Patterns of migratory connectivity in Vaux's Swifts at a northern migratory roost: a multi-isotope approach

Running head: Vaux's Swift migratory connectivity

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ABSTRACT

The strength of migratory connectivity between breeding, stopover, and wintering areas can have important implications for population dynamics, evolutionary processes, and conservation. For example, patterns of migratory connectivity may influence the vulnerability of species and populations to stochastic events. For many migratory songbirds, however, we are only just beginning to understand patterns of migratory connectivity. Here, we investigate the potential strength of migratory connectivity within a population of Vaux's Swifts (*Chaetura vauxii*). Vaux's Swifts, like many aerial insectivores, are currently experiencing population declines, and a mass mortality event at a spring migratory roost on Vancouver Island, British Columbia, Canada, resulted in the death of over 1,000 individuals, representing some 2% of the British Columbia population. From these individuals, we examined variation in three stable-isotopes (δ^{2}H, δ^{13}C, and δ^{15}N) from claw samples in order to determine whether spring migrants showed inherent isotopic similarity of the habitats they used on their Mexican and Central American wintering grounds. Our results indicated the presence of two to three broad isotopic clusters, suggesting that Vaux's Swifts migrating through Vancouver Island most likely originated from two or three over-wintering locales or habitat types. We found no evidence of sex- or morphology-based segregation, suggesting that these different groups likely share a similar over-wintering ecology and thus may be equally vulnerable to stochastic events or habitat loss on the wintering grounds. Our results highlight the need for more studies on the non-breeding season ecology and migratory connectivity of this species.

Keywords: Vaux's Swift, migratory connectivity, cluster analysis, stable isotope, conservation, roost

INTRODUCTION

Migratory birds move annually between breeding, wintering, and stopover sites that can be separated by hundreds or thousands of kilometers. As such, they face an array of both natural and human-mediated environmental challenges. Thus, for effective conservation, it is essential to take an annual cycle approach that addresses factors that influence populations throughout the year and across broad geographic scales (Webster 2002, Webster and Marra 2005). Stressors occurring during one phase of the annual cycle can have carry-over effects to subsequent phases of the annual cycle, affecting both individual- and population-level dynamics (Marra et al. 1998, Sillett et al. 2000, Reudink et al. 2009a). Population level carry-over effects can manifest through density-dependent population regulation (Fretwell 1972, Ratikainen et al. 2008), large-scale climatic cycles (Sillett et al. 2000, Wilson et al. 2011), and habitat loss (Norris 2005). Examples of carry-over effects influencing individual fitness include wintering habitat-mediated differences in reproductive success (Reudink et al. 2009a) and natal dispersal (Studds et al. 2008). Understanding patterns of migratory connectivity, defined as the extent to which individuals from the same wintering site migrate to the same breeding site, and vice versa (Marra et al. 2010), may be important for understanding species- or population-specific responses to anthropogenic disturbances (e.g., land-use change, agricultural development), as well as large-scale selective pressures such as climate change (Webster et al. 2002).

Migratory populations may be especially vulnerable to stochastic events when they display high migratory connectivity, particularly if population size is also small. For example, Kirtland's Warblers (*Setophaga kirtlandii*) exhibit extremely strong connectivity between breeding populations in Michigan, USA, and wintering populations in the Bahamas and Turks and Caicos (Bocetti et al. 2014). As a consequence, the species experiences pronounced carry-over effects whereby drier winters delay arrival and nest initiation on the breeding grounds, ultimately resulting in fewer offspring fledged (Rockwell et al. 2012). Intensive management of the species on the breeding grounds has led to population increases, but continued long-term recovery is likely dependent on active management in the Bahamas as well (Wunderle et al. 2010), especially as climate change is predicted to increase drought severity in the Caribbean (Rockwell et al. 2012).

For small migratory birds, making geographic connections for individuals and populations between different phases of their annual cycle can be exceedingly difficult. In recent years, information gleaned from traditional bird ringing has been greatly enhanced with the use of intrinsic markers such as genetic (e.g., Ruegg et al. 2014) and stable isotope (Hobson 2011) techniques, as well as combinations of these techniques (Chabot et al. 2012, Rundel et al. 2013). Stable isotopes of hydrogen (δ^2H) in particular have been instrumental in migratory connectivity research owing to geographically predictable patterns of isotopic variation that are reflected in animal tissue and thus provide markers of origin for where the tissue was grown (Hobson et al. 2012a). Such intrinsic markers have the advantage that birds need only be captured once, and they may be especially useful as a first step to understanding patterns of migratory connectivity in poorly-studied species or in remote areas (Hobson et al. 2014, Pekarsky et al. 2015). The advent of new extrinsic marker technologies such geolocators and GPS loggers has led to higher resolution information on migratory behaviour for certain species (Stutchbury et al. 2009). However, use of these extrinsic markers is complicated by the need to recover data loggers and remains impractical and often prohibitively expensive for large-scale studies (Arlt et al. 2013, Bridge et al. 2013, Hobson et al. 2014). In addition, extrinsic markers cannot be used to infer historic patterns of migratory connectivity. In contrast, intrinsic markers can be extremely useful for forensic studies, including dietary reconstruction (Nocera et al. 2012, Blight et al. 2014) and the inference of migratory connectivity from historic specimens (Hobson et al. 2010). Given the paucity of information on population- or region-specific migratory connectivity for many species of conservation concern, there is a pressing need to apply intrinsic and/or extrinsic marker approaches to evaluate factors limiting populations throughout their annual cycle (Hobson et al. 2014).

Like many other species of aerial insectivore, breeding populations of the Vaux's Swift (*Chaetura vauxii*) have undergone significant declines (Nebel et al. 2010). Recent estimates from the North American Breeding Bird Survey suggest a population decline of -2.2%/year in Canada since the 1970s (Environment Canada 2014). Vaux's Swifts breed throughout western North America, but exhibit an unusual roosting strategy during migratory journeys between their western North American breeding grounds and their southern Mexican and Central American wintering grounds (Bull and Collins 2007). During migration, groups of Vaux's Swifts roost communally in tree cavities or, more commonly, in abandoned industrial chimneys, where they can remain for several days to weeks (Bull and Collins 2007). Migratory roosts may contain hundreds to thousands of individuals. During winter, Vaux's Swifts also appear to occupy large roost sites (Bull and Collins 2007). However, little is known about migratory connectivity for Vaux's Swift and it remains unknown whether swifts from particular wintering areas migrate north together and stop at the same roost sites (strong migratory connectivity), or whether migratory roost sites are comprised of individuals arriving from multiple locations across their winter range (weaker migratory connectivity). The degree of connectivity can have important implications for conservation (e.g. Sheehy et al. 2011), and if large concentrations of individuals winter together and move together throughout migration, they may be particularly vulnerable to stochastic events. In this study, we used intrinsic markers to study putative migratory connectivity in migrating Vaux's Swifts. We sampled claws (grown during the over-wintering period) from a large group of communally-roosting Vaux's Swifts that perished due to accidental causes at a spring migratory roost site on Vancouver Island, British Columbia, Canada. We used cluster analyses based on multiple stable isotope (δ^2H, δ^{13}C, and δ^{15}N) markers to examine support for the existence of a single versus multiple wintering origins in this population of swifts.

