

Stable isotope ratios indicate that body condition in migrating passerines is influenced by winter habitat

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Recd 19.09.03; Accptd 23.10.03; Published online 19.01.04

Although predicted some time ago, there has been little success in demonstrating that the overall fitness of migratory birds depends on the combined influences of their experiences over all seasons. We used stable carbon isotope signatures ($\delta^{13}\text{C}$) in the claws of migrating black-throated blue warblers *Dendroica caerulescens* to infer their wintering habitats and investigated whether winter habitat selection can be linked to condition during migration. Resident bird species with low $\delta^{13}\text{C}$ corresponded to selection of more mesic habitats, and migrating birds with low $\delta^{13}\text{C}$ were in better condition than conspecifics with higher $\delta^{13}\text{C}$ signatures. These findings concur with empirical observations on the wintering grounds, where dominants (mostly males) tend to exclude subordinates from mesic areas (considered to be high-quality habitats). We believe that variation in condition during migration may be one of the key factors determining differences in arrival times and condition at the breeding areas, which in turn have a major influence on reproductive success.

Keywords: migration; condition; seasonal interaction; stable isotope

1. INTRODUCTION

It is generally agreed that early arrival on the breeding grounds and good body condition are important determinants of fitness in migratory birds (e.g. Lundberg & Alatalo 1992). Fretwell (1972) suggested that, in seasonal environments, events throughout the year might interact to produce these patterns. However, because of the difficulty in tracking such animals, it is unclear to what extent interactions among seasons may impact upon migratory species, particularly during migration (Webster *et al.* 2002).

Recent developments in the use of stable isotopes have allowed researchers to investigate, for the first time, seasonal interactions in migrant songbirds. Marra *et al.* (1998) demonstrated that American redstarts *Setophaga ruticilla* wintering in mesic, high-quality habitats were not only in better condition and migrated earlier than conspecifics wintering in poorer xeric habitats, but they arrived earlier on the breeding grounds (inferred from comparatively lower stable-carbon-isotope signatures in their pectoral muscle).

Stable isotope ratios in the environment vary according to several well-established processes and thus can be used as local habitat or broad-scale geographical markers in the study of migration (Hobson 1999). For example, the ratio of $^{13}\text{C} : ^{12}\text{C}$ (expressed as $\delta^{13}\text{C}$) in consumer tissues varies according to the proportions of C3, C4 and CAM plants within the foraging area (Hobson 1999). Tissues that remain metabolically inert after formation (e.g. feather or claw) will hold these habitat signatures indefinitely (Hobson & Clark 1992; Bearhop *et al.* 2002). Therefore sampling of claw material in spring can allow inferences to be made about winter habitat selection (Bearhop *et al.* 2003).

We examine whether seasonal interactions explain variability in the condition of black-throated blue warblers *Dendroica caerulescens* (BTBW's) during spring migration. Breeding in eastern North America, the BTBW is a small, insectivorous passerine that winters in the northern Caribbean. We predicted that birds in better condition during northwards migration had wintered in more mesic habitats (high-quality wintering sites). Thus, condition should be negatively correlated with stable carbon isotope signatures of claws, because low $\delta^{13}\text{C}$ values tend to indicate more mesic habitats.

The BTBW is ideally suited to a study of this nature. Its breeding and wintering ecology are relatively well known (e.g. Sillit & Holmes 2002), studies have already reported values for $\delta^{13}\text{C}$ in this species (Chamberlain *et al.* 1997; Rubenstein *et al.* 2002) and it has a relatively small wintering range almost entirely in the Greater Antilles with a small population in the Yucatan peninsula. This is beneficial, since changes in the proportions of C3, C4 and CAM plants produce large-scale spatial variation in $\delta^{13}\text{C}$ (Still *et al.* 2003) and can confound smaller-scale habitat-specific signals when examining tissues synthesized away from the sampling site.

2. METHODS

We trapped 25 BTBW's on North Andros, Bahamas (24°50' N 77°53' W) in mist nets, during 2 days in April 2000 when adverse weather produced a 'fall' of migrant birds. Few birds were likely to have wintered locally, since no BTBW's were caught in the area on the 2 days preceding the fall and only one on the day following. Each individual was weighed (to the nearest 0.5 g), and measured (maximum wing chord, to the nearest millimetre). Subcutaneous fat reserves were estimated via examination of the tracheal pit using a six-point scale and birds were also assigned one of four pectoral muscle scores, both adapted from Redfern & Clark (2001).

