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SHORT COMMUNICATIONS

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ESTIMATING NATAL ORIGINS OF MIGRATORY JUVENILE GOLDEN EAGLES USING STABLE HYDROGEN ISOTOPES

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The difficulty in determining the geographic origins of migratory birds and identifying their source populations has limited the understanding of the ecology of many North American species (Viverette et al. 1996, Meehan et al. 2001, Hobson et al. 2009). In North America, Golden Eagles (*Aquila chrysaetos*) are widespread; they breed predominately in the West from northern Alaska to central Mexico, and occupy a wide range of habitats from arctic tundra to deserts (Kochert et al. 2002). Most raptors, including Golden Eagles, typically occur in low breeding densities, have large home ranges, and nest in remote areas, making it difficult to assess populations using traditional methods such as Breeding Bird Surveys and Christmas Bird Counts (Fuller and Mosher 1981, Titus and Fuller 1990, Rosenfield et al. 1991, Meehan et al. 2001). Consequently, annual migration counts along ridges during fall migration are the traditional means of monitoring migratory Golden Eagle populations. Analysis of long-term migration count data suggests a downward trend in observed numbers for this species. This has raised concern about the status of migratory Golden Eagles in western North America (Kochert and Steenhof 2002, Hoffman and Smith 2005, Smith et al. 2008, Tilly 2008). However, interpreting migration count data can be difficult (McCaffery and McIntyre 2005) and requires knowledge of the source populations, which is incomplete for a majority of migration count sites (Hoffman and Smith 2003). By identifying the geographic origins of the avian migrants, it may be possible to relate migration count trends to a particular regional population of interest (Viverette et al. 1996, Meehan et al. 2001).

Recently, there has been a call for more information on Golden Eagles in the West (Pagel et al. 2010). Downward trends in Golden Eagle numbers at migration count sites may be the result of factors on breeding or wintering grounds, or both, across western North America (Kochert and Steenhof 2002, Hoffman and Smith 2005, Smith et al. 2008, Pagel et al. 2010). However, one of the biggest obstacles in accurately interpreting raptor migration counts in relation to population status is determining origins and destinations of migrants, and this poses difficulties when addressing conservation issues and developing management strategies (Kochert and Steenhof 2002, Hoffman and Smith 2005).

Stable hydrogen isotope analysis utilizing the weighed growing season average precipitation (δ^2H_p) can help reveal natal origins of hatch-year (hereafter referred to as juvenile) Golden Eagles captured at annual migration count sites or on wintering grounds. In addition, it can be used to estimate the natal origins of eagles found as mortalities (e.g., on wind farms) and determine whether those individuals were from northern migratory or nonmigratory populations. The analysis utilizes predictable continent-wide distributions of deuterium abundance occurring in rainfall. These geographically specific ratios of deuterium are then transferred from precipitation through the food chain to upper trophic level consumers such as birds and mammals (Hobson and Wassenaar 1997, 1999; Hobson 2005; Wassenaar 2008). A number of studies have used stable hydrogen isotope ratios to estimate the breeding latitudes of Neotropical migratory songbirds (e.g., Kelly et al. 2002, Rubenstein et al. 2002, Wassenaar and Hobson 2000a), and more recently an increasing number of raptor species (Meehan et al. 2001, Lott et al. 2003, Meehan et al. 2003, Smith et al. 2003, DeLong et al. 2005, Smith and Dufty 2005, Hobson et al. 2009, Ruyck et al. 2013, Wittenberg et al. 2013). Although relatively new, stable hydrogen isotope analysis has proven to be successful and revolutionized our understanding of

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migratory ecology for many avian species (Meehan et al. 2001, Kelly et al. 2005, Wunder et al. 2009, VanWilgenburg and Hobson 2011, but see Smith et al. 2009).

