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PAPER

ANTHROPOLOGY; CRIMINALISTICS

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Traveling There and Back Again: A Fingernail's Tale*,†,‡

ABSTRACT: This study focuses on the examination of stable isotope biomarkers in human fingernails of travelers who ventured away from Salt Lake City, UT and subsequently returned. Here, we describe three case studies examining: individual travelers (n = 4), paired travelers (n = 2), and a traveler (n = 1) who repeatedly left and returned. Our case studies examined the influences of dietary and drinking water inputs on fingernail stable isotope profiles for reconstructing travel histories. Stable isotope data were compared with theoretical and predictive models that laid the groundwork for anthropological and forensic geospatial reconstruction of travel histories. Drinking water inputs strongly influenced stable isotope profiles and the patterns observed were consistent with reported travel histories. Finally, observed fingernail stable isotope ratio data were consistent with modeled rates of stable isotope turnover and transition to expected values for the new environment. The results of our study further support the use of fingernail tissues for travel history reconstruction.

KEYWORDS: forensic science, forensic anthropology, stable isotope ratio analysis, travel history reconstruction, fingernail, keratin protein

Anthropological and forensic studies have primarily used stable isotope biomarkers to help reconstruct life history events of individuals or populations of individuals. The breadth of published literature is vast, looking at prehistoric, historic, and modern human populations to examine questions related to dietary inputs and mobility (1–6). These studies measured stable isotope ratios in human tissues ranging from bone, tooth enamel, and collagen (more predominant in ancient remains) to keratin tissues (hair and fingernails). Keratin tissues have mainly been used in modern human studies to examine movement and diets within a population, as hair can be viewed as a linear tape recorder of travel events (7).

Diet and travel history reconstruction using stable isotope biomarkers focus on two inputs: (1) dietary and (2) drinking water. As "you are what you eat" isotopically (8,9), the stable isotope measurement of body tissues allows for the indirect analysis of broad food groups that have been consumed. Stable carbon and nitrogen isotope ratios have been used to infer dietary patterns of human populations (10–14) based on the broad differences in C_3 and C_4 photosynthetic pathways of plants (15) and the trophic enrichment of consumers (16,17). Similar to our dietary inputs, "you are also what you drink," in that the oxygen and hydrogen isotopes in a person's diet reflects their direct and indirect water consumption through drinking and foods waters, which are tied to geographic locations (18–20).

Understanding the basis of how stable isotope inputs influence keratin stable isotope composition have helped create a theoretical framework and provided the physiological basis that allows us to predict and model stable isotope turnover in tissues, to understand how stable isotopes are incorporated into different tissues, and to better understand how variations in stable isotope abundances relate to geospatial patterns (18,21–23). Specifically, Cerling et al. (22) focused on using a reaction progress variable model to describe how different stable isotope pools influenced temporal variations in stable isotope ratios of different tissues. The reaction progress model also predicted how long it would take for the stable isotope ratios of tissues to turnover and equilibrate to a new input source (diet or location) (22). While O'Grady et al. (23) examined how particular sources of water (drinking, food, or recycled body water) are integrated into keratin tissues to create predictive models, Ehleringer et al. (18) and Bowen et al. (21) made the connection between the water sources (drinking water) and geospatial patterns. These theoretical relationships have allowed historic anthropologic and modern forensic disciplines to understand more about the diets and movements of ancient peoples and to assist law enforcement in the identification of human remains using stable isotope biomarkers.

Here, we highlight the use of stable isotope biomarkers in human fingernails to examine travel histories through case studies of Salt Lake City, UT residents who left, traveled, and returned. In particular, we were interested in both testing the theoretical models related to isotope turnover and to evaluate connections between tap water inputs and geospatial relationships in

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travelers. Here, we present the results from three types of travelers: (1) single individuals, (2) paired individuals, and (3) a repeated traveler using human fingernail clippings.

Materials and Methods

Participant Recruitment

In 2015, a total of six participants were recruited from a larger study of resident and nonresident populations in Salt Lake City, UT (24). Travelers are defined as Salt Lake City residents (i.e., resided consistently in the Salt Lake City region for a minimum of 9 months, all participants had been residents for 2+ years), who ventured away from Utah and then returned. Participants in this group included individuals who traveled to Ecuador, Panama, Indonesia, Australia/New Zealand, and India.