METHODS

Field Methods

On May 9, 2012, a mass mortality event occurred at a migratory roost in Cumberland, British Columbia, Canada, used annually by Vaux's Swifts during spring migration. Approximately 1,350 Vaux's Swifts perished from suffocation after being trapped in the roost. That single mortality event represented a loss of between 1.5-2.7 % of published population size estimates (Summers and Gebauer 1995, Partners in Flight Science Committee 2013) for Vaux's Swifts in British Columbia. From those individuals, we randomly sub-sampled 98 individuals for analysis. From each individual we measured wing length, tail length, and tail pin length, and we sexed each individual via laparoscopy. We then sampled 2mm (0.30 to 0.40 mg) of tissue from the tip of the central claw, ensuring we avoided the quick, of each foot from each individual for isotope analysis. Though claws grow continuously and, due to their conical growth pattern, incorporate some material grown at slightly different times, analysis of the claw tip should accurately reflect the isotopic environment over an extended time period (weeks to months) preceding sampling (Mazerolle and Hobson 2005, Hahn et al. 2012). For instance, based on changes in δ^2H after arrival on the breeding grounds, Fraser et al. (2008) estimated isotopic values in claws of

Parulids may reflect the non-breeding area for 3-7 weeks after the birds arrive on the breeding grounds. Hahn et al. (2014) also provide empirical evidence demonstrating that in Palearctic-African migratory passerines the distal claw tip should reflect isotopic environments over a few months prior to sampling, with typical claw growth rates of 0.03-0.05 mm d^{-1}. Though data on migration timing of Vaux's Swifts are sparse, major flight passages occur during mid-April to late May in California (Small 1994), with birds arriving in Oregon from late April to late May (Bull and Collins 2007 and references therein). Thus, claws sampled from birds that perished during spring migration on May 9 should reflect wintering conditions, as has been demonstrated with passerines captured both on migration (Bearhop et al. 2004) and upon arrival on the breeding grounds (Reudink et al. 2009a,b).

Stable Isotope Analysis

We analyzed the stable isotope ratios of three naturally-occurring elements that are incorporated predictably into an animal's diet. First, we analyzed δ^2H which has been used extensively in studies of avian migration and connectivity because it is linked to both latitude and elevation, with ^2H being relatively less abundant at more northern latitudes and higher elevations (Hobson 2011). Second, we used $\delta^{13}C$, which varies in animal tissues with habitat, where individuals occupying habitats with a higher proportion of C4 plants or under higher water stress exhibit less negative $\delta^{13}C$ signatures (Lajtha and Marshall 1994, Cerling et al. 1997; Still and Powell 2010). Finally, we used $\delta^{15}N$, where ^{15}N is preferentially incorporated into the tissues of consumers and thus biomagnifies with increasing trophic levels, leading to an increase in $\delta^{15}N$ signatures (Post 2002, Poupin et al. 2011). In addition, $\delta^{15}N$ may reflect the relative aridity of a biome, with $\delta^{15}N$ being negatively correlated with rainfall and positively correlated with temperature (Sealy et al. 1987, Craine et al. 2009). All stable isotope analyses were conducted at the Smithsonian

Institution OUSS/MCI Stable Isotope Mass Spectrometry Facility in Suitland, Maryland, USA. Claws were washed in a 2:1 chloroform:methanol solution, then air-dried and allowed to acclimate to lab atmospheric conditions in a fume hood for 72 hours prior to sample preparation. Samples were pyrolyzed in a Thermo TC/EA elemental analyzer (Thermo Scientific, Waltham, Massachusetts, USA) at 1,350 °C and analyzed using a Thermo Delta V Advantage isotope ratio mass spectrometer. For stable hydrogen isotope analysis, we ran four calibrated standards for every 10 samples, including the hydrogen standard International Atomic Energy Agency CH-7 and three additional standards (KHS: -54.1 ± 2.3‰, CBS: -197.3 ± 2.0‰, Spectrum keratin: -121.6 ± 2.5‰). Non-exchangeable δ^2H values were corrected to keratin standards following Wassenaar and Hobson (2003), and were repeatable to within 3 ± 2‰ (mean ± SD, n = 10). For stable carbon and nitrogen analysis, we ran two in-house standards (acetanalide and urea) for every 10 samples. Stable isotope values are expressed in parts per thousand (‰) deviations from international standards VSMOW (hydrogen), PDB (carbon), and air (nitrogen) by the following equation:

$dX = \{[(R_{unknown}-R_{standard})-1] \times 1000\}$,

where X is the isotope ratio of interest (δ^2H, $\delta^{13}C$, or $\delta^{15}N$) and R is the corresponding ratio (^2H:^1H, ^{13}C:^{12}C or ^{15}N:^{14}N). Carbon and Nitrogen samples were repeatable to within ± 0.2 ‰ based on repeated measurements of standards.

Statistical Analysis

There are many different methods available for determining the optimal number of clusters in a dataset. For this reason, we made use of the package 'NbClust' in program R (R Development Core Team, Vienna, Austria), which provides the user with results from 30 indices aimed at determining the number of clusters into which the data should be split (Charrad et al. 2014). Clustering validity indices combine information such as intra and inter-cluster variation, geometric or statistical properties of the data, and dissimilarity or similarity measurements. For a detailed description of all 30 indices, see Charrad et al. (2014). Two widely-used clustering algorithms are available in the NbClust package: *k*-means and hierarchical agglomerative clustering. In *k*-means clustering, observations are assigned to initial cluster centers, which are then iteratively updated until the cluster centers no longer change and the within-cluster sum of squares is minimized (MacQueen 1967). In hierarchical agglomerative clustering, each observation begins in its own cluster, and pairs of clusters are joined based on a distance measure and an agglomeration criterion (Székely and Rizzo 2005). Because of the differences in these clustering approaches and a lack of *a priori* reasons to hypothesize a set number of clusters in our data, we used both clustering algorithms. We used a Euclidean distance (square distance between two vectors; Seber 1984) in both clustering algorithms and examined the relative support for division of the data into 2-10 clusters based on the three isotopic markers (δ^{2}H, δ^{13}C, and δ^{15}N). For hierarchical agglomerative clustering, we used the Ward agglomeration method (Ward 1963), which minimizes the total within-cluster variance. We also repeated the above procedures after removing six multivariate outliers, assessed using the robust Mahalanobis distance (Varmuza and Filzmoser 2009).

After the optimal number of clusters was identified by each clustering approach, we examined the partitioning of observations (individual birds) into clusters. We performed multivariate analysis of variance (MANOVA) to verify significant differences among the selected clusters in δ^{2}H, δ^{13}C, and $\delta\ ^{15}$N ratios. We then used a Pearson's chi square test to examine whether the proportion of claws from male and female birds differed among clusters, and we used a t test or analysis of variance (ANOVA) to examine whether wing length or tail length varied among clusters.

Finally, we used a model-based hierarchical clustering procedure within the package 'mclust' in R (Fraley et al. 2012) to determine whether the optimal number(s) of clusters selected by the above procedures was preferred over no clustering at all. Mclust uses an expectation-maximization (EM) algorithm to estimate the finite mixture models that correspond to different numbers of clusters, and uses the Bayesian information criterion (BIC) to select the best number of clusters. Importantly, a single cluster can also be considered. We evaluated the support for 1-10 clusters.

