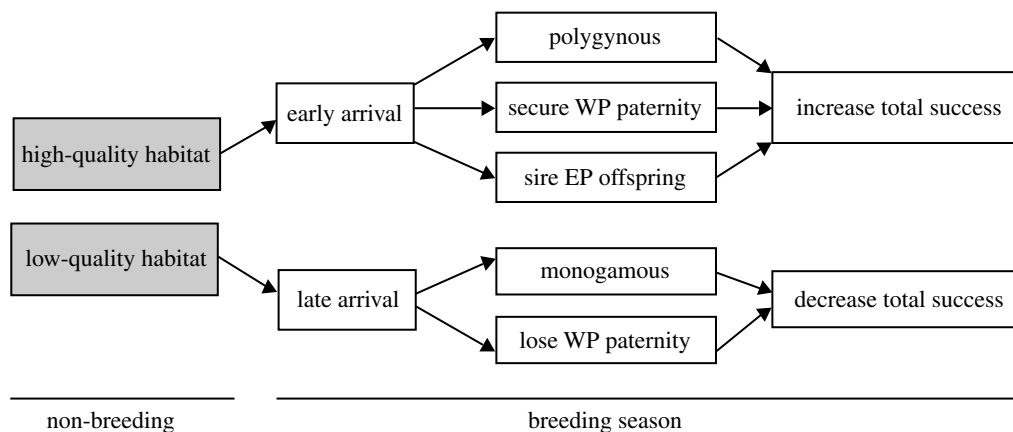


Figure 1. Diagram illustrating the predicted pathway by which winter habitat quality may carry over to the breeding season to influence total genetic success.

is likely to considerably underestimate the variance in true (genetic) reproductive success (Albecht *et al.* 2007; Webster *et al.* 2007) and miss the critical factors influencing individual fitness and the opportunity for sexual selection.

The study of sexual selection has traditionally focused on the events and behaviours immediately surrounding copulation (i.e. courtship through insemination) and relegated events spatially or temporally separated from copulation to the realm of natural selection. We investigate whether the process of sexual selection may be influenced by non-breeding season carry-over effects through variation in arrival dates. Arrival scheduling may be particularly important for species that employ a mixed-mating strategy, whereby polygyny and EP paternity depend heavily on individuals arriving early enough to pair and secure paternity at the primary nest and advertise for a secondary female and/or EP partners (Spottiswoode *et al.* 2006).

In this study, we investigate whether carry-over effects from the non-breeding season may influence polygyny and EP paternity through variation in arrival dates on the breeding grounds in a long-distance neotropical migratory bird, the American redstart (figure 1). Previous studies have shown that American redstarts employ a mixed-mating strategy with high levels of EP paternity (59% of broods, 40% of offspring; Perrault *et al.* 1997), and a moderate level of polygyny (5–16% of males; Secunda & Sherry 1991). We predict that by overwintering in a high-quality habitat, birds will arrive on the breeding grounds earlier, will be more likely to achieve polygyny and will secure more paternity at their own nest. Additionally, early arriving males should sire more EP offspring (EPO) in the nests of late-arriving males. Ultimately, we predict that early arrival on the breeding grounds will result in increased realized fertilization success and fledging success (figure 1).

2. MATERIAL AND METHODS

(a) Field data collection

Fieldwork was conducted in May–July 2004–2007 at Queen's University Biological Station, Chaffey's Lock, Ontario, Canada (44°34' N, 76°19' W). Our study area is composed of mixed-deciduous forest, dominated by sugar maple (*Acer saccharum*) and Eastern hop hornbeam (*Ostrya virginiana*). When males arrive on the breeding grounds, they immediately begin singing for territory advertisement and to attract

females. Each year, from May 1 to 31, we surveyed our 60 ha study area daily from 06.00 to 12.00, detecting males by the presence of singing and subsequent visual identification. Arrival date was standardized as the number of days after the first male arrived (first-male arrival date = 0). All adults were captured in mist-nets within 7 days of arrival by simulating territorial intrusions using song playbacks accompanied with a decoy. Once captured, American redstarts were individually marked with a single Canadian Wildlife Service aluminium band and two to three colour bands. We then extracted 50 μ l of blood for paternity analysis by piercing the brachial vein and clipped 2–3 mm of the central claw for stable-isotope analysis (2006 and 2007 only).

Upon arrival, all males were observed and mapped for at least 20–30 min d^{-1} throughout the breeding season to determine territory boundaries and pairing date. Females typically begin nest building within a few days of pairing. Once nest building began, we monitored nest status every other day, noting the onset of egg laying, number of eggs laid, hatching and fledging success. Males were monitored daily to detect individuals that paired with secondary females (i.e. polygynous mating). At day 5 after hatching, we banded nestlings with a single aluminium band and collected 15–20 μ l of blood for paternity analysis. Offspring from nests that were too high to access on day 5 were captured on the day of fledging.

American redstart males exhibit delayed plumage maturation, wherein males resemble females during their first breeding season and do not mature into the full adult breeding plumage until their second prebasic moult, which follows their first breeding season. Owing to the differences in plumage, and the fact that the first-year American redstarts have greatly reduced reproductive performance (Sherry & Holmes 1997; M. W. Reudink 2007 unpublished data), we limited our analyses to only adult (after second-year or ASY) males.