Fingernail clippings were collected for a period of 9–24 months. Demographic surveys (sex, age, travel history, and dietary questions) were also collected from participants throughout the study period to monitor changes in diet and additional travel when they returned to Salt Lake City. The University of Utah's Institutional Review Board approved this study (IRB protocol 00035524). All participants provided written consent to participate in the study and were free to choose which sample(s) and demographic information to provide. These individual choice decisions account for the variation in sample sizes for analyses presented in this study.

Fingernail Samples

Sample Collection—Fingernail samples were collected from traveler participants in the same manner as described in Mancuso and Ehleringer (24). In brief, participants collected their fingernail clippings for a period of 9–24 months, which were then stored in labeled coin envelopes. Participants were asked to record the date in which they clipped their fingernails and separate clippings from right versus left hands. This distinction was requested because there is evidence that fingernails on an individual's dominant hand grow faster (25,26). To maintain consistency among participants, we used fingernail clipping from the dominant hand for all analyses whenever possible. Participants were instructed to clip their fingernails as they normally would (dates between clippings averaged at 15 ± 6 days for all participants) and to collect all samples over the study period. Sample processing and isotope analyses were conducted at the Stable Isotope Ratio Facility for Environmental Research (SIRFER) at the University of Utah, Salt Lake City, UT.

Stable Isotope Analyses

All stable isotope analyses were analyzed at SIRFER (http://sirfer.utah.edu). Fingernail clippings were cleaned using methods similar to that described by O'Connell et al. (1). Briefly, fingernail clippings were cleaned of any visible surface dirt using deionized (DI) water. Afterward, samples were washed twice (for 60 and 30 min) in a 2:1 chloroform:methanol mixture to remove lipids and other surface contaminants. Lastly, they were rinsed with DI water for 20–40 min in an ultrasonic bath. Cleaned samples were dried for 24–48 h at 65°C in a convection oven.

For measurements of stable carbon and nitrogen isotope ratio values, fingernail samples were cut into 750 μg (±10%) pieces and placed into tin capsules for analyses. Fingernail samples and laboratory reference materials were analyzed using an elemental analyzer attached to an isotope ratio mass spectrometer (EA-IRMS, Thermo Scientific, Bremen, Germany) operated in continuous flow mode. The laboratory reference materials for carbon and nitrogen isotopes consisted of two glutamic acids and ground bovine muscle. All stable isotope ratios are reported using the standard δ-notation relative to an international standard (carbon isotopes: Vienna Pee Dee Belemnite (VPDB) and nitrogen isotopes: AIR) in units per mil (‰) using the following: $\delta X = (R_{sample}/R_{standard} - 1)*1{,}000$, where X is the isotope of interest, R_{sample} and $R_{standard}$ are the molar ratios of the heavy to the light isotopes (e.g., $^{13}C/^{12}C$) of the sample and international standard, respectively. The analytical precision (1σ), based on long-term measurements of internal laboratory reference materials for $\delta^{13}C$ and $\delta^{15}N$, was 0.1‰ for both stable isotopes.

Fingernail preparation and measurements of oxygen and nonexchangeable hydrogen were performed in a similar manner as described by Mancuso and Ehleringer (24). In brief, 150 μg (±10%) of samples were loaded in duplicate into silver capsules that had been treated to remove oxides. Samples for oxygen and hydrogen isotope analyses and associated reference and QA/QC keratin were equilibrated and stored under vacuum following established principle of identical treatment methods (27,28). A set of three laboratory keratin reference materials, for which the hydrogen isotope ratios of the nonexchangeable hydrogen fraction and total oxygen isotope ratios were known, were also loaded in a similar manner following the principle of identical treatment.

The oxygen and hydrogen isotope values of the fingernail clippings were measured using a temperature conversion elemental analyzer (TC/EA, Thermo Scientific, Bremen, Germany) attached to an isotope ratio mass spectrometer (IRMS, Delta Plus XL, Thermo Scientific, Bremen, Germany). All results for δ^2H data are expressed on a non-exchangeable hydrogen basis and both δ^2H and $\delta^{18}O$ are expressed in per mil (‰) units relative to the international standard Vienna Standard Mean Ocean Water (VSMOW). The analytical precision (1σ), based on long-term measurements of internal laboratory reference materials for $\delta^{18}O$ and δ^2H was 0.2‰ and 1.1‰, respectively.