The δ^2H, δ^{13}C, and δ^{15}N isotope ratios were standardized prior to analysis by subtracting by their mean and dividing by their standard deviation. Standardization is recommended in cluster analysis when variables are on different scales to minimize the effects outliers and so cluster formation is not overly influenced by variables with greater absolute variation (Milligan and Cooper 1988). Although isotope ratios represent the ratio of heavy to light isotopes in a sample divided by the ratio of a standard, and are always expressed in the same units (‰), differences in the natural abundances and fractionation of heavy and light isotopes among elements lead to differences in the magnitude of the range of typical δ values (Fry 2006). Thus, standardization was useful in giving equal weight to variation among the three isotope ratios. All analyses were performed in R version 3.1.1 and means are shown as mean ± SD.

RESULTS

Stable isotopic values (δ^2H, δ^{13}C, and δ^{15}N) were available from claws of 98 individual swifts. All three elements showed a considerable variation in all three isotopes (Supplementary Material

Table S1). Mean isotope ratios were -37.9 ± 12.4 ‰ for δ^2H, -21.6 ± 0.6 ‰ for δ^{13}C, and 7.3 ± 0.9 ‰ for δ^{15}N. Correlations among isotope ratios were r = -0.09 (p = 0.37) for δ^2H and δ^{13}C, r = -0.14 (p = 0.17) for δ^2H and δ^{15}N, and r = 0.53 (p < 0.001) for δ^{13}C and δ^{15}N. Of those 98 individuals with isotope data, we were unable to determine sex for one individual. There was a nearly-even number of males and females in our sample (n = 47 females and 50 males). Males and females did not differ in δ^2H (t_{95} = -0.02, p = 0.98), δ^{13}C (t_{95} = -0.98, p = 0.33), or δ^{15}N (t_{95} = -0.18, p = 0.86). Mean wing length was 113.5 ± 2.8 mm and did not differ between males and females (males: 113.4 ± 2.7, females: 113.7 ± 2.8; t_{95} = -0.46, p = 0.65). Wing length was not correlated with δ^2H (r = -0.06, p = 0.59), δ^{13}C (r = 0.16, p = 0.12), or δ^{15}N (r = 0.16, p = 0.12). Mean tail length was 36.2 ± 2.3 mm and did not differ between males and females (males: 35.8 ± 1.8, females: 36.5 ± 2.7; t_{95} = -1.53, p = 0.13). Tail length was not correlated with δ^2H (r = 0.00, p = 0.99) or δ^{13}C (r = -0.07, p = 0.49), but it was negatively correlated with δ^{15}N (r = -0.23, p = -0.02).

When using the full dataset, 11 out of 30 clustering validity indices (37%) proposed two as the optimal number of clusters in the *k*-means clustering procedure. The next highest-ranked numbers of clusters were three and four, which received support from six (20%) and five (17%) clustering indices, respectively. Three was deemed the optimal number of clusters by the majority (10 or 33%) of indices in the hierarchical agglomerative procedure, but a large proportion of indices (9 or 30%) also supported two clusters. The only alternative clustering that received support was a 10 cluster solution with support from five (17%) indices. Results of the *k*-means clustering procedure were similar when outliers were removed, with two deemed the optimal number of clusters by the majority (9 or 30%) of indices. For the hierarchical agglomerative procedure, two was identified as the optimal number (8 or 27% of indices), with three receiving the second-most support (5 or 17% of indices). Overall, these results suggest that the isotope data grouped most naturally into two or three clusters, and thus we used the full dataset for the remainder of the analyses. Although the selection of the optimal number of clusters was not unanimous among indices, similar results are obtained from simulated datasets even when distinct non-overlapping clusters are used (Milligan & Cooper 1985, Charrad et al. 2014). Furthermore, according to the BIC, the model-based clustering procedure suggested that the optimal number of clusters was two (Supplementary Material Table S2).

We examined the classification of individual claws into two or three clusters using the results from *k*-means clustering or hierarchical agglomerative clustering, respectively. The clusters were significantly different from one another for all three isotope ratios simultaneously when partitioned into two (MANOVA $F_{3,93} = 52.5$, $p < 0.001$) or three clusters ($F_{3,93} = 36.4$, $p < 0.001$). The two cluster solution resulted from a split between swifts with higher $\delta^{15}N$ and $\delta^{13}C$ ratios and those with lower $\delta^{15}N$ and $\delta^{13}C$ ratios, with little influence of $\delta^{2}H$ on clustering (Table 1). The three cluster solution suggested an additional division of the swifts with low $\delta^{15}N$ and $\delta^{13}C$ ratios into those with higher or lower $\delta^{2}H$ ratio (Figures 1, 2). There was no association between sex and cluster membership when the claws were grouped into two ($\chi^2_1 = 0$, $p = 1$) or three ($\chi^2_2 = 1.70$, $p = 0.42$) clusters. There was no association between wing length and cluster membership for two ($t_{96} = 0.62$, $p = 0.54$) or three (ANOVA $F_{2,95} = 0.70$, $p = 0.50$) clusters. Similarly, there was no association between tail length and cluster membership for two ($t_{96} = -0.48$, $p = 0.64$) or three (ANOVA $F_{2,95} = 1.60$, $p = 0.21$) clusters.

DISCUSSION

Patterns of migratory connectivity in long-distance migratory birds can have important implications for population dynamics, evolutionary processes, and effective conservation strategies (Webster and Marra 2005). Using multiple stable isotopes ($\delta^{13}C$, $\delta^{15}N$, and $\delta^{2}H$) from claw samples of Vaux's Swifts at a spring migratory roost, our goal was not to assign individuals to a particular over-wintering locality or localities, which would have been exceedingly difficult due to poor resolution in isoscapes of Mexico and Central America (Bowen et al. 2005) and our lack of known-origin tissues from these areas. Rather we asked whether Vaux's Swifts at a single migratory roost likely came from a single winter location, which would indicate stronger migratory connectivity, or multiple winter locations, which would indicate weaker migratory connectivity. We found evidence in support of two or possibly three broad isotopic clusters on the over-wintering grounds from the single migratory roost on Vancouver Island, British Columbia, Canada. While alternative explanations for our observations are possible (see below), we tentatively interpret our findings as likely demonstrating differences in over-wintering locations used by Vaux's Swifts in our sample.

Understanding patterns of migratory connectivity in small, long-distance migratory birds has been exceedingly challenging, but the use of stable isotope analysis over the past two decades has revolutionized our ability to establish patterns of connectivity (Hobson 2005). Generally, this approach relies on the assignment of tissues to spatially explicit isoscapes generated from precipitation isoscapes (Bowen and West 2008) or known-origin individuals sampled across their range (e.g., Hobson et al. 2009c, Hobson et al. 2012b). Recently, these methods have been enhanced through the use of Bayesian statistical methods and GIS-based models of precipitation isoscapes, as well as additional information such as band recoveries, to improve assignments by creating probabilistic regions of origin (Hobson et al. 2009a, b, Van Wilgenburg and Hobson 2011). However, most studies focus on assigning breeding origins to winter-captured individuals, rather than vice versa, because breeding ground isoscapes, particularly in North America, are better delineated than winter ground isoscapes (Bowen et al. 2005). This poses a problem for assigning winter origins to individuals sampled within breeding or migratory populations, such as in our study. Furthermore, the particular tissue chosen for sampling must be reflective of the area of origin that is of interest to the researcher. This again poses difficulties for assigning winter origins when many North American bird species, including Vaux's Swifts, moult their feathers on or near the breeding grounds (Pyle 1997; but see for e.g. Kelly et al. 2008, Quinlan and Green 2011). Thus, in our study, we chose to sample claws because they reflect conditions several weeks prior to sampling, a time during which the birds should have been present on the wintering grounds (Bearhop 2004, Reudink et al. 2009a,b).