(b) Winter habitat quality and stable-carbon isotope analysis

American redstarts are widespread across Central America, northern South America, and are especially abundant throughout the Caribbean (Sherry & Holmes 1997). Although the specific overwintering location of our breeding population is unknown, stable-hydrogen isotope analysis indicates that birds breeding in southeastern Ontario probably overwinter in the Caribbean (Norris *et al.* 2006).

Table 1. American redstart microsatellite data characterization from CERVUS v. 2.0 (Marshall *et al.* 1998) over the 4 years of this study (2004–2007).

locus	no. of alleles	expected heterozygosity (h_e)	observed heterozygosity (h_o)	probability of maternal exclusion	probability of paternal exclusion	null allele frequency
<i>Dpu01</i>	29	0.945	0.866	0.789	0.885	+0.043
<i>Dpu03</i>	13	0.479	0.460	0.133	0.302	+0.018
<i>Dpu05</i>	27	0.949	0.824	0.808	0.894	+0.070
<i>Dpu15</i>	19	0.880	0.772*	0.611	0.759	+0.065
<i>Dpu16</i>	17	0.904	0.906	0.671	0.804	+0.002
all loci	21 (avg)	0.831	0.764	0.996	> 0.999	+0.040

*Significantly different from expected (goodness-of-fit test: $X^2 = 31.90$, $p < 0.001$).

On the wintering grounds in Jamaica, American redstarts are ubiquitous across the island and defend small territories (less than 0.25 ha; Sherry & Holmes 1997) throughout the non-breeding season. Quality of these habitats is defined primarily by water availability (Marra *et al.* 1998): high-quality habitats such as mangrove forest and wet upland forest have high water availability throughout the non-breeding season, while low-quality second-growth scrub and citrus plantations are highly seasonal and extremely water stressed during much of the non-breeding period (Marra *et al.* 1998; Studts & Marra 2005). These different habitats are patchily distributed across the island and movement between habitat types is negligible.

Stable-carbon isotope signatures of plants in the tropics vary by habitat type owing to the differences in plant water stress and photosynthetic system (Lajtha & Marshall 1994). These signatures are transferred up the food chain and eventually incorporated into birds' tissues (Marra *et al.* 1998). Owing to the differences in water availability and plant water stress in these different habitats, American redstart territory quality during the non-breeding period is well described by $\delta^{13}\text{C}$ signatures (Marra *et al.* 1998; Reudink *et al.* 2009). By capturing birds upon arrival on the breeding grounds, we can infer the quality of winter habitat; more negative $\delta^{13}\text{C}$ signatures are indicative of higher quality territories (Marra *et al.* 1998; Norris *et al.* 2004; Reudink *et al.* 2009). Stable-carbon isotopes in claw tissue turnover at the rate of weeks to months, making claws an ideal tissue to sample across the migratory period (Bearhop *et al.* 2003, 2004). We analysed only birds that were captured within 7 days of arrival from May 1 to 31, to ensure carbon isotope signatures reflected winter habitat type. Claw samples were weighed, converted to CO_2 in an oxidation/reduction furnace, separated by gas chromatography, then measured for $\delta^{13}\text{C}$ with an isotope ratio mass spectrometer (Lajtha & Marshall 1994; Reudink *et al.* 2009). All isotope analyses were conducted at the Queen's University Facility for Isotope Research (Kingston, ON, Canada).

(c) Paternity analysis

We collected blood samples from putative parents and offspring and stored the samples in Queen's lysis buffer (Seutin *et al.* 1991; 2005–2007) or on blotting paper (2004). DNA was extracted using an Invitrogen Blood and Tissue Kit. gDNA was then quantified via agarose gel electrophoresis and diluted or concentrated to approximately $10 \text{ ng } \mu\text{l}^{-1}$. All loci were amplified using a Biometra Thermogradient or Biometra UNOII PCR machine under the following conditions: 94°C for 3 min followed by 35

cycles of 94°C for 15 s, 58°C for 15 s, 72°C for 30 s, and a final extension of 72°C for 10 min. Each sample included $1 \mu\text{l}$ DNA ($10 \text{ ng } \mu\text{l}^{-1}$), $1 \mu\text{l}$ $10\times$ Qiagen PCR buffer, $0.03 \mu\text{l}$ (100 mM) dNTPs, $0.03 \mu\text{l}$ (100 μM) forward primer, $0.03 \mu\text{l}$ (100 μM) reverse primer, $0.025 \mu\text{l}$ M13 F 700IRD licor primer, $0.005 \mu\text{l}$ (5 U μl^{-1}) Taq polymerase, brought up to $10 \mu\text{l}$ total volume with sterile ddH₂O. Amplified samples were run on a Licor IR2 Global Sequencer and allele scoring was conducted by a trained observer blind to the identity of individuals.