We used stable hydrogen isotope analysis to address three questions about Golden Eagle migratory ecology: (1) can patterns in natal origins of juvenile Golden Eagles captured during fall migration be identified? (2) do migrant juvenile Golden Eagles exhibit an association between latitudinal origin and passage date? and (3) are there any sex-related differences concerning latitudinal origins and passage dates?

STUDY AREA

The Rocky Mountain Front (RMF) stretches from Alaska, U.S.A. to Mexico and holds the largest known concentration of migratory Golden Eagles known on earth (Tilly 2008, Sherrington 2009). The interface of the Rocky Mountains and Great Plains creates a recognizable migration corridor from Alaska to Mexico. In Montana, it is characterized by north-south ridges often leading to distinct geographic bottlenecks where migrating raptors concentrate. We conducted our research at three sites along the RMF in Montana during fall migration: (1) Rogers Pass (47°4.52′N, 112°22.73′W) on the Continental Divide near Lincoln, Montana; (2) Nora Ridge (47°1.14′N, 112°24.28′W) 6.5 km southwest of Rogers Pass and; (3) Grassy Mountain (46°18.74′N, 111°7.13′W) located in the Big Belt Mountains approximately 128 km southeast of Rogers Pass. All three sites are located in the Helena National Forest and are representative of the RMF flyway in Montana.

METHODS

We captured juvenile Golden Eagles using bow-nets with Rock Pigeons (*Columba livia*) as lures (Bloom 1987). We began trapping each year on approximately 15 September and continued daily, through 31 October, 2004–2007. Daily observations and trapping efforts began at approximately 0930 H and finished at approximately 1700 H, weather permitting. We identified juvenile eagles in the hand by plumage (Bloom and Clark 2001). We determined sex via morphometric measurements (Bortolotti 1984, Edwards and Kochert 1986). We validated sex determinations based on morphometric measurements for a subset of our sample via DNA analysis (Zoogen Services, Inco., Davis, California U.S.A.).

We collected 3-cm feather clippings from the crural (upper leg) region of each Golden Eagle captured because of feather abundance in the crural region and ease of sampling. We banded captured individuals with a unique United States Geological Survey (U.S.G.S.) aluminum leg band and fitted them with alphanumerically coded vinyl wing-tags. We sent feather samples to the Colorado Plateau Stable Isotope Laboratory for stable isotope analysis. To limit variability, only the distal end of the non-rachis vane material was analyzed (Wassenaar and Hobson 2006). Using standardized lab protocols, feathers were first cleaned with chloroform-methanol solution and then air-dried to enable exchangeable hydrogen to equilibrate with ambient laboratory moisture, equilibrating for a minimum of 7–10 d prior to analysis (Chamberlain et al. 1997, Wassenaar and Hobson 2000b, 2003). Feathers were then packed in silver capsules and pyrolized using a Themo Finnigan TC/EA elemental analyzer (Thermo Electron Corporation, Boston, Massachusetts, U.S.A.) and sample hydrogen content was analyzed using a Delta Plus XL mass spectrometer (Thermo Electron Corporation, Boston, Massachusetts, U.S.A.) in continuous flow mode at 1400°C. The non-exchangeable hydrogen isotope ratios were calculated via comparative equilibration using keratin standards after Wassenaar and Hobson (2003). Values of δ^2H_f were normalized on the VSMOW-SLAP scale using three powdered keratin standards obtained from Environment Canada, Saskatoon, Saskatchewan: bowhead whale baleen (BWB: $\delta^2H = -108‰$), chicken feather standard (CFS: $\delta^2H = 147.5‰$), and cow hoof standard (CHS: $\delta^2H = -187‰$; L. Wassenaar pers. comm.). Analytical error of non-exchangeable hydrogen isotope values was ±4%.