Despite the relatively high variation in all three isotopic signatures (δ^2H range: 66 ‰; δ^{15}N range: 5.8 ‰; δ^{13}C range: 3.5 ‰; Supplementary Material Table S1), our analysis provided support for Vaux's Swift claws fitting into two or three isotopically distinct clusters (Figure 2), with strongest support for two clusters when outliers were removed. When partitioned into two clusters, the claws separated based on those with higher δ^{13}C and δ^{15}N and those with lower δ^{13}C and δ^{15}N. Variation in δ^{13}C and δ^{15}N most likely relate to broad-scale habitat differences, as food-web δ^{13}C signatures are strongly related to the distribution of C3 and C4 vegetation (Lajtha and Marshall 1994), and food-web δ^{15}N signatures are associated with soil exposure and climate (Nadelhoffer and Fry 1994). While we cannot exclude the possibility that birds were separated in part on the basis of dietary shifts and niche specialization, broad-scale differences in δ^{13}C, δ^{15}N, and δ^2H appear to be consistent with Vaux's Swift using habitats that follow climatic/moisture gradients. Regardless of the clustering algorithm, birds associated with cluster 2 were enriched in ^{13}C by ~0.9 ‰ over individuals from cluster 1. Simultaneously, birds associated with cluster 2 were also enriched in ^{15}N by ~ 1.6 ‰ compared to birds from cluster 1 (Table 1). Assuming $\delta^{13}C$ follows similar gradients to those reported elsewhere (e.g. Marra et al. 1998), these data would appear to suggest that birds in cluster 2 originated from hotter and drier habitats than birds in cluster 1.

The agglomerative clustering approach suggested that birds could be further partitioned into a third cluster on the basis of splitting claws that had both low $\delta^{13}C$ and $\delta^{15}N$ values based on those with higher $\delta^{2}H$ (cluster 3) and versus lower $\delta^{2}H$ values (cluster 1, see Supplementary Material Table S1). Latitudinal and altitudinal gradients in $\delta^{2}H$ are well-established and among the strongest of isotopic gradients (Hobson 2005), and our results thus suggest that individuals that had low $\delta^{13}C$ and $\delta^{15}N$ were likely further segregated along an latitudinal or possibly an altitudinal gradient. Although a species-specific $\delta^{2}H$ claw tissue isoscape is lacking for the Vaux's Swift over-wintering range, a $\delta^{2}H$ feather isoscape developed for House Sparrows (*Passer domesticus*) in Mexico showed more positive $\delta^{2}H$ signatures in the northeast of the country and more negative signatures in the south and high altitude regions (Hobson et al. 2009c). Overall, our results indicate that individuals from the migratory population of swifts on Vancouver Island may have originated from a small number of different latitudes and/or habitats.

Breeding populations that are more strongly connected to specific wintering areas, particularly those experiencing degradation, may be most vulnerable to population declines (Dolman and Sutherland 1995, Webster and Marra 2005), although a recent study found that the strength of connectivity in breeding populations of Barn Swallows (*Hirundo rustica*) in North America was unrelated to their population trend (García-Pérez and Hobson 2014). Our data suggest that further research to estimate the strength of connectivity across breeding populations of Vaux's Swifts is now warranted, and might help explain patterns of decline if extrinsic markers could be deployed along a gradient of declining versus increasing populations (*sensu* Fraser et al. 2012 and Hobson et al. 2015).

We also asked whether swifts showed evidence of segregation based on sex or morphology during the over-winter period, as has been observed in several other passerines, such as Hooded Warblers (*Setophaga citrine*; Morton 1990), Prairie Warblers (*Setophaga discolor*; Latta and Faaborg 2001), and American redstarts, that segregate based on habitat, and White-throated Sparrows (*Zonotrichia albicollis*; Mazerolle and Hobson 2007) and Hermit Thrushes (*Catharus guttatus*; Stouffer and Dwyer 2003), that segregate based on latitude. For example, American Redstarts (*Setophaga ruticilla*) exhibit marked age- and sex-biased habitat segregation, with older males disproportionately inhabiting high-quality mangrove forests (Parish and Sherry 1994, Marra 2000) – a characteristic also easily discerned via stable isotope analysis of muscle (Marra et al. 1998), blood (Norris et al. 2004), and claw samples (Reudink et al. 2009a,b). Furthermore, larger females were more likely to inhabit high-quality mangrove territories due to dominance-mediated habitat segregation (Marra 2000). Latitudinal segregation, on the other hand, may be related to trade-offs between body size or wing size/shape and migration distance (García Peiró 2003; Mazerolle and Hobson 2007). Our results suggest no evidence for sex- or morphology-based segregation, at least when considering wing and tail length, and indeed there was no evidence of differences in wing size between males and females, suggesting that male and female Vaux's Swifts likely share a similar over-wintering ecology and thus may be equally vulnerable to stochastic events or habitat loss.

The utility of triple-isotope isoscapes for assigning origins to South American and African wintering birds has recently been demonstrated (Hobson et al. 2012b, García-Pérez and Hobson 2014), and our results indicate that the approach has potential for identifying Mexican and Central-American wintering clusters as well. Taken together, our results highlight the need for additional research on the over-wintering ecology and behaviour of this poorly-studied long-distance migrant and the need for calibrating tissue specific isoscapes to better assign Nearctic-Neotropical migratory birds to wintering localities. Our data suggest that birds migrating through Vancouver Island likely over-winter in two to three isotopically similar regions or habitat types, and these predictions should now be validated. Wintering ground sampling would facilitate nominal assignment (*sensu* Wunder 2012) to two or three spatially delineated geographic regions, or alternatively might facilitate spatially explicit multivariate assignment approaches (e.g., García-Pérez and Hobson 2014, Veen et al. 2014).