Paternity analysis was conducted using five microsatellite loci (*Dpu01*, *Dpu03*, *Dpu05*, *Dpu15* and *Dpu16*) originally isolated from yellow warblers (*Dendroica petechia*; Dawson *et al.* 1997, table 1). Over the 4 years of this study, we analysed DNA from 265 offspring from 75 nests and all putative parents. The use of five highly variable microsatellite loci ensured a high probability of paternity exclusion (>0.999 , table 1). Owing to limitations in detecting 2 bp differences and the relatively high frequency of null alleles, we followed the conservative approach of Reudink *et al.* (2006): offspring were excluded only if they mismatched the putative sire at more than 2 bp and at two or more loci. All EPO were then compared with all sampled potential sires in the population to assign paternity using CERVUS v. 2.0 (Marshall *et al.* 1998). All assignments were then double-checked by hand by two trained, independent observers. EP paternity was assigned when the putative sire matched at least four out of five loci within 2 bp. Mismatches at single loci were only allowed when the mismatch was due to a likely null allele. Total fertilization success was calculated as the number of WP offspring at the primary and secondary (if polygynous) nests and the number of EPO sired. Some offspring died prior to fledging and we were unable to ascertain the identity of all fledged offspring. Thus, we calculated genetic fledging success by multiplying the number of offspring fledged by the proportion of WP offspring for the primary and secondary nests and added the number of EPO \times proportion fledged at the EP nest.

(d) Predicting paternity and polygyny

To illustrate the potential carry-over effects resulting from holding a territory in tropical winter habitats that vary in quality, we created a simple model based on $\delta^{13}\text{C}$ signatures of tissues from individuals collected in four different winter habitats (wet forest, mangrove, citrus and scrub; *sensu* Norris *et al.* 2004). Because winter habitat quality is unlikely to influence paternity directly, but rather through factors associated with arrival timing (see Norris *et al.* 2004), we first predicted average arrival dates for birds arriving from

those four winter habitat types based on the regression of $\delta^{13}\text{C}$ on arrival. Next, calculated the probability of polygyny, proportion paternity and realized fledging success based on regressions of polygyny, proportion paternity and realized fledging success on arrival.

(e) Statistical analysis

All statistical analyses were performed in JMP v. 5.1 (SAS Institute 2006) and SAS v. 8.2 (SAS Institute 1999). We used a mixed model with individual as a random effect and $\delta^{13}\text{C}$ and year as main effects to test the relationship between $\delta^{13}\text{C}$ and arrival. Because some individuals were present in multiple years, we tested whether arrival date predicted binary reproductive variables (EP paternity (y/n) and polygyny/monogamy) using logistic regression with individual as a repeated measure and year as a random effect. To test whether arrival date predicted linear response variables (fertilization success and fledging success), we used a mixed model with the reproductive variable as the response, year as a random effect, individual as a repeated measure and standardized arrival date as a linear covariate. If year was not found to have a significant effect on the model, we excluded the year term and used a mixed model with individual as a random effect and standardized arrival date as a linear covariate. To test for a relationship between the arrival date and the proportion of WP offspring, we used a non-parametric mixed model with proportion paternity as a response variable, year as a random effect, individual as a repeated measure and standardized arrival date as a linear covariate (PROC MIXED in SAS). It was necessary to take a non-parametric approach as our proportion paternity data were highly skewed and could not be transformed. Comparisons between WP and EPO were analysed using matched-pairs *t*-tests.

3. RESULTS

(a) Winter habitat quality and arrival date

Adult males that overwintered in high-quality habitats (more negative claw $\delta^{13}\text{C}$ signatures) in 2006–2007 arrived on the breeding grounds earlier than males that overwintered in low-quality habitats (mixed model: claw $\delta^{13}\text{C}$: $n=43$, $F=7.56$, $p=0.009$; year: $n=43$, $F=0.36$, $p=0.55$; figure 2a).

(b) Paternity

Out of the 75 nests analysed, 32 (43%) contained one or more EPO and 56 out of the 239 (23%) offspring analysed were EP. A subset of males (9/75) was polygynous, but there was no significant difference in paternity at the nest of the primary female or the secondary female (first females: 28 out of 66 (42%) of nests contained EPO, 52 out of 209 (25%) of offspring were EPO; second females: 4 out of 9 (45%) of nests contained EPO, 4 out of 30 (13%) of offspring were EPO; Fisher's exact probability test for the presence of EPO: $n=9$; $r^2=0.42$, $p=0.17$; paired *t*-test for proportion EPO: $n=9$, $t=0.58$, $p=0.58$). Realized success (WP offspring at each nest + EPO sired) for 62 adult males with complete reproductive data ranged from zero to seven offspring (mean = 3.13 ± 0.24 s.e.).