Lott and Smith (2006) used over 250 known-origin raptor feathers to create a raptor-specific reference map of δ^2H_f for North America. Because there can be differences in δ^2H_f detection between labs or even within the same lab from different sample runs, we calibrated our samples using a smaller subset of known-origin samples used by Lott and Smith to make their map ($n = 22$). For an anecdotal comparison, we sampled feathers from two Golden Eagles that had been in captivity >10 yr in Montana and fed exclusively local fauna (K. Davis pers. comm.). Both individuals had isotope values of −114‰, which supported established isotope levels for Montana. We measured δ^2H_f at our lab (Northern Arizona State Isotope Laboratory) for 22 known-origin feathers that Lott and Smith used to create their reference map. We then used simple linear regression to relate the δ^2H_f measured at our lab to the δ^2H_f measured at the Lott and Smith lab. We used the regression equation to calibrate our samples from juvenile Golden Eagles. As suggested by Lott and Smith (2006), we incorporated error into the prediction by assessing an upper and lower range for the δ^2H_f values of ±8‰.

To map the estimated origin of the captured eagles, we grouped the calibrated δ^2H_f values into three classes for ease of display, and reported the percent of birds estimated to originate from each class. We plotted these three classes using data from the Lott and Smith (2006) raptor-specific isoscape reference map, to spatially interpret natal origins of migratory juvenile Golden Eagles. We tested whether juvenile Golden Eagles exhibited migratory timing patterns using simple linear regression with capture date as the independent variable and δ^2H_f as the dependent variable. Additionally, we tested for sex differences in δ^2H_f using a two sample *t*-test.

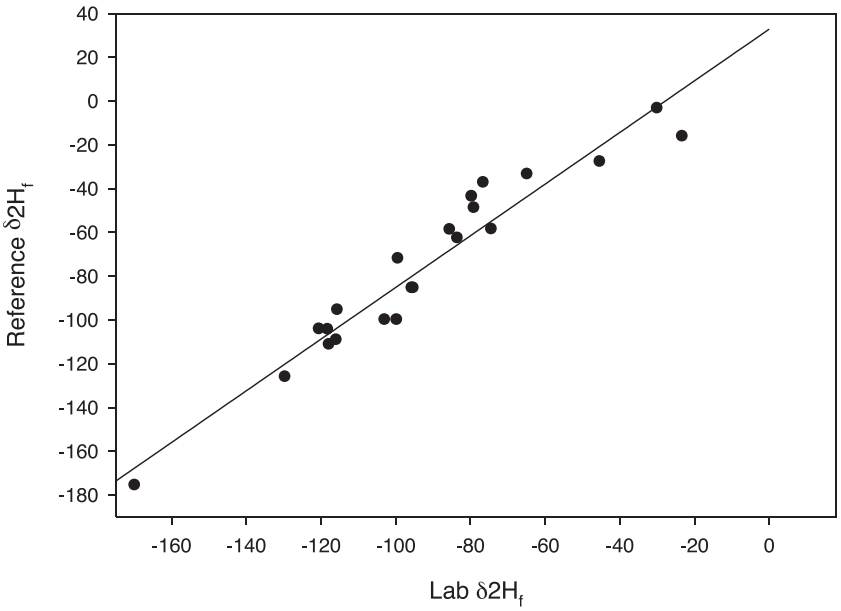

Figure 1. Relationship between the δ^2H_f measured in the known origin samples at our lab (Northern Arizona State Isotope Laboratory) to the δ^2H_f reported for the same sample Lott and Smith used to create the raptor specific reference map.

RESULTS AND DISCUSSION

We captured and collected feather samples from 50 juvenile Golden Eagles during fall migration. We sampled 12 eagles in 2004 and 13 in 2005 from our Rogers Pass study site, 5 in 2006 and 14 in 2007 from Nora Ridge, and 6 in 2007 from Grassy Mountain. We used a subsample of 25 eagles to verify our sex determinations from morphometrics via DNA tests and 100% of sex assignments were correct. We found that our analysis of the reference feather samples slightly underestimated δ^2H_f compared to Lott and Smith (2006) but still exhibited good fit with these data ($P < 0.0001$, $r^2 = 0.937$; Fig. 1). Consequently, we used the regression equation $\hat{y} = 32.92 + 1.18x$ to calibrate our results to the raptor-specific map of δ^2H_f where x is the measured δ^2H_f (‰) and $\hat{y}$ is the calibrated δ^2H_f (‰).