A length of claw (1–2 mm) was clipped from each individual. Bearhop *et al.* (2003) demonstrated that claws grow at ca. 0.04 mm d⁻¹ and that there was no correlation between $\delta^{13}\text{C}$ values of BTBW claw collected during the spring and those in tertials grown in the breeding season the previous autumn. Thus, claw tips in spring probably reflect winter habitat selection. The third tertial of one wing was also clipped by 4–10 mm in each individual. Since they were grown during the post-breeding moult, $\delta^{13}\text{C}$ in these feathers reflects habitat selection during the previous (1999) breeding season.

To determine habitat-specific isotope signatures, claws were clipped from insectivorous thick-billed vireos *Vireo crassirostris* (TBVI), a resident found in the same types of habitat as BTBW's

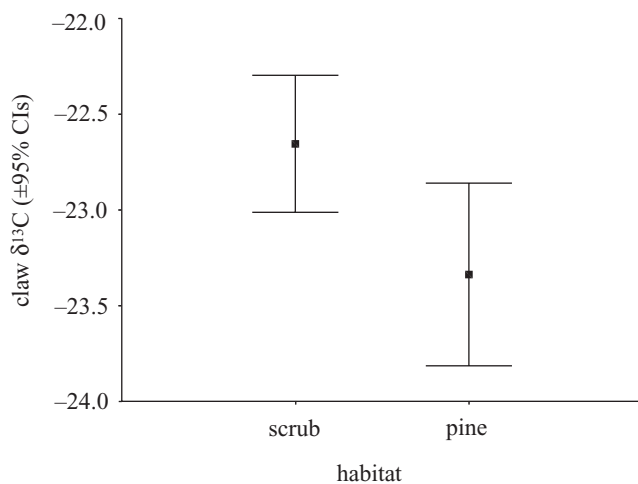


Figure 1. The mean $\delta^{13}\text{C}$ ($\pm 95\%$ confidence interval (CI)) in the claws of TBVIs living in pine forest are significantly lower than those living in scrub.

during winter. The size of TBVI home range is unknown, but a resident congener in the Yucatan is known to defend territories throughout the year (Greenberg *et al.* 1993). Samples were collected at multiple sites on three islands separated by almost 4° of latitude (Long Island: $22^\circ 52' \text{N}$, $74^\circ 58' \text{W}$; Andros and Abaco: $26^\circ 31' \text{N}$, $77^\circ 11' \text{W}$) and assigned one of two habitat types (scrub, with a high prevalence of xerophytic shrubs, and pine forest *Pinus caribaea*). To ensure that they were isotopically representative of the sampling habitat, claw $\delta^{13}\text{C}$ of TBVIs from pine were compared with those collected from resident species found only in this habitat; Bahama yellowthroat *Geothlypis rostrata*, olive-capped warbler *Dendroica pityophila*, yellow-throated warbler *Dendroica dominica* (Bahamian subspecies only) and pine warbler *Dendroica pinus*. TBVIs were the only exclusively resident species captured in scrub.

Isotopic analyses of claws and feathers were conducted using the protocol described in Bearhop *et al.* (2003). Repeat analysis of an internal standard showed that $\delta^{13}\text{C}$ can be measured with accuracy and a precision of 0.2‰ or less.

Data from resident birds were analysed using a generalized linear model (GLM) with claw $\delta^{13}\text{C}$ as the dependent variable, latitude of the sampling locality as a covariate and habitat as a factor (scrub or pine). A measure of 'migratory condition' was derived from a principal components analysis using the correlation between fat and muscle score. Principal component 1 (PC1) accounted for 85% of the variance and was selected as a condition index (factor loadings for fat and muscle were both 0.92). Large fat stores and pectoral muscles should be advantageous to birds during migration, since they provide fuel and power. Thus, they should be honest indicators of migratory condition and are unlikely to be affected by state-dependent regulation, particularly during unplanned stopovers (as is the case here).

To investigate the link between winter habitat and migratory condition, PC1 along with wing-length and mass were used as covariates in a generalized linear mixed model (GLMM), where age (second year, after second year) and sex (two levels) were random factors and the dependent variable was claw $\delta^{13}\text{C}$. The effects of sex and age on migratory condition were investigated using a two-way ANOVA. Associations between condition and migration distances were assessed by regressing PC1 upon $\delta^{13}\text{C}$ signature of tertial feathers; the latter values are correlated with breeding latitude and wintering longitude (Rubenstein *et al.* 2002). The power of this regression analysis was also estimated.