(c) Arrival date and paternity

Adult males that sired all the offspring at their own nest arrived earlier on the breeding grounds than males that

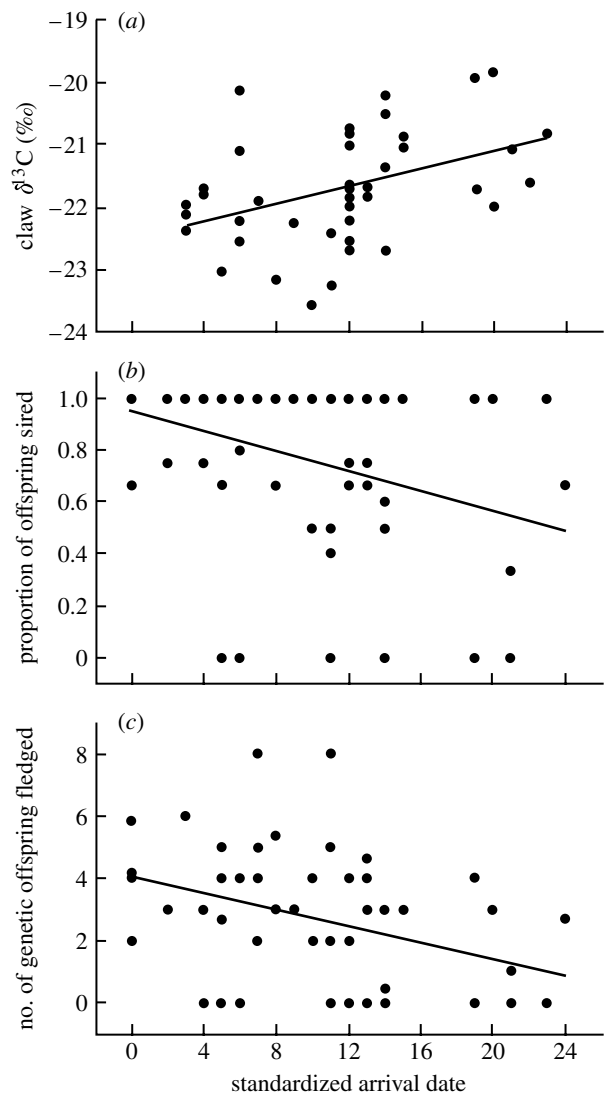


Figure 2. Significant relationships between standardized arrival date (number of days after first male to arrive) on the breeding grounds and (a) claw $\delta^{13}\text{C}$, (b) proportion of offspring sired by the social male at a nest and (c) total genetic fledging success (within pair + EPO).

lost paternity (logistic regression with correlated data; arrival: $n=64$, $z=2.00$, $p=0.045$; year: $n=64$, $z=-0.65$, $p=0.52$). Arrival date of adult males was also significantly correlated with the proportion of a WP offspring male sired (non-parametric random effects mixed model: $n=64$, $\chi^2=5.95$, $p=0.01$; figure 2b). At nests that lost paternity, EP sires arrived on average 4.19 ± 1.83 s.d. days earlier than the males they cuckolded (matched-pairs *t*-test: $n=26$, $t=-2.28$, $p=0.03$). However, an examination of all adult males revealed no difference in arrival date between males that sired EPO and those that did not (logistic regression: arrival: $n=115$, $\chi^2=0.53$, $p=0.47$; year: $n=115$, $\chi^2=2.59$, $p=0.11$).

(d) Arrival date and polygyny

Males that achieved polygyny arrived significantly earlier than males who remained socially monogamous (logistic regression with correlated data: $n=115$ (86 monogamous, 29 polygynous), arrival: $z=-2.04$, $p=0.04$; year: $z=-1.78$, $p=0.08$; figure 3). These results did not change qualitatively when we removed the non-significant year effect ($z=-2.59$, $p=0.01$).

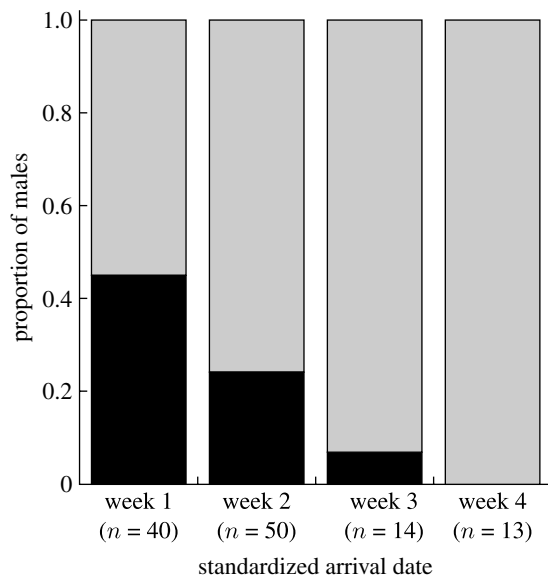


Figure 3. Proportion of adult males arriving during weeks 1–4 of arrival that were polygynous (black bars) and monogamous (grey bars) during the season.

(e) Realized success

Males that arrived early had higher, although not significantly, realized fertilization success (mixed model; arrival: $n=65$, $F=3.14$, $p=0.11$; year: $n=65$, $F=0.17$, $p=0.92$) and fledging success (mixed model; arrival: $n=65$, $F=3.50$, $p=0.09$; year: $n=65$, $F=2.08$, $p=0.17$; figure 2c). When we removed the non-significant year effects and tested the relationships between arrival and realized fertilization success and realized fledging success using individual as a random effect and arrival as a linear covariate, we found that males that arrived earlier had significantly higher fertilization (mixed model; $n=65$, $F=4.03$, $p=0.05$) and realized fledging success (mixed model; $n=65$, $F=7.52$, $p=0.008$).