Stable Isotope Turn Over and Reaction Progress Modeling of Oxygen Isotope Ratio Values

To examine the turnover and integration of a new water signal in the fingernail clippings of our traveler population, we used the 2-pool reaction progress variable model (short term, $t_{1/2}$ = 2 days and long term, $t_{1/2}$ = 30 days) as described by Cerling et al. (22). For most participants, the initial starting value (d_i) for the model was derived from all of the fingernail clippings from the resident mean population [$\delta^{18}O$: 8.9 ± 1.3‰, (24)] in Salt Lake City and the first equilibration value (d_e) reflected the calculated isotope value for the location that they traveled to. On their return to Salt Lake City, the return starting value (d_{ir}) reflected the modeled isotope value before they left and the return equilibrium value (d_{er}) was again taken from the Salt Lake City resident mean population.

To calculate the predicted precipitation oxygen isotope value for the travel location of our participants, we used the Online Isotopes in Precipitation Calculator (version, OIPC3.1, http://wateriso.utah.edu/waterisotopes/pages/data_access/oipc.html) to determine the mean annual precipitation for that location (29–31). The calculated annual precipitation stable isotope value served as a proxy for the isotope values of local drinking water, as local precipitation is often used for municipal tap waters

(19,30,32–34). The oxygen precipitation isotope value was applied to the human scalp hair prediction model to determine the oxygen isotope value (18,21). As the relationship between drinking water and fingernail and scalp hair keratin proteins are similar to each other (24), we used the hair model as a proxy for what the oxygen isotope ratio could be in fingernails from the traveled to location.

Fingernail development, growth rates, and isotope incorporation were under similar constraints that we have previously described (24). The same set of parameters that were specific to fingernail samples were applied to the traveler samples in the same manner [e.g., time "lag" for isotope incorporation of the traveled to location and the earliest time we could anticipate seeing the new isotope signature based on the average fingernail growth rates and nail matrix lengths (24–26,35)].

Statistics

Statistical significance and graphic output were generated using PRISM v 5.0 (GraphPad Software). Data were summarized as mean ± standard deviation (SD). Normality of distributions was assessed using the Shapiro–Wilks test. Comparisons between individual travelers were calculated using a Mann–Whitney test. The observed and predicted fingernail oxygen isotope ratios were analyzed using a linear regression analysis. Differences were statistically significant at the alpha level of 0.05.

Results/Discussion

There have been few longitudinal studies examining fingernail isotope ratios in traveler populations. While our case study population sizes may be limited, they reflect unique travel patterns and specifically examine isotope changes over a period of 9 months to a year. Our study focused on fingernail stable isotope profiles of individuals who were residents of Salt Lake City and who subsequently traveled to various locations outside of the United States before returning. We were interested in evaluating how dietary and drinking water inputs would relate to their travel history and allow us to look for patterns associated with their movements. In particular, we compared fingernail stable isotope ratio values to theoretical models that related stable isotope incorporation into human tissues and their relation to geospatial patterns. Here, we discuss the findings in three case studies looking at individual (n = 4), group (n = 2), and repeated travel (n = 1) histories.

All by Myself—Individual Travel Patterns

Participants and Travel History—Here, we describe the travel histories of individual residents of Salt Lake City, UT who traveled to various locations outside of the United States and returned. Individuals traveled to Panama (participant 4, from March to June 2015), Central India (participant 9, from mid-February to end April 2015), Indonesia (participant 15, September to December 2015), and Australia/New Zealand (participant 53, from mid-June to September 2015) before returning. Samples from each individual reflected the time they were away, their return to Salt Lake City, and the adjustment in fingernail stable isotope ratios back to that of the resident population values.

Dietary Isotopes—Surprisingly, individuals did not exhibit significant changes in their dietary isotope (carbon and nitrogen stable isotope ratios) profiles over the study period, nor were there changes during their time away from Salt Lake City (Fig. 1). The intra-individual variation among participants was low (0.2–0.3‰) and within the analytical precision of the measurement, and isotope values were consistent throughout the study period. Fingernail stable isotope values for participant 53, who went to Australia/New Zealand, did have greater fluctuations in their individual stable carbon and nitrogen isotope values throughout the study (mean values: −17.4 ± 1.0‰ and 9.5 ± 0.6‰). While there was no indication of dietary preference or eating changes during the study period, the changes in carbon and nitrogen isotope ratios for this individual could reflect increased consumption of marine and seafood protein sources, resulting in more enriched carbon and nitrogen stable isotope ratio values (17,36,37).