The mean predicted δ^2H_f for all juvenile Golden Eagle feather samples was −145.5‰ (range −103.3‰ to −176.4‰, SD = 16.4‰; Fig. 2). We incorporated error into the prediction by assessing an upper and lower range for the δ^2H_f values of ±8‰ (Fig. 3). Our results indicated 46% ($n = 23$) of migratory, juvenile Golden Eagles we sampled had predicted δ^2H_f values ≤ −150‰ with possible percentages ranging from 26% to 62% after accounting for error. Additionally, 20% ($n = 10$; range 14 to 24%) had predicted δ^2H_f values between −150‰ and −140‰. Thus, 66% had predicted natal areas located in the Yukon and western Northwest Territories, Canada, to eastern Alaska (Fig. 4). Based on the isoscape map, it is possible that some individuals may have come from the region of the eastern Northwest Territories and Nunavut provinces, Canada; however, it is unlikely given limited breeding records from that area (Kochert et al. 2002). Furthermore, concurrent satellite tracking studies of adult Golden Eagles captured at our study sites ($n = 18$) confirmed summer ranges were all west of central Northwest Territories (R. Domenech unpubl. data). We found no association between time of capture and calibrated δ^2H_f ($P = 0.2928$) or between sex and calibrated δ^2H_f ($P = 0.1349$) or sex and time of capture ($P = 0.4404$; Fig. 5).

Previously, researchers utilizing hydrogen stable isotope technology for animal migration studies relied on using a simple model of hydrogen stable isotopes in precipitation (δ^2H_p) that did not account for elevation, latitudinal and other sources of variation (Meehan et al. 2003, Lott and Smith 2006). Lott and Smith (2006) developed the raptor-specific deuterium map to allow for more accurate estimation of natal origins of raptors by incorporating both elevationally explicit growing-season precipitation data (Bowen et al. 2005) and an actual known-origin raptor feather base map. We found that plotting calibrated deuterium ratios on the Lott and Smith (2006) base map was useful for estimating natal origins of juvenile Golden Eagles migrating along the RMF in Montana. The results were further supported by several years of satellite tracking data of Golden Eagles using the RMF (R. Domenech unpubl. data).

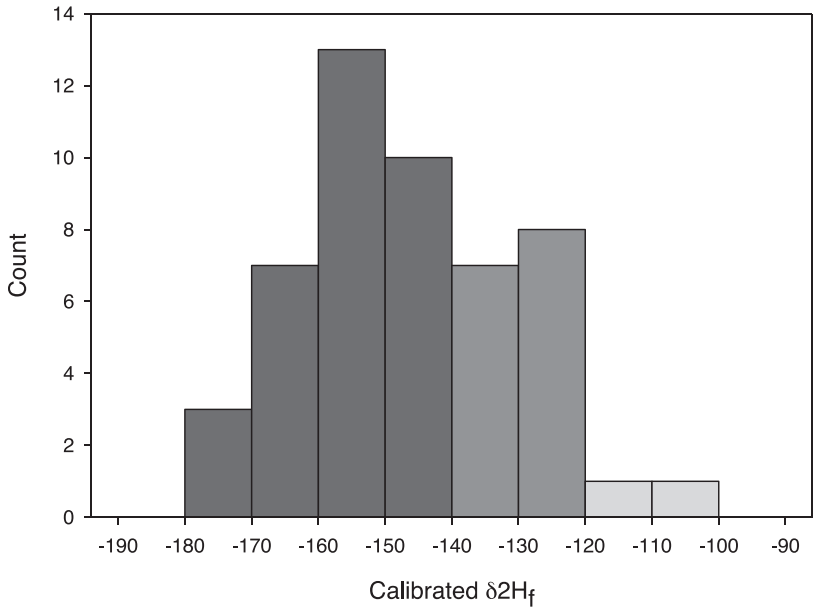

Figure 2. Distribution of δ^2H_f (‰) in juvenile Golden Eagles captured during migration along the Rocky Mountain Front in Montana (mean = –145.5‰, SD=16.4‰, n=50). The shades of the bins on the histogram indicate the predicted natal origin regions in Fig. 4.