(f) Predicting paternity, polygyny and total fledging success

We calculated the average (mean \pm s.e.) $\delta^{13}\text{C}$ signatures of the American redstarts from four tropical habitat types (from wet to dry: wet forest ($\delta^{13}\text{C} = -24.35 \pm 0.11$; $n=15$), mangrove ($\delta^{13}\text{C} = -22.80 \pm 0.16$; $n=34$), citrus ($\delta^{13}\text{C} = -22.16 \pm 0.20$; $n=16$), scrub ($\delta^{13}\text{C} = -21.69 \pm 0.16$; $n=24$); Marra et al. 1998; Norris et al. 2004; Marra unpublished data). We then calculated the average (mean \pm s.e.) arrival dates for birds arriving from those four habitat types, based on the regression of $\delta^{13}\text{C}$ on arrival date for birds arriving on the breeding grounds (figure 2a). Next, we used those four arrival dates (days 4.61 ± 0.29 , 8.83 ± 0.44 , 10.55 ± 0.55 and 11.83 ± 0.43) to predict the proportion of offspring an individual was likely to sire based on the regression of the proportion of WP offspring on the arrival date (figure 2b). We then predicted the probability of being polygynous based on a regression of the probability of polygyny on the arrival date. Finally, we predicted realized fledging success based on a regression of realized fledging success on the arrival date (figure 2c). Our model suggests that males arriving on the breeding grounds from high-quality winter habitats sire 14 per cent more offspring at their primary nest and are 19 per cent more likely to be polygynous than males arriving later from low-quality habitats (figure 4a,b). Ultimately,

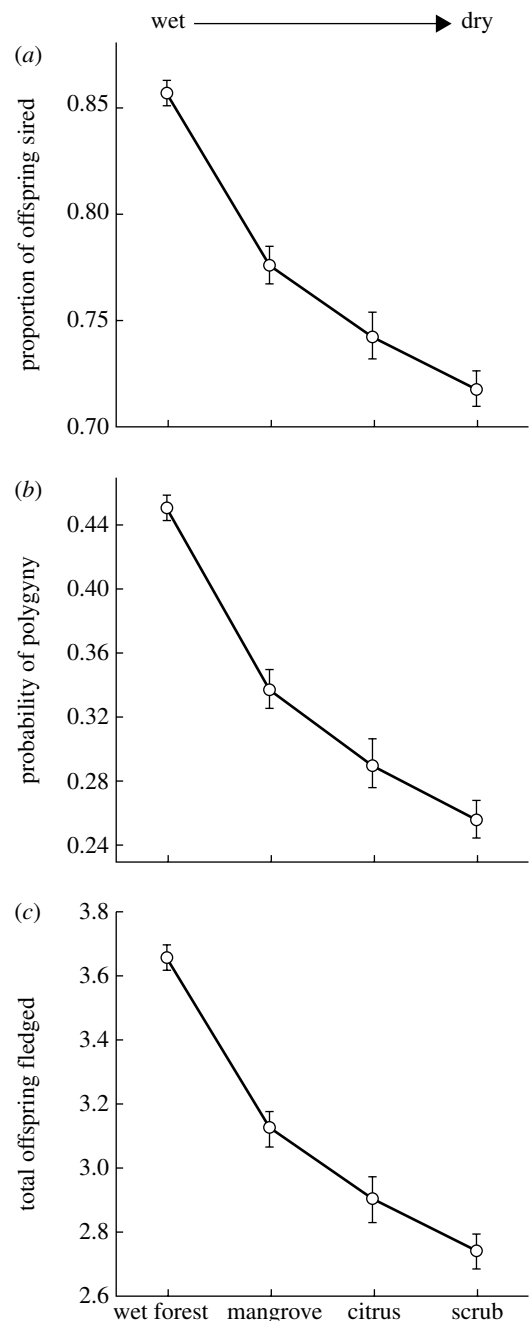


Figure 4. Predicted values (mean \pm s.e.) for (a) proportion of offspring sired by WP males, (b) probability of achieving polygyny and (c) total number of genetic offspring fledged for males arriving from four different tropical habitat types. Hypothetical arrival dates for individuals wintering in the four tropical habitat types were obtained by predicting arrival dates for the four given $\delta^{13}\text{C}$ values based on the regression of $\delta^{13}\text{C}$ and arrival date. Those four arrival dates (days 4.61 ± 0.29 , 8.83 ± 0.44 , 10.55 ± 0.55 and 11.83 ± 0.43 , respectively) were then used to predict (a) the proportion of offspring sired, (b) the probability of being polygynous and (c) the total number of genetic offspring fledged.

males overwintering in the high-quality winter habitats fledge approximately one additional offspring than birds overwintering in the poor-quality habitats (figure 4c).