Recent work highlights the potential for keratinous tissues to vary isotopically with diet (Fraser et al. 2011, Voigt et al. 2013, Voigt et al. *in press*, Soto et al. 2013) and microhabitat (Fraser et al. 2011). Thus, while we interpret our results primarily in the context of migratory connectivity, we cannot preclude the possibility that multivariate differences in the isotopic composition of claws could stem from variation in microhabitat, behaviour, or dietary preferences among individuals. Given the high motility of Vaux's swift while foraging on the wing (Bull et al. 2007), it is plausible that our results could also stem from variation in degree of foraging over aquatic versus terrestrial or forested versus grassland or agricultural habitats. Like other aerial insectivores, Vaux's swifts forage broadly over forest canopy, meadows and open water (Bull and Collins 2007) and their diets likely reflect the composition of aerial plankton from major insect emergences from both the aquatic and terrestrial environments. Within their wintering grounds, these major habitat types should be relatively isotopically distinct owing to major isotopic differences in $\delta^{13}C$ and $\delta^{15}N$ between forest canopy (primarily using a C_3 phytosynthetic pathway) versus grasslands and pastures dominated by C_4 plants (Still and Powell 2010). It is important to note that the distribution of forest versus grassland, pasture and agricultural lands displays a great deal of geographic structure (Dixon et al. 2014, Ellis and Ramankutty 2008), and thus it is probable that isotopic differences due to differences in habitat selection would also covary with any spatial segregation. Regardless of whether the patterns we observed owe solely to differences in migratory connectivity or variation in local habitat selection, our results point to relatively limited variation in over-wintering niches of Vaux's swifts. Thus, a better understanding of the overwintering ecology Vaux's swifts is necessary to inform conservation. Wintering ground work on habitat use and diets of Vaux's swifts or performing multi-isotope assays on claws from birds fitted with miniaturized GPS tags (Hallworth and Marra 2015) would shed light on the mechanistic explanations for our observations.

Like most migratory aerial insectivores (Nebel et al. 2010), Vaux's Swifts are in decline. Given the unique roosting ecology of Vaux's Swifts, they are particularly susceptible to stochastic events as exemplified by this one mass mortality, which may represent a loss of ~1.5 – 2.7% of the British Columbia population of Vaux's Swifts in a single evening. This exceptional vulnerability coupled with increasing habitat loss and alteration in the tropics (Hansen et al. 2013) make the need for further research to determine patterns of migratory connectivity and identify critical winter and migratory roost sites crucial for future conservation planning.

Acknowledgements We thank S. Joly for assistance in data collection. Funding was provided by a Natural Sciences and Engineering Research Council of Canada (NSERC) Discovery Grant and a TRU Internal Research Fund Award to M.W.R. and a National Science Foundation (NSF) Grant to P.P.M. Birds were collected under a Canadian salvage permit (BC-SA-0049-12).

LITERATURE CITED

Arlt, D., M. Low, and T. Part (2013). Effect of Geolocators on Migration and Subsequent Breeding Performance of a Long-Distance Passerine Migrant. PLOS ONE 8:e82316.

Bearhop, S., G. M. Hilton, S. C. Votier, and S. Waldron (2004). Stable isotope ratios indicate that body condition in migrating passerines is influenced by winter habitat. Proceedings of the Royal Society B: Biological Sciences 271:S215–S218.

Blight, L. K., K. A. Hobson, T. Kurt Kyser, and P. Arcese (2014). Changing gull diet in a changing world: a 150-year stable isotope ($\delta^{13}C$, $\delta^{15}N$) record from feathers collected in the Pacific Northwest of North America. Global Change Biology 21:1497-1507.

Bocetti, C. I., M.D. Deahn and H. F. Mayfield (2014). Kirtland's Warbler (*Setophaga kirtlandii*), The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology; Retrieved from the Birds of North America Online: http://bna.birds.cornell.edu/bna/species/019

Bowen, G. J., L. I. Wassenaar, and K. A. Hobson (2005). Global application of stable hydrogen and oxygen isotopes to wildlife forensics. Oecologia 143:337-348.

Bowen, G.J., and J.B. West (2008). Isotope landscapes for terrestrial migration research. *In*: Tracking animal migration with stable isotopes. Eds: K.A. Hobson and L.I. Wassenaar. Academic Press, Amsterdam.

Bridge, E. S., J. F. Kelly, A. Contina, R. M. Gabrielson, R. B. MacCurdy, and D. W. Winkler (2013). Advances in tracking small migratory birds: a technical review of light-level geolocation. Journal of Field Ornithology 84:121-137.

Bull, E. L., and C. T. Collins (2007). Vaux's Swift (*Chaetura vauxi*), The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology; Retrieved from the Birds of North America Online: http://bna.birds.cornell.edu/bna/species/077

Cerling, T. E., J. M. Harris, B. J. MacFadden, M. G. Leaky, J. Quade, V. Eisenmann, and J. R. Ehrlinger (1997). Global vegetation change through the Miocene/Pliocene boundary. Nature 389:153-158.

Chabot, A. A., K. A. Hobson, S. L. Van Wilgenburg, G. J. McQuat, and S. C. Lougheed (2012). Advances in Linking Wintering Migrant Birds to Their Breeding-Ground Origins Using Combined Analyses of Genetic and Stable Isotope Markers. PLOS ONE 7:e43627.

Charrad, M., N. Ghazzali, V. Boiteau, and A. Niknafs (2014). NbClust: an R package for determining the relevant number of clusters in a data set. Journal of Statistical Software 61:1-36.

Craine, J. M., A. J. Elmore, M. P. M. Aidar, M. Bustamante, T. E. Dawson, E. A. Hobbie, A. Kahmen, M. C. Mack, K. K. McLauchlan, A. Michelsen, G. B. Nardoto, L. H. Pardo, J. Peñuelas, P. B. Reich, E. A. G. Schuur, W. D. Stock, P. H. Templer, R. A. Virginia, J. M. Welker, and I. J. Wright (2009). Global patterns of foliar nitrogen isotopes and their relationships with climate, mycorrhizal fungi, foliar nutrient concentrations, and nitrogen availability. New Phytologist 183:980–992.

Dixon, A. P., D. Faber-Langendoen, C. Josse, J. Morrison, and C. J. Loucks (2014). Distribution mapping of world grassland types. Journal of Biogeography 41: 2003-2019.

Dolman, P. M., and W. J. Sutherland (1995). The response of bird populations to habitat loss. Ibis 137:S38–S46.

Ellis, E. C., and N. Ramankutty (2008). Putting people in the map: anthropogenic biomes of the world. Frontiers in Ecology and the Environment 6: 439–447.

Environment Canada (2014). North American Breeding Bird Survey - Canadian Trends Website, Data-version 2012. Environment Canada, Gatineau, Quebec.

Fraley, C., A. E. Raftery, T. B. Murphy, and L. Scrucca (2012). mclust Version 4 for R: Normal Mixture Modeling for Model-Based Clustering, Classification, and Density Estimation. Technical Report No. 597, Department of Statistics, University of Washington.

Fraser, K. C., T. K. Kyser, R. J. Robertson, and L. M. Ratcliffe (2008). Season patterns in hydrogen isotopes of claws from breeding wood-warblers (Parulidae): utility for estimating migratory origins. Avian Conservation and Ecology 3:2.

Fraser, K. C., E. A. McKinnon, A. W. Diamond, and L. Chavarría (2011). The influence of microhabitat, moisture and diet on stable-hydrogen isotope variation in a Neotropical avian food web. Journal of Tropical Ecology 27: 563-572.

Fraser, K. C., B. J. Stutchbury, C. Silverio, P. M. Kramer, J. Barrow, D. Newstead, N. Mickle, B. F. Cousens, J. C. Lee, D. M. Morrison, T. Shaheen, P. Mammenga, K. Applegate, and J. Tautin (2012). Continent-wide tracking to determine migratory connectivity and tropical habitat associations of a declining aerial insectivore. Proceedings of the Royal Society of London B: Biological Sciences, rspb20122207.