Inter-individual differences in carbon and nitrogen isotope ratios over the course of the study did suggest variations in individual diets. Nitrogen isotope ratios were similar among the four individuals (~9.4–9.5‰), which were consistent with their reported omnivore dietary preference classification. Carbon

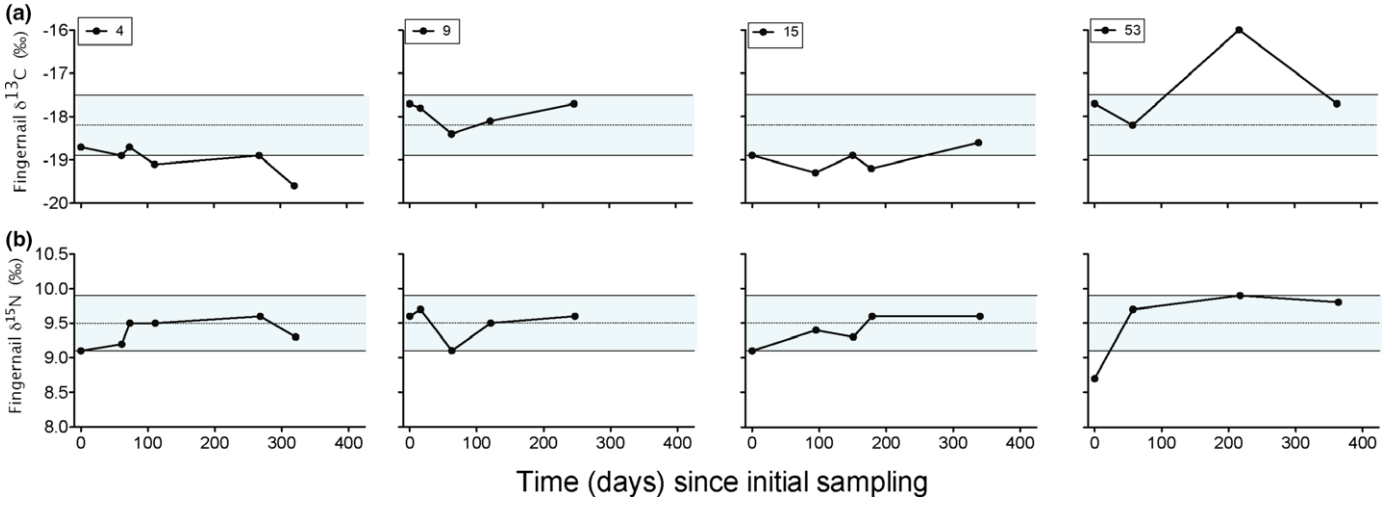

FIG. 1—*Fingernail (a) carbon and (b) nitrogen isotope ratios of travelers over time. Blue rectangle reflects the Salt Lake City, UT resident group mean ± SD (24). [Color figure can be viewed at wileyonlinelibrary.com]*

isotope ratios suggested differences among participants in the proportions of C_3 and C_4 food sources within their diets (−19.0‰ to −17.4‰). The differences in carbon isotope ratios could be influenced by the proportion of fast food and marine seafood that they regularly reported eating as part of their diet, which would result in more positive carbon isotope ratios (38–40). However, the exact explanation is unknown.

Drinking Water and Travel History Reconstruction—We analyzed the oxygen and hydrogen isotope ratios of the fingernail clippings of our participants over the study period and compared their profiles with that of the Salt Lake City resident isotope mean (24) to examine periods when they did or did not fall within the range of resident values. We have shown only the oxygen isotope values as the hydrogen isotope ratio results showed similar relationships (Fig. 2 and Figure S1). Here, we highlight the stable isotope incorporation in fingernails from participants 4 and 53, who traveled to Panama and Australia/New Zealand. Fingernail oxygen isotope ratios from both individuals reflect periods when fingernail clippings were developed in Salt Lake City and when they were away. Isotope profiles were consistent with their travel histories. In addition, when we compared observational data and modeled predictions, we saw that stable isotope observations for both individuals generally fit the modeled values well ($\delta^{18}O_{obs} = 1.15 \times \delta^{18}O_{pred} - 1.892‰$, $r^2 = 0.7463$, $p < 0.0001$); however, during the turnover period and convergence of the resident population mean the model occasionally overestimated the observed values (Fig. 3).