4. DISCUSSION

Our results indicate that arrival timing on the breeding grounds, driven by conditions that experienced thousands

of kilometres away on tropical wintering grounds, may influence the rates of polygyny and EP paternity in American redstarts, suggesting that non-breeding season events may carry over to influence the process of sexual selection. Specifically, we demonstrate that not only does winter territory quality influence apparent success through arrival timing (wherein variation in apparent success is driven largely by predation; Norris *et al.* 2004), but also carry-over effects from winter may influence behavioural processes such as female mate choice and male–male competition. Males overwintering in high-quality habitats arrive earlier on the breeding grounds than males overwintering in low-quality habitats (inferred by stable-carbon isotope analysis). In turn, early arriving males sire a higher proportion of their own offspring, sire EPO in the nests of late-arriving males, are more likely to achieve polygyny and ultimately fledge a greater number of genetic offspring. These results suggest that early male arrival, driven by winter habitat quality, has the potential to directly influence evolutionary processes (Spottiswoode *et al.* 2006).

Several species of migratory birds are returning earlier to the breeding grounds and recent work has suggested that long-distance migrants breeding in Scandinavia have advanced arrival dates as a result of rapid, climate-driven evolutionary change (Jonzen *et al.* 2006; but see Both 2007). Our data indicate that early arriving males may increase fledging success by approximately 25 per cent through variation in polygyny and EP paternity, suggesting that early arrival is strongly favoured by selection and may be a mechanism by which rapid evolutionary change may occur.

Previous studies of American redstarts on the wintering grounds have shown that individuals holding territories in poor-quality habitats delay their spring departure northwards from the wintering grounds (Marra *et al.* 1998; Studds & Marra 2005), and arrive on the breeding grounds later (Marra *et al.* 1998; Norris *et al.* 2004; this study) and in poorer condition (Marra *et al.* 1998). Obtaining a territory in high-quality habitat during the winter is to some degree driven by individual quality, which could suggest that the relationship between winter territory quality and reproductive success is driven, at least in part, by individual quality rather than a carry-over effect of winter habitat quality. However, Studds & Marra (2005) provided strong experimental evidence showing that American redstarts overwintering in low-quality habitats can ‘turn into’ high-quality birds through an experimental upgrade in habitat quality. When birds were removed from high-quality habitats that were subsequently resettled by birds from low-quality habitats, those new settlers were in better condition throughout the overwinter period and ultimately advanced their spring departure dates relative to birds that remained in low-quality habitats (Studds & Marra 2005). While disentangling the effects of individual quality from the effects of habitat quality is onerous, these data strongly support the idea that spring departure and subsequent arrival on the breeding grounds are influenced by ecological conditions during winter, not just individual condition/quality. Future studies that track individuals throughout the annual cycle will allow us to explicitly identify the impacts of altering territory quality on arrival date and eventual reproductive success.

While previous work on our breeding population of American redstarts has shown that late arrival is associated with reduced apparent reproductive success (Norris *et al.* 2004), our study demonstrates a strong relationship between winter territory quality and rates of EP paternity and polygyny through variation in arrival scheduling and suggests that non-breeding season events may influence sexual selection. While experimental work is still needed, we suggest that the costs of wintering in a low-quality territory are compounded by the mixed-mating strategy employed by American redstarts. Late arrival on the breeding grounds appears to lead not only to a shortened breeding season and higher probability of nest predation (Lozano *et al.* 1996; Norris *et al.* 2004), but also to an increased probability of cuckoldry from early arriving males. Furthermore, late-arriving males are less likely to mitigate the costs of lost paternity through either EP copulations or polygyny. Our model suggests that the acquisition of a poor-quality territory during winter may result in a paternity loss of approximately 14 per cent and a 19 per cent lower probability of polygynous mating, ultimately resulting in an over 25 per cent reduction in total fledging success, or one less successful offspring fledged per season (figure 4c). To our knowledge, this is the first study to demonstrate that genetic success may be influenced by non-breeding season events in a migratory songbird.

Previous studies have suggested that protandry and early male arrival is favoured by sexual selection (Thusius *et al.* 2001; Coppack *et al.* 2006; Kokko *et al.* 2006; Spottiswoode *et al.* 2006), where early arriving individuals increase their probability of obtaining mates (Lozano *et al.* 1996), achieving polygyny (Hasselquist 1998) and increasing reproductive performance (Norris *et al.* 2004; Smith & Moore 2005). Early arrival may also influence EP paternity through density-dependent effects and breeding synchrony/asynchrony (Westneat *et al.* 1990; Birkhead & Møller 1992; Chuang *et al.* 1999; Lindstedt *et al.* 2007). However, these studies have generally assumed that individual variation in the arrival date is dependent on individual quality and condition during migration, largely ignoring the factors affecting variation in arrival scheduling, such as non-breeding season carry-over effects. Ample evidence has now accumulated, which suggests that spring migration scheduling and condition during migration are influenced by the conditions experienced during the non-breeding season (Marra *et al.* 1998; Bearhop *et al.* 2004; Norris *et al.* 2004; Saino *et al.* 2004a; Studds & Marra 2005). We suggest that processes that can influence the opportunity for sexual selection, such as variation in EP paternity and polygyny (Andersson 1994), should be viewed not only in terms of the events and processes occurring during the breeding season, but rather as a continual process that may be influenced by events occurring throughout the annual cycle.

All methods in this study complied with the laws of Canada.