We acknowledge there are some pitfalls to be considered when applying these techniques which include: the age of the study species, prey types, natal origins in proximity to large bodies of water (e.g., along ocean shorelines) and other variables (Smith et al. 2009, Ruyck et al. 2013, Whittenberg et al. 2013). Smith et al. (2009) report concerns with between-lab reproducibility of δ^2H_f. Although there was some variability between the two labs analyzing

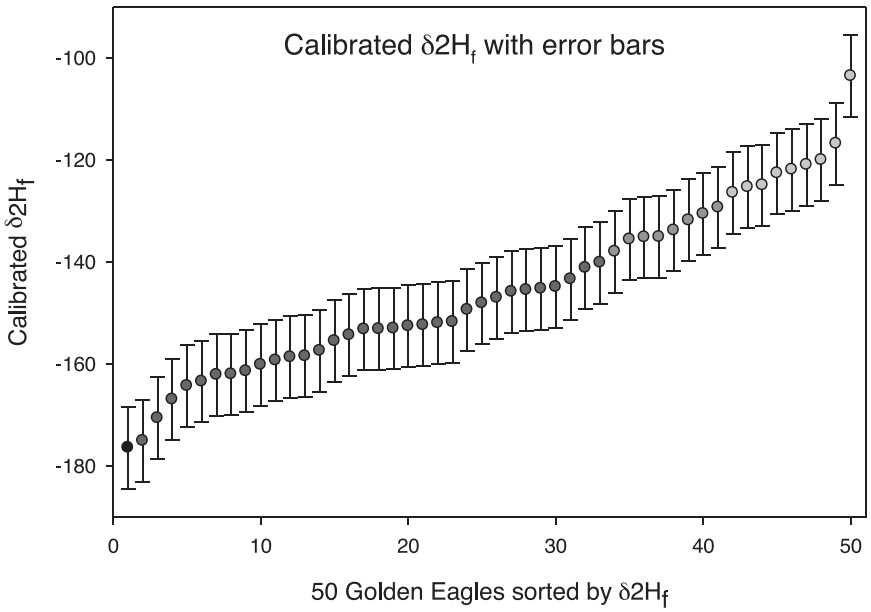

Figure 3. Predicted δ^2H_f values (‰) for the juvenile, migratory Golden Eagles sampled. We incorporated error into the prediction by assessing an upper and lower range for the δ^2H_f values of ±8‰.