The samples from participant 15, displayed some curious patterns. This participant traveled to Indonesia and while there became ill with Dengue Fever and was hospitalized for approximately 1 week. During this time, intravenous fluids were provided to re-hydrate the individual. Observed fingernail oxygen isotope ratios were compared to the model predictions (Figure S2A). Here, we see an underestimation during the turnover period. It is possible that the incorporation of the Indonesian water-isotope signal was muted as the intravenous fluids altered the body water composition (23) and therefore influenced the keratin tissues of the individual. This explanation would account for the delayed the integration of the local water-isotope signature until the end of the travel period, where the oxygen isotope ratios agreed with the predicted values for keratin hair samples based on their precipitation values from that region (Figure S2A). While this medical event is unique in our study, it could mimic an important influence on isotope signatures of keratin tissues when asking questions related to modern human movement or the recreation of travel history of unidentified remains. Further research related to the impact of intravenous fluids on body water isotopes and incorporation into keratin tissues may help us better understand the influence additional intravenous fluids on travel reconstruction.

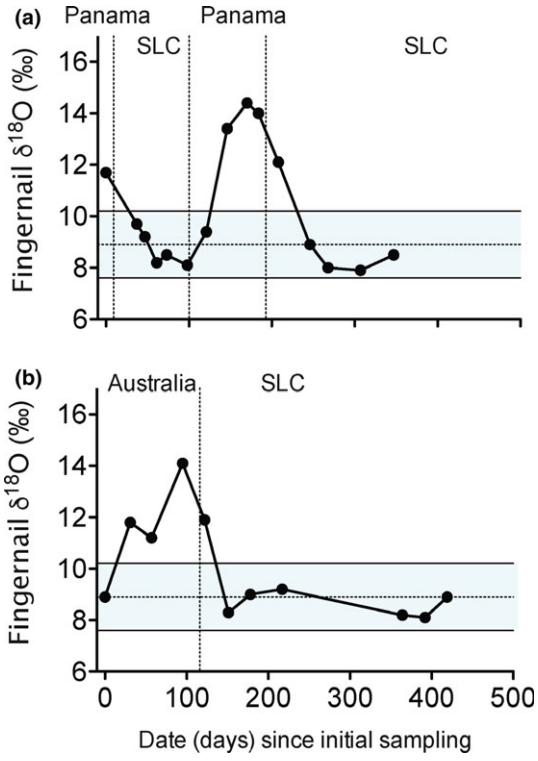

FIG. 2—*Fingernail oxygen isotope ratios from (a) participant 4 and (b) participant 53 and location where the fingernails were developed. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24). [Color figure can be viewed at wileyonlinelibrary.com]*

A Tale of Two Travelers—The Galapagos Story

Participants and Travel History—In the beginning of January 2015, two individuals (participants 5 and 6) traveled to the Galapagos Islands, Ecuador from Salt Lake City, UT for a period of 4.5 months before returning. Both individuals were residents of the Salt Lake City region prior to traveling. Fingernail clippings

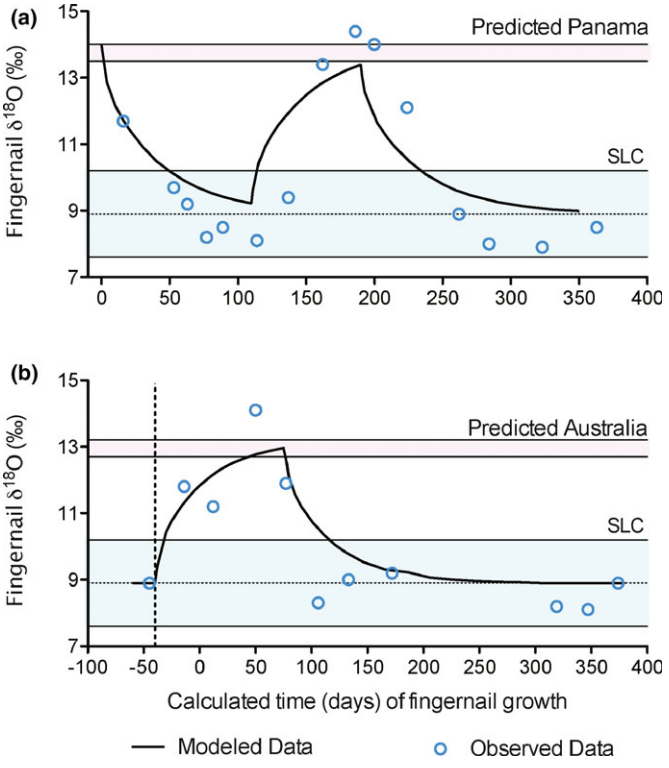

FIG. 3—*Fingernail oxygen isotope ratios of reaction progress modeled data (solid line) and observed traveler data (open blue circle) (a) participant 4 (b) 53 over time. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24) and pink box reflects the predicted oxygen isotope value of scalp hair (18) using calculated precipitation isotope values from the OPIC (29) for the travelers given region. [Color figure can be viewed at wileyonlinelibrary.com]*