Fretwell, S. D. (1972). Populations in a seasonal environment. Princeton, New Jersey: Princeton University Press.

Fry, B. (2006). Stable Isotope Ecology. Springer, New York, NY.

García Peiró, I. (2003). Intraspecific variation in the wing shape of the long-distance migrant Reed Warbler *Acrocephalus scirpaceus*: effects of age and distance of migration. Ardeola 50:31-37.

García-Pérez, B., and K. A. Hobson (2014). A multi-isotope (δ^2H, δ^{13}C, δ^{15}N) approach to establishing migratory connectivity of Barn Swallow (*Hirundo rustica*). Ecosphere 5:art21

Hahn, S., B. J. Hoye, H. Korthals, and M. Klaasen (2012). From food to offspring down: tissue-specific discrimination and turn-over of stable isotopes in herbivorous waterbirds and other avian foraging guilds. PLOS One 7:e30242.

Hallworth, M. T., and P. P. Marra. 2015. Miniaturized GPS tags identify non-breeding territories of a small breeding migratory songbird. Scientific Reports 5: art 11069. doi:10.1038/srep11069

Hahn, S., D. Dimitrov, S. Rehse, E. Yohannes, and L. Jenni (2014). Avian claw morphometry and growth determine the temporal pattern of archived stable isotopes. Journal of Avian Biology 45:202-207.

Hansen, M. C., P. V. Potapov, R. Moore, M. Hancher, S. A. Turubanova, A. Tyukavina, D. Thau, S. V. Stehman, S. J. Goetz, T. R. Loveland, A. Kommareddy, A. Egorov, L. Chini, C. O. Justice, and J. R. G. Townshend (2013). High-resolution global maps of 21st-century forest cover change. Science 342:850-853.

Hobson, K. A. (2005). Stable isotopes and the determination of avian migratory connectivity and seasonal interactions. Auk 122:1037-1048.

Hobson, K. A. (2011). Isotopic ornithology: a perspective. J. Ornithol. 152 (Suppl 1):S49–S66.

Hobson, K. A., M. B. Wunder, S. L. Van Wilgenburg, R. G. Clark, & L. I. Wassenaar (2009a). A method for investigating population declines of migratory birds using stable isotopes: origins of harvested lesser scaup in North America. PLOS ONE e:7915.

Hobson, K. A., H. Lormée, S. L. Van Wilgenburg, L. I. Wassenaar, J. M. Boutin (2009b). Stable isotopes (δD) delineate the origins and migratory connectivity of harvested animals: the case of European woodpigeons. Journal of Applied Ecology 46:572-581.

Hobson, K. A., S. L. Van Wilgenburg, K. Larson, & L. I. Wassenaar (2009c). A feather hydrogen isoscape for Mexico. Journal of Geochemical Exploration 102:167-174.

Hobson, K. A., R. Greenberg, S. L. Van Wilgenburg, and C. Mettke-Hoffman (2010). Migratory connectivity in the Rusty Blackbird: isotopic evidence from feathers of historical and contemporary specimens. Condor 112:778-788.

Hobson, K. A., S. L. Van Wilgenburg, L. I. Wassenaar, K. Larson (2012a). Linking hydrogen (δ^2H) isotopes in feathers and precipitation: sources of variance and consequences for assignment to isoscapes. PLOS ONE 7:e35137.

Hobson, K. A., S. L. Van Wilgenburg, L. I. Wassenaar, R. L. Powell, C. J. Still, and J. M. Craine (2012b). A multi-isotope (δ^{13}C, δ^{15}N, δ^2H) feather isoscape to assign Afrotropical migrant birds to origins. Ecosphere 3:art44

Hobson, K. A., S. L. Van Wilgenburg, J. D. Toms, J. Faaborg, C. Rengifo, Y. Aubry, and R. B. Aguilar (2014). Connecting breeding and wintering grounds of Neotropical migrant songbirds using stable hydrogen isotopes: A call for an isotopic atlas of migratory connectivity. Journal of Field Ornithology 85:237-257.

Hobson, K. A., K. J. Kardynal, S. L. Van Wilgenburg, G. Albrecht, A. Salvadori, M. D. Cadman, F. Liechti, and J. D. Fox. (2015). A continent-wide migratory divide in North American breeding Barn Swallows (*Hirundo rustica*). PLOS One 10: e0129340.

Kelly, J. F., M. J. Johnson, S. Langridge, and M. Whitfield (2008). Efficacy of stable isotope ratios in assigning endangered migrants to breeding and wintering sites. Ecological Applications 18:568-576.

Latta, S. C. and J. Faaborg (2001). Winter site fidelity of Prairie Warblers in the Dominican Republic. Condor 103:455-468.

Lajtha, K. & Marshall, J. D. (1994). Sources of variation in isotopic composition in plants. In Stable isotopes in ecological and environmental sciences (eds K. Lathja & J. D. Marshall), pp. 1–21. London, UK: Oxford University Press.

MacQueen, J. B. (1967). Some methods for classication and analysis of multivariate observations. In LML Cam, J Neyman (eds.), Proceedings of the Fifth Berkeley Symposium on Mathematical Statistics and Probability 1:281-297.

Marra, P. P., K. A. Hobson, and R. T. Holmes (1998). Linking winter and summer events in a migratory bird by using stable-carbon isotopes. Science 282:1884–1886.

Marra, P. (2000). The role of behavioral dominance in structuring patterns of habitat occupancy in a migrant bird during the nonbreeding season. Behavioral Ecology 11:299–308.

Marra, P. P., C. E. Studds, and M. Webster (2010) Migratory connectivity. In: Breed MD, Moore J (eds) Encyclopedia of animal behavior. Academic, Oxford, pp 455–461.

Mazerolle, D. F., and K. A. Hobson (2005). Estimating origins of short-distance migrant songbirds in North America: contrasting inferences from hydrogen isotope measurements of feathers, claws, and blood. Condor 107:280–288.

Mazerolle, D. F., and K. A. Hobson (2007). Patterns of differential migration in white-throated sparrows evaluated with isotopic measurements of feathers. Canadian Journal of Zoology 85:413-420.

Milligan, G. W., and M. C. Cooper (1985). An examination of procedures for determining the number of clusters in a data set. Psychometrika 50:159-179.

Milligan, G. W., and M. C. Cooper (1988). A study of standardization of variables in cluster analysis. Journal of Classification 5:181-204.

Morton, E.S. (1990). The evolution of habitat segregation by sex in the Hooded Warbler: experiments on proximate causation. American Naturalist 135: 319-333.

Nadelhoffer, K. J., and B. Fry (1994). Nitrogen isotope studies in forest ecosystems. Pages 22–44 in Stable Isotopes in Ecology and Environmental Science (K. Lajtha and R. H. Michener, Eds.). Blackwell Scientific Publications, Oxford.

Nebel, S., A. Mills, J. D. McCracken, and P. D. Taylor (2010). Declines of Aerial Insectivores in North America Follow a Geographic Gradient. Avian Conservation and Ecology 5:1.