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REFERENCES

- Albecht, T., Schnitzer, J., Kreisinger, J., Exnerova, A., Bryja, J. & Munclinger, P. 2007 Extrapair paternity and the opportunity for sexual selection in long-distant migratory passerines. *Behav. Ecol.* **18**, 477–486. (doi:10.1093/beheco/arm001)
- Andersson, M. 1994 *Sexual selection*. Princeton, NJ: Princeton University Press.
- Bearhop, S., Furness, R. W., Hilton, G. M., Votier, S. C. & Waldron, S. 2003 A forensic approach to understanding diet and habitat use from stable isotope analysis of (avian) claw material. *Funct. Ecol.* **17**, 270–275. (doi:10.1046/j.1365-2435.2003.00725.x)
- Bearhop, S., Hilton, G. M., Votier, S. C. & Waldron, S. 2004 Stable isotope ratios indicate that body condition in migrating passerines is influenced by winter habitat. *Biol. Lett.* **271**, 215–218.
- Birkhead, T. R. & Møller, A. P. 1992 *Sperm competition in birds: evolutionary causes and consequences*. London, UK: Academic Press.
- Both, C. 2007 Comment on 'rapid advance of spring arrival dates in long-distance migratory birds'. *Science* **315**, 598b. (doi:10.1126/science.1136148)
- Chuang, H. C., Webster, M. S. & Holmes, R. T. 1999 Extrapair paternity and local synchrony in the black-throated blue warbler. *Auk* **116**, 726–736.
- Coppack, T., Tøttrup, A. P. & Spottiswoode, C. 2006 Degree of protandry reflects level of extrapair paternity in migratory songbirds. *J. Ornithol.* **147**, 260–265. (doi:10.1007/s10336-006-0067-3)
- Dawson, R. J. G., Gibbs, H. L., Hobson, K. A. & Yezerinac, S. M. 1997 Isolation of microsatellite DNA markers from a passerine bird, *Dendroica petechia* (the yellow warbler), and their use in population studies. *Heredity* **79**, 506–514. (doi:10.1038/hdy.1997.190)
- Greenberg, R. & Marra, P. P. (eds) 2005 *Birds of two worlds: the ecology and evolution of migration*. Baltimore, MD: Johns Hopkins University Press.
- Hasselquist, D. 1998 Polygyny in great reed warblers: a long-term study of factors contributing to male fitness. *Ecology* **79**, 2376–2390.
- Jonzen, N., Linden, A., Ergon, T., Knudsen, E., Vik, J. O. & Rubolini, D. 2006 Rapid advance of spring arrival dates in long-distance migratory birds. *Science* **312**, 1959–1961. (doi:10.1126/science.1126119)
- Kelly, J. F., Atudorei, V., Sharp, Z. D. & Finch, D. M. 2002 Insights into Wilson's warbler migration from analyses of hydrogen stable-isotope ratios. *Oecologia* **130**, 216–221.
- Kokko, H., Gunnarsson, T. G., Morrell, L. J. & Gill, J. A. 2006 Why do female migratory birds arrive later than males? *J. Anim. Ecol.* **75**, 1293–1303. (doi:10.1111/j.1365-2656.2006.01151.x)
- Lajtha, K. & Marshall, J. D. 1994 Sources of variation in isotopic composition in plants. In *Stable isotopes in ecological and environmental sciences* (eds K. Lajtha & J. D. Marshall), pp. 1–21. London, UK: Oxford University Press.
- Lindstedt, E. R., Oh, K. P. & Badyaev, A. V. 2007 Ecological, social, and genetic contingency of extrapair behaviour in a socially monogamous bird. *J. Avian Biol.* **38**, 214–223. (doi:10.1111/j.0908-8857.2007.03889.x)
- Lozano, G. A., Perrault, S. & Lemon, R. E. 1996 Age, arrival date and reproductive success of male American redstarts *Setophaga ruticilla*. *J. Avian Biol.* **27**, 164–170. (doi:10.2307/3677146)
- Marra, P. P., Hobson, K. A. & Holmes, R. T. 1998 Linking winter and summer events in a migratory bird by using stable-carbon isotopes. *Science* **282**, 1884–1886. (doi:10.1126/science.282.5395.1884)
- Marra, P. P. & Holberton, R. L. 1998 Corticosterone levels as indicators of habitat quality: effects of habitat segregation in a migratory bird during the non-breeding season. *Oecologia* **116**, 284–292.
- Marra, P. P. & Holmes, R. T. 2001 Consequences of dominance-mediated habitat segregation in a migratory passerine bird during the non-breeding season. *Auk* **118**, 92–104. (doi:10.1642/0004-8038(2001)118[0092:CODMHS]2.0.CO;2)
- Marshall, T. C., Slate, J., Kruuk, L. E. B. & Pemberton, J. M. 1998 Statistical confidence for likelihood-based paternity inference in natural populations. *Mol. Ecol.* **7**, 639–655. (doi:10.1046/j.1365-294x.1998.00374.x)
- Norris, D. R., Marra, P. P., Kyser, T. K., Sherry, T. W. & Ratcliffe, L. M. 2004 Tropical winter habitat limits reproductive success on the temperate breeding grounds in a migratory bird. *Proc. R. Soc. Lond. B* **271**, 59–64. (doi:10.1098/rspb.2003.2569)
- Norris, D. R., Marra, P. P., Bowen, G. J., Ratcliffe, L. M., Royle, J. A. & Kyser, T. K. 2006 Migratory connectivity of a widely distributed Neotropical–Nearctic songbird. *Ornithol. Monogr.* **61**, 14–28.
- Perrault, S., Lemon, R. E. & Kuhnlein, U. 1997 Patterns and correlates of extrapair paternity in American redstarts (*Setophaga ruticilla*). *Behav. Ecol.* **8**, 612–621. (doi:10.1093/beheco/8.6.612)
- Reudink, M. W., Mech, S. G. & Curry, R. L. 2006 Extrapair paternity and mate choice in a chickadee hybrid zone. *Behav. Ecol.* **17**, 56–62. (doi:10.1093/beheco/ari098)
- Reudink, M. W., Studds, C. E., Marra, P. P., Kyser, T. K. & Ratcliffe, L. M. 2009 Plumage brightness predicts non-breeding season territory quality in a long-distance migratory songbird, the American redstart *Setophaga ruticilla*. *J. Avian Biol.* **40**, 34–41. (doi:10.1111/j.1600-048X.2008.04377.x)
- Rubenstein, D. R. & Hobson, K. A. 2004 From birds to butterflies: animal movement patterns in stable isotopes. *Trends Ecol. Evol.* **19**, 256–263. (doi:10.1016/j.tree.2004.03.017)
- SAS Institute 1999 *SAS/STAT users guide, v. 8.2*. Cary, NC: SAS Institute.
- SAS Institute 2006 *JMP statistical discovery 6.0.2*. Pacific Grove, CA: Duxbury.
- Saino, N., Szep, T., Ambrosini, R., Romano, M. & Møller, A. P. 2004a Ecological conditions during winter affect sexual selection and breeding in a migratory bird. *Proc. R. Soc. Lond. B* **271**, 681–686. (doi:10.1098/rspb.2003.2656)
- Saino, N., Szep, T., Romano, M., Rubolini, D., Spina, F. & Møller, A. P. 2004b Ecological conditions during winter predict arrival date at the breeding quarters in a trans-Saharan migratory bird. *Ecol. Lett.* **7**, 21–25. (doi:10.1046/j.1461-0248.2003.00553.x)
- Secunda, R. C. & Sherry, T. W. 1991 Polyterritorial polygyny in the American redstart. *Wilson Bull.* **103**, 190–203.
- Seutin, G., White, B. N. & Boag, P. T. 1991 Preservation of avian blood and tissue samples for DNA analyses. *Can. J. Zool.* **69**, 82–90. (doi:10.1139/z91-013)
- Sherry, T. W. & Holmes, R. T. 1997 American redstart (*Setophaga ruticilla*). In *The birds of North America* (eds A. Poole & F. B. Gill). Washington, DC: Academy of Natural Science, Philadelphia and American Ornithological Union.