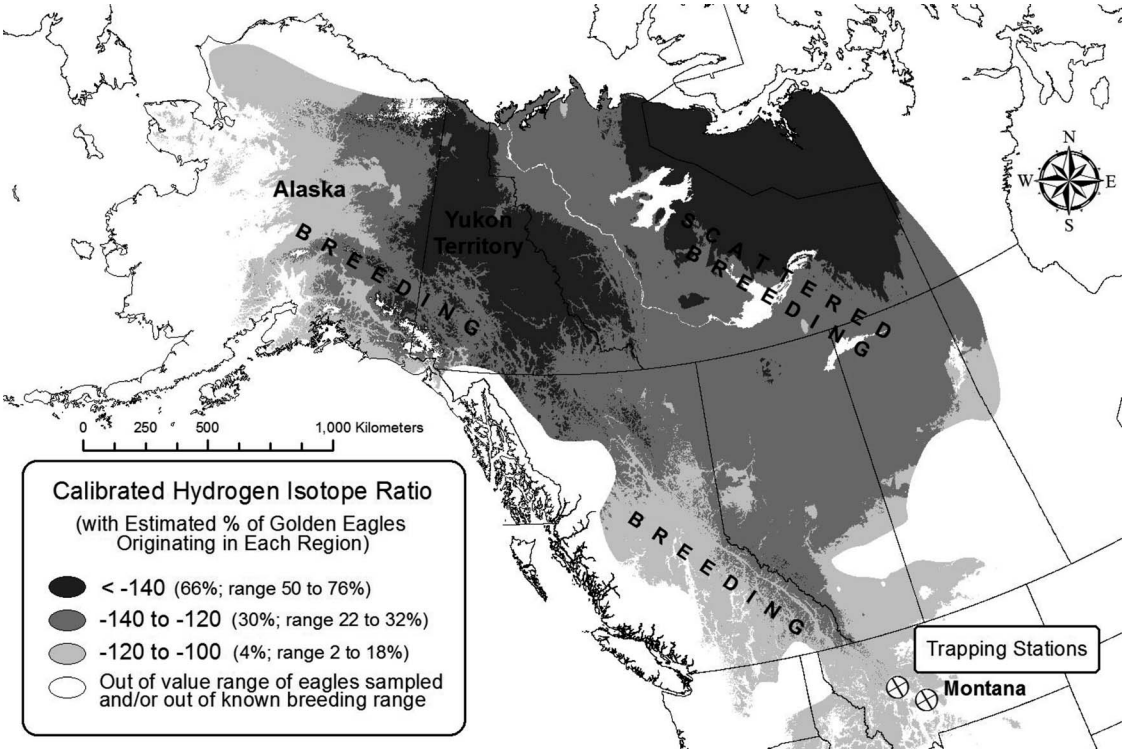

Figure 4. Raptor-specific deuterium map adapted from Lott and Smith (2006) indicating natal origin regions for juvenile, migratory Golden Eagles captured along the Rocky Mountain Front in Montana.

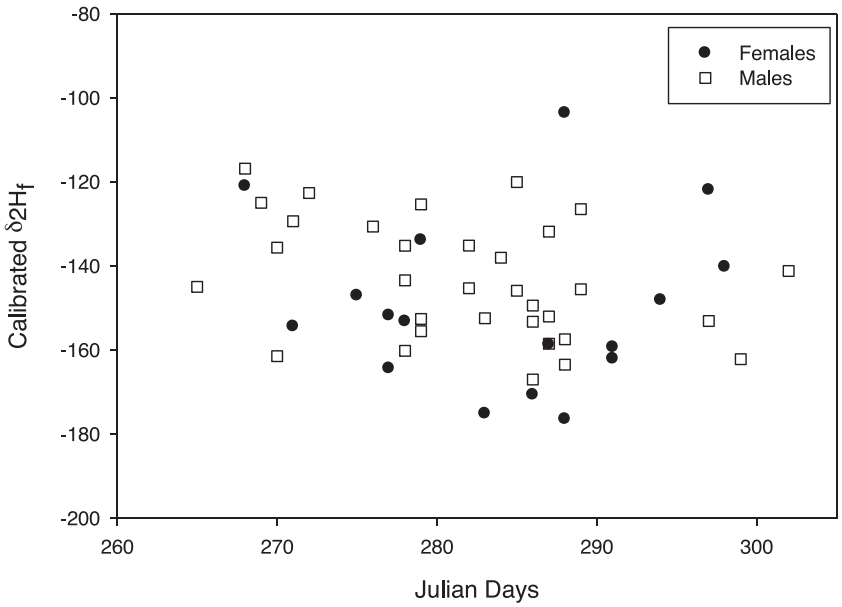

Figure 5. Relationship between time of arrival and calibrated δ^2H_f (‰) and gender and calibrated δ^2H_f. Females are represented by a dark circle, males are represented by an open square.