Nocera, J.J., J.M. Blais, D.V. Beresford, L.K. Finity, C. Grooms, L.E., Kimpe, K. Kyser, N. Michelutti, M.W. Reudink, and J.P. Smol. 2012. Historical pesticide applications coincided with an altered diet of aerially foraging insectivorous chimney swifts. Proceedings of the Royal Society B: Biological Sciences 278:3114-3120.

Norris, D. R. (2005). Carry-over effects and habitat quality in migratory populations. Oikos, 109:178-186.

Norris, D. R., P. P. Marra, T. K. Kyser, T. W. Sherry, and L. M. Ratcliffe (2004). Tropical winter habitat limits reproductive success on the temperate breeding grounds in a migratory bird. Proceedings of the Royal Society B: Biological Sciences 271:59–64.

Parrish, J., and T. Sherry (1994). Sexual Habitat Segregation by American Redstarts Wintering in Jamaica - Importance of Resource Seasonality. Auk 111:38–49.

Partners in Flight Science Committee (2013). Population Estimates Database, version 2013. Available at http://rmbo.org/pifpopestimates. Accessed on April 26, 2015.

Pekarsky, S., A. Angert, B. Haese, M. Werner, K. A. Hobson, and R. Nathan (2015). Enriching the isotopic toolbox for migratory connectivity analysis: a new approach for migratory species breeding in remote or unexplored areas. Diversity and Distributions 21:416-427.

Post, D. M. (2002). Using stable isotopes to estimate trophic position: models, methods, and assumptions. Ecology 83:703–718.

Poupin, N., C. Bos, F. Mariotti, J.-F. Huneau, D. Tome, and H. Fouillet (2011). The nature of the dietary protein impacts the tissue-to-diet ^{15}N discrimination factors in laboratory rats. PlOS One 6:e28046.

Pyle, P. 1997. Identification guide to North American Birds, Part I. Slate Creek Press, Bolinas, CA.

Quinlan, S. P., and D. J. Green (2011). Variation in deuterium (δD) signatures of yellow warbler Dendroica petechial feathers grown on breeding and wintering grounds. Journal of Ornithology 152:93-101.

R Core Team (2013). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/

Ratikainen, I. I., J. A. Gill, T. G. Gunnarsson, W. J. Sutherland, and H. Kokko (2008). When density dependence is not instantaneous: theoretical developments and management implications. Ecology Letters 11:184-198.

Reudink, M. W., P. P. Marra, T. K. Kyser, P. T. Boag, K. M. Langin, and L. M. Ratcliffe (2009a). Non-breeding season events influence sexual selection in a long-distance migratory bird. Proceedings of the Royal Society B. 276:1619-1626.

Reudink, M. W., C. E. Studds, T. K. Kyser, P. P. Marra, and L. M. Ratcliffe (2009). Plumage brightness predicts non-breeding season territory quality in a long-distance migratory songbird, the American redstart *Setophaga ruticilla*. Journal of Avian Biology 40:34-41.

Rockwell, S. M., C. I. Bocetti, and P. P. Marra (2012). Carry-Over Effects of Winter Climate on Spring Arrival Date and Reproductive Success in an Endangered Migratory Bird, Kirtland's Warbler (*Setophaga kirtlandii*). Auk 129:744-752.

Ruegg, K., E. C. Anderson, K. L. Paxton, V. Apkenas, S. Lao, R. B. Siegel, D. F. DeSante, F. Moore, and T. B. Smith (2014). Mapping migration in a songbird using high-resolution genetic markers. Molecular Ecology 23:5726-5736.

Rundel, C.W., M. B. Wunder, A. H. Alvarado, K. C. Ruegg, R. Harrigan, A. Schuh, J. F. Kelly, R. B. Siegel, D. F. DeSante, T. B. Smith, and J. Novembre (2013). Novel statistical methods for integrating genetic and stable isotope data to infer individual-level migratory connectivity. Molecular Ecology 22:4163-4176.

Sealy, J. C., N. J. van der Merwe, J. A. Lee-Thorp, J. L. Lanham (1987). Nitrogen isotopic ecology in southern Africa: implications for environmental and dietary tracing. Geochimica et Cosmochimica Acta 51:2707–2717.

Seber GAF (1984). Multivariate Observations. John Wiley & Sons, New York.

Sheehy, J., C. M. Taylor, and D. R. Norris (2011). The importance of stopover habitat for developing effective conservation strategies for migratory animals. Journal of Ornithology 152: 161-168.

Sillett, T. S., R.T. Holmes, and T.W. Sherry (2000). Impacts of a global climate cycle on the population dynamics of a migratory songbird. Science 288:2040-2042.

Small, A. (1994). California Birds: status and distribution. Ibis Publishing, Temecula, California.

Soto, D. X., L. I. Wassenaar, and K. A. Hobson (2013). Stable hydrogen and oxygen isotopes in aquatic food webs are tracers of diet and provenance. Functional Ecology 27: 535-543.

Still, C. J., and R. L. Powell. (2010). Continental-scale distributions of vegetation stable carbon isotope ratios. In: West, J. B., G. J. Bowen, T. E. Dawson, and K. P. Tu (eds). Isoscapes: understanding movements, pattern and process on Earth through isotope mapping. Springer, New York, pp.179–194.

Stouffer, P. C., G. M. Dwyer (2003). Sex-biased winter distribution and timing of migration of Hermit Thrushes (*Catharus guttatus*) in Eastern North America. Auk 120:836-847.

Studds, C. E., T. K. Kyser, and P. P. Marra (2008). Natal dispersal driven by environmental conditions interacting across the annual cycle of a migratory songbird. Proceedings of the National Academy of Sciences USA 105:2929-2933.

Stutchbury, B. J., S. A. Tarof, T. Done, E. Gow, P. M. Kramer, J. Tautin, J. W. Fox, V. Afanasyev (2009). Tracking long-distance songbird migration by using geolocators. Science 323:896–896

Summers, K., and M. Gebauer (1995). Status of Vaux's Swift in British Columbia. Wildlife Working Report WR-67. Ministry of Environment, Victoria, BC.

Székely, G. J., M. L. Rizzo (2005). Hierarchical clustering via joint between-within distances: extending Ward's minimum variance method. Journal of Classification 22:151-183.

Van Wilgenburg, S. L., and K. A. Hobson (2011). Combining stable-isotope (δD) and band recovery data to improve probabilistic assignment of migratory birds to origin. Ecological Applications 21:1340-1351.

Varmuza, K., and P. Filzmoser (2009). Introduction to Multivariate Statistical Analysis in Chemometrics. CRC Press, Boca Raton, FL.

Veen, T., M. B. Hjernquist, S. L. Van Wilgenburg, K. A. Hobson, E. Folmer, L. Font, and M. Klassen (2014). Determining the winter grounds of hybrid flycatchers using a multi–isotope (δ^2H, δ^{13}C, δ^{15}N) assignment approach. PLOS 9:e98075.

Voigt, C. C., D. Lehmann, and S. Greif (*in press*). Stable isotope ratios of hydrogen separate mammals of aquatic and terrestrial food webs. Methods in Ecology and Evolution. DOI: 10.1111/2041-210X.12414

Voigt, C. C., K. Schneeberger, and A. Luckner (2013). Ecological and dietary correlates of stable hydrogen isotope ratios in fur and body water of syntopic tropical bats. Ecology 94:346–355.