3. RESULTS

Habitat had a significant effect on $\delta^{13}\text{C}$ (claw) of TBVIs (figure 1: GLM, $F_{1,22} = 6.3$, $p = 0.019$), with mean values in pine (mean \pm s.d. = $-23.4 \pm 0.6\text{‰}$, $n = 9$) lower than those found in scrub (mean \pm s.d. = $-22.5 \pm 0.6\text{‰}$, $n = 14$). However, the latitude of capture was not significantly related to $\delta^{13}\text{C}$ (claw) (GLM, $F_{1,22} = 0.463$, $p = 0.503$). Mean carbon isotope values in the claws of TBVIs living in pine (mean \pm s.d. = $-23.4 \pm 0.6\text{‰}$, $n = 9$)

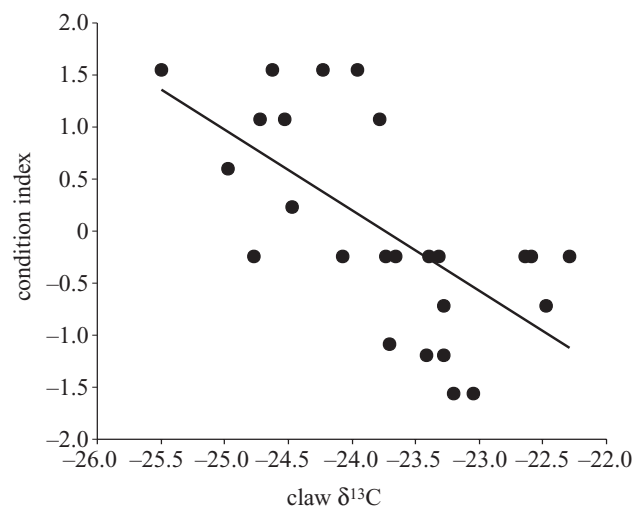


Figure 2. There is a highly significant relationship between condition and claw $\delta^{13}\text{C}$ (indicating winter habitat use) in BTBW's.

were not significantly different from those of other resident species caught in the same habitat (mean \pm s.d. = $-23.2 \pm 0.7\text{‰}$, $n = 31$, $t_{38} = 0.496$, $p = 0.62$). Thus, higher $\delta^{13}\text{C}$ (claw) should be indicative of wintering in a more xeric habitat.

A GLMM indicated no significant sex- and marginally non-significant age-related influences on $\delta^{13}\text{C}$ in the claws of BTBW's (sex: $F_{1,21} = 1.61$, $p = 0.22$; age: $F_{1,22} = 3.56$, $p = 0.07$). Similar patterns were found for wing length and body mass (mass: $F_{1,19} < 0.001$, $p = 0.988$; wing: $F_{1,20} = 0.71$, $p = 0.41$). However, birds with better migratory condition indices tended to have lower $\delta^{13}\text{C}$ (claw), suggesting more mesic wintering areas (figure 2: $F_{1,23} = 16.48$, $r^2 = 0.42$, $p < 0.001$). There was a significant effect of sex upon the migratory condition of individuals, with males tending to have higher condition indices than females. ($F_{1,23} = 9.66$, $p = 0.005$; mean \pm s.d.: males, 0.65 ± 0.93 ; females, -0.44 ± 0.81), with no significant association between age and migratory condition ($F_{1,22} = 0.27$, $p = 0.61$).

There was no significant relationship between $\delta^{13}\text{C}$ of tertial feathers and migratory condition ($F_{23,1} = 0.69$, $p = 0.414$). Standard errors indicate that we could detect relationships with slopes of less than -0.039 or greater than 0.401 and, with a sample size of 25, relationships with r^2 -values as low as 0.15 would have been significant.

4. DISCUSSION

BTBW's that had wintered in habitats with lower $\delta^{13}\text{C}$ signatures were in better body condition during migration with respect to fat reserves and pectoral muscle mass (figure 2), implying that individuals in better condition spent the winter in more moist forest habitats, as has been found in American redstarts (Marra *et al.* 1998).

There are two lines of complementary evidence to support our interpretation:

- (i) stable isotope signatures in the claws of resident Bahamian birds, where TBVIs from relatively mesic pine forest had lower mean $\delta^{13}\text{C}$ values (figure 1) than those caught in drier scrub;

- (ii) studies of BTBW's in their wintering areas indicate that dominant individuals tend to displace subordinates from forest into scrub habitats (Wunderle 1995; Sherry & Holmes 1996).