- Smith, R. J. & Moore, F. R. 2005 Arrival timing and seasonal reproductive performance in a long-distance migratory landbird. *Behav. Ecol. Sociobiol.* **57**, 231–239. (doi:10.1007/s00265-004-0855-9)
- Spottiswoode, C., Tøttrup, A. P. & Coppack, T. 2006 Sexual selection predicts advancement of avian spring migration in response to climate change. *Proc. R. Soc. B* **273**, 3023–3029. (doi:10.1098/rspb.2006.3688)
- Studds, C. E. & Marra, P. P. 2005 Nonbreeding habitat occupancy and population processes: an upgrade experiment with a migratory bird. *Ecology* **86**, 2380–2385. (doi:10.1890/04-1145)
- Studds, C. E., Kyser, T. K. & Marra, P. P. 2008 Natal dispersal driven by environmental conditions interacting across the annual cycle of a migratory songbird. *Proc. Natl Acad. Sci. USA* **105**, 2929–2933. (doi:10.1073/pnas.0710732105)
- Thusius, K. J., Peterson, K. A., Dunn, P. O. & Whittingham, L. A. 2001 Male mask size is correlated with mating success in the common yellowthroat. *Anim. Behav.* **62**, 435–446. (doi:10.1006/anbe.2001.1758)
- Webster, M. S., Marra, P. P., Haig, S. M., Bensch, S. & Holmes, R. T. 2002 Links between worlds: unraveling migratory connectivity. *Trends Ecol. Evol.* **17**, 76–83. (doi:10.1016/S0169-5347(01)02380-1)
- Webster, M. S., Tarvin, K. A., Tuttle, E. M. & Pruett-Jones, S. 2007 Promiscuity drives sexual selection in a socially monogamous bird. *Evolution* **61**, 2205–2211. (doi:10.1111/j.1558-5646.2007.00208.x)
- Westneat, D. F., Sherman, P. W. & Morton, M. L. 1990 The ecology and evolution of extra-pair copulations in birds. *Curr. Ornithol.* **7**, 331–369.
- Yohannes, E., Hobson, K. A. & Pearson, D. J. 2007 Feather stable-isotope profiles reveal stopover habitat selection and site fidelity in nine migratory species moving through sub-Saharan Africa. *J. Avian Biol.* **38**, 347–355. (doi:10.1111/j.0908-8857.2007.03927.x)