the same feathers, the values in our data range of more negative values (i.e., northern latitudes) showed minimal variability, compared to less negative values (i.e., southern latitudes; Fig. 1). This supports other data that show predicting natal origins is more problematic in southerly latitudes with more positive isotopic values (Smith et al. 2009). Finally, unlike some raptors, such as accipiters and falcons, which may feed largely on migratory species, Golden Eagle prey species are generally much more localized. For example, largely nonmigratory species such as jackrabbits (*Lepus* spp.), snowshoe hares (*Lepus americanus*), marmots (*Marmota* spp.), ground squirrels (*Spermophilopsis* spp.), and ungulates would have isotopic values that more accurately reflect the location of juvenile Golden Eagle origin. As such, we reason that this technique may have more utility for Golden Eagles than some other raptor species.

Utilizing stable isotope analysis can be an important tool for: (1) helping link Golden Eagle migration count data with population origin and status; and (2) identifying important breeding areas of migratory Golden Eagles. Approximately two-thirds of the juvenile Golden Eagles we sampled were likely to have natal origins in and around the Yukon and Northwest Territories, Canada, to east and northeast Alaska, suggesting this area may be an important breeding area for migratory Golden Eagles utilizing the RMF. This is corroborated by multiple breeding studies (McIntyre 1995, Young et al. 1995, McIntyre and Adams 1999, McIntyre 2002, 2008).

We encourage researchers to incorporate stable-hydrogen isotope analysis during fall migration, wintering, and mortality studies on Golden Eagles. Widespread use has potential to greatly increase our understanding about the natal origins of migratory Golden Eagles which will aid in better interpretation of trends in Golden Eagle migration count data and population status throughout North America. In addition, employing this technique on juvenile Golden Eagles found as mortalities (e.g., on wind farms) would yield an estimation of natal origin, as well as providing important insight into whether mortalities are from local (i.e., sedentary) or migratory (i.e., northern) populations.

ESTIMA DEL LUGAR DE NACIMIENTO DE INDIVIDUOS MIGRANTES JUVENILES DE *AQUILA CHRYSAETOS* MEDIANTE EL USO DE ISÓTOPOS DE HIDRÓGENO

RESUMEN.—Utilizamos análisis de isótopos estables de hidrógeno para estimar el origen natal de individuos juveniles de *Aquila chrysaetos* capturados durante la migración de otoño a lo largo de la cadena frontal de las Montañas Rocosas en Montana, EEUU. Recolectamos muestras de plumas de 50 individuos menores a un año (juveniles) en numerosos lugares de migración otoñal entre 2004-2007. Analizamos el radio de deuterio (δ^2H_f) en las plumas mostrando los resultados en partes por mil [‰]. Se utilizó un modelo de regresión lineal simple para calibrar nuestros cocientes de isótopos obtenidos de las águilas migrantes con un mapa base de deuterio específico para rapaces. Esto nos permitió realizar inferencias acerca del lugar de origen natal de individuos juveniles de *A. chrysaetos* capturados durante la migración otoñal. Nuestro análisis evidenció que el origen natal se extendió desde la Cadena Brooks en Alaska hasta el Norte de Montana. Sin embargo, el 66% (rango 50-76%) de los individuos que analizamos tuvo su origen probable en áreas ubicadas en Yukón y en los Territorios del Noroeste, Canadá, y en una pequeña porción del Este de Alaska (≤ -140 δ^2H_f). No observamos ninguna diferencia en las fechas de paso migratorio asociada al sexo o con la latitud del lugar de nacimiento. Nuestro estudio sugiere que el análisis de isótopos estables puede ser una herramienta útil para relacionar los censos migratorios y los datos de tendencias de *A. chrysaetos* con su estatus poblacional cuando se consideran múltiples lugares de censo en migración así como las áreas de invernada a lo largo y ancho de América del Norte.

[Traducción del equipo editorial]

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