By implication, mesic forest is the better wintering habitat for this species. The consequences for individuals wintering in sub-optimal habitats may be higher mortality or poorer condition (Marra *et al.* 1998; Sillet & Holmes 2002).

Studies have also shown that winter habitat segregation in BTBW's is often sex biased, with males dominant in primary forest and females and subordinates occupying drier scrub (e.g. Wunderle 1995). Indeed, in our analyses, males were in better condition than females, although we detected no direct effect of sex on winter habitat use as inferred from $\delta^{13}\text{C}$ (possibly owing to lack of statistical power).

Birds in better condition during migration may benefit over conspecifics in several ways. First, these individuals probably have more resources to devote to reproduction when they arrive on the breeding grounds. However, it may also permit shorter inward migration times and earlier arrival on the breeding grounds, allowing access to the highest-quality territories. This is because birds in high migratory condition can spend less time refuelling during stopovers (Gannes 2002) or make longer continuous migration flights.

Marra *et al.* (1998) suggested that dominance-mediated habitat segregation during winter influences the time of arrival of American redstarts at breeding grounds and condition on arrival. Since individuals wintering in lower-quality habitats were in poorer breeding condition on arrival, winter habitat may be a limiting factor for this species. Our study provides evidence that such habitat forcing may also occur with BTBW's, suggesting that the results of Marra *et al.* (1998) have some generality for migratory *Dendroica* warblers.

However, it is important that we consider factors that may confound our interpretation. As we are inferring habitat selection remotely, we cannot directly demonstrate that the observed variation in condition is a consequence of differences in winter habitat use.

Variation in $\delta^{13}\text{C}$ occurs as large-scale latitudinal trends, as well as local habitat-related variability owing to changes in the proportions of C3, C4 and CAM plants. Thus, the relationship shown in figure 2 could be a consequence of large-scale gradients in $\delta^{13}\text{C}$ signatures, whereby sites further south in the wintering range have higher $\delta^{13}\text{C}$, independent of local habitat. In this scenario, birds in poorer condition during migration through the Bahamas would simply have been migrating for longer, from more Southerly wintering sites, and hence would have used up a larger proportion of their initial energy reserves. This is unlikely to be the case for two reasons.

- (i) Across the wintering range of BTBW's (10° of latitude), variability in $\delta^{13}\text{C}$ (vegetation) is at a local scale and there are no obvious large-scale latitudinal patterns (Still *et al.* 2003).
- (ii) There was no effect of latitude at capture on $\delta^{13}\text{C}$ (claw) of the resident TBVIs. Although this range is smaller than for BTBW's, it is sufficiently large that any latitudinal variability in $\delta^{13}\text{C}$ should be detectable.

However, large-scale longitudinal variability in $\delta^{13}\text{C}$ may exist across the wintering range (Still *et al.* 2003). Since the birds were sampled in the central–northern part of the wintering range, the winter residence could have been in a range of longitudes further east. Breeding season $\delta^{13}\text{C}$ values covary with wintering longitude (Rubenstein *et al.* 2002). In our study, there was no significant relationship between individual condition and $\delta^{13}\text{C}$ values representative of the previous breeding season (from tertial feathers), despite considerable statistical power. The absence of any such relationship suggests that our observed results cannot be driven by large-scale longitudinal gradients. Moreover, breeding season $\delta^{13}\text{C}$ also covaries with breeding latitude and thus the non-significant relationship also indicates that birds in better condition were not carrying more reserves because they had further to travel to their breeding areas.

Marra *et al.* (1998) demonstrated that winter habitat selection could influence fitness in migratory birds. Here, we have demonstrated a mechanism that may be one of the drivers of this phenomenon: variation in winter habitat results in systematic variation in condition during return migration. This in turn is likely to result in differences in arrival date at the breeding grounds and in condition on arrival, both of which influence fitness. This paper demonstrates the strong connectivity between wintering and breeding season ecology in many migratory species, and provides rare evidence of the applicability of the seasonal interaction ideas of Fretwell (1972) to migratory birds. Our research emphasizes the need to consider winter ecology and habitat use in population models and ultimately in conservation management of such species.

Acknowledgements

The authors are grateful to Eric Carey, Luther Brown, Colin McLeod, Pericles Mailiss, Maurice Minnis, Dickie Riley, Bahamas Department of Agriculture, Bahamas Natural History Society, Carnegie Trust, RSPB, Percy Sladen Memorial Trust, Glasgow Natural History Society/Blodwen Lloyd Binns Trust and Professor Tony Fallick. S.B. is funded by a NERC postdoctoral fellowship.

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