Ward, J. H. (1963). Hierarchical grouping to optimize an objective function. Journal of the American Statistical Association 58:236-244.

Wassenaar, L. I., and K. A. Hobson (2000). Stable-carbon and hydrogen isotope ratios reveal breeding origins of red-winged blackbirds. Ecological Applications 10:911–916.

Webster, M. S., P. P. Marra, S. M. Haig, S. Bensch, and R. T. Holmes (2002). Links between worlds: unraveling migratory connectivity. Trends in Ecology and Evolution 17:76-83.

Webster, M. S., and P. P. Marra (2005). The importance of understanding migratory connectivity. In: Birds of Two Worlds: The Ecology and Evolution of Temperate-

Tropical Migration Systems. R. Greenberg and P. P. Marra (Eds). Johns Hopkins University. Pp 199-209.

Wilson, S., S. L. LaDeau, A. P. Tøttrup, and P. P. Marra (2011). Range-wide effects of breeding- and nonbreeding-season climate on the abundance of a Neotropical migrant songbird. Ecology 92:1789–1798.

Wunder, M. B. (2012). Determining geographic patterns of migration and dispersal using stable isotopes in keratins. Journal of Mammalogy 9:360-367.

Wunderle, J. M., D. Currie Jr., E. H. Helmer, D. N. Ewert, J. D. White, T. S. Ruzycki, B. Parresol, and C. Kwit (2010). Kirtland's warblers in anthropogenically disturbed early-successional habitats on Eleuthera, the Bahamas. Condor 112:123-137.

TABLE 1. Mean (±SD) stable isotope ratios of Vaux's Swift claws partitioned into two or three clusters based on *k*-means or hierarchical agglomerative clustering, respectively. Cluster numbers (1-3) corresponds to those shown in Figure 1.

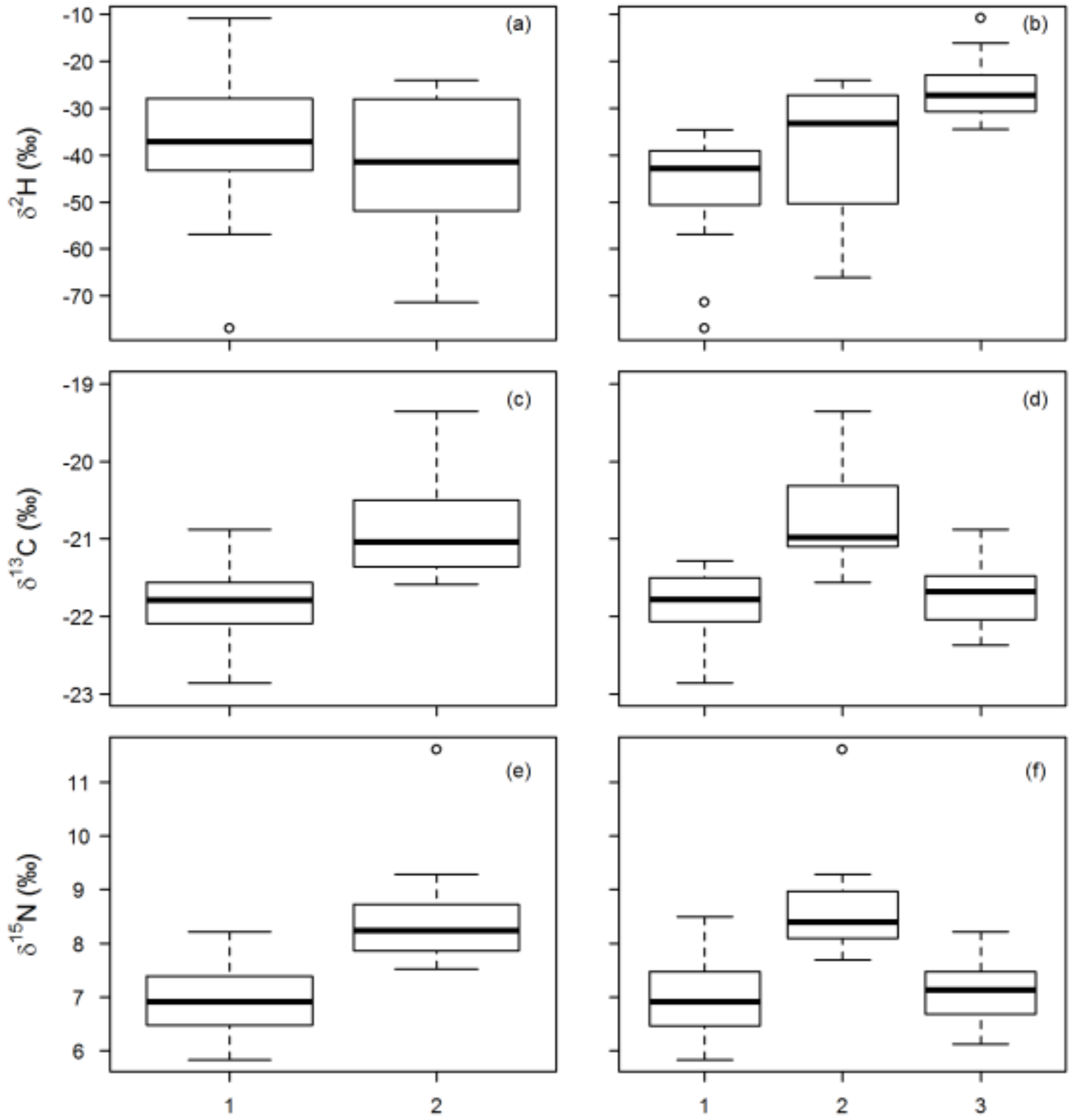
	Two clusters			Three clusters		
Cluster no.	δ^{2}H	δ^{13}C	δ^{15}N	δ^{2}H	δ^{13}C	δ^{15}N
1	-36.5 (11.6)	-21.8 (0.4)	7.0 (0.5)	-45.6 (8.9)	-21.8 (0.4)	7.0 (0.6)
2	-42.0 (13.8)	-20.9 (0.6)	8.4 (0.9)	-38.7 (12.3)	-20.7 (0.6)	8.6 (0.9)
3	-	-	-	-26.3 (5.6)	-21.7 (0.4)	7.1 (0.5)

FIGURE LEGENDS

FIGURE 1. Variation in Vaux's Swift claw stable isotopes of hydrogen (δ^{2}H: a, b), carbon (δ^{13}C: c, d), and nitrogen (δ^{15}N: e, f) when claws were clustered into 2 (a, c, e) or 3 (b, d, f) clusters. Shown are the median, interquartile range (IQR), and outliers (> 1.5 x IQR).

FIGURE 2. Clustering of stable isotope ratios of Vaux's Swift claws into two (left panel) or three (right panel) clusters based on *k*-means or hierarchical agglomerative clustering, respectively.

FIGURE 1



(a)
(b)
(c)
(d)
(e)
(f)
δ^2H (‰)
-10
-20
-30
-40
-50
-60
-70
δ^{13}C (‰)
-19
-20
-21
-22
-23
δ^{15}N (‰)
11
10
9
8
7
6
1
2
1
2
3

FIGURE 2