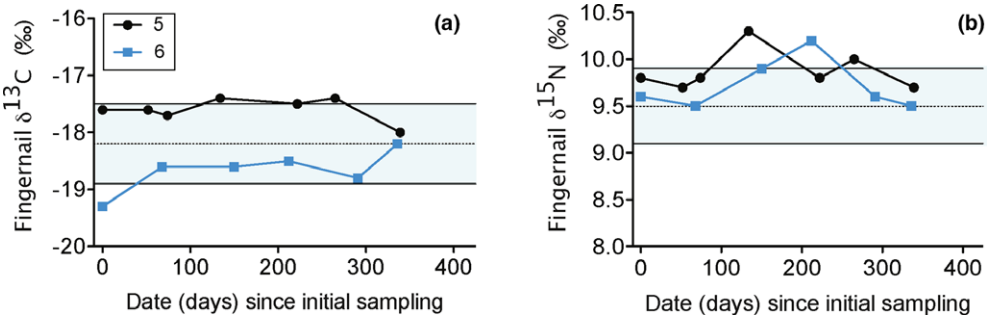

FIG. 4—*(a) Carbon and (b) nitrogen fingernail isotope profiles of participants 5 and 6 over time. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24). [Color figure can be viewed at wileyonlinelibrary.com]*

were collected starting from February 2015 to January 2016 and were analyzed for dietary (carbon and nitrogen stable isotope ratios) and drinking water (oxygen and hydrogen stable isotope ratios) inputs that would reflect their time in the Galapagos and Salt Lake City. The fingernail clippings provided us with a timeline of their dietary and travel history from ~ November 2014 to October 2015 based on the time lag associated with fingernail development, maturation, and growth to the free edge of the nail (24,25). This would allow us to examine inputs from their residential period in Salt Lake City, travel, and equilibration to the Galapagos environment, and their return to the Salt Lake City.

Dietary Isotopes—Over the course of the study, we did not observe differences in the intra-individual dietary isotopes. The minor fluctuations in dietary isotope values did not indicate significant dietary changes between individuals over the study, nor did we detect any differences in stable isotope ratios that corresponded with the time that they resided in the Galapagos. The inter-carbon and nitrogen isotope values (Fig. 4) for participant 5 ($-17.6 \pm 0.2‰$ and $9.9 \pm 0.2‰$) and participant 6 ($-18.7 \pm 0.4‰$ and $9.7 \pm 0.3‰$) displayed significant differences between the mean carbon isotope ratios ($p = 0.003$), but not for the mean nitrogen isotope ratio values ($p = 0.171$) over the study period. The carbon isotope ratio differences indicate that participant 5 incorporated (direct or indirectly) more C_4 foods (~69% of carbon isotopes being derived from C_4 sources) in their diet than compared to participant 6 (~62%). Based on their dietary surveys, both individuals had omnivore diets with similar estimates of meat and vegetable/fruit intakes. The overall lower carbon isotope ratios of participant 6 could be attributed to the consumption of grass fed meats or foods containing fewer C_4 byproducts, compared to participant 5, who did not report high levels of marine seafood consumption that could account for their elevated carbon isotope ratio (37,41,42).

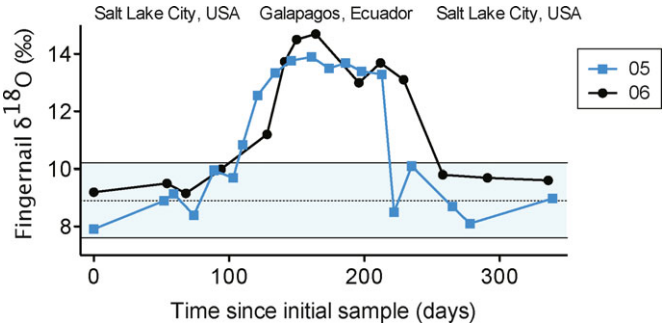

FIG. 5—*Oxygen isotope ratios of traveler fingernails and location where the fingernails were developed. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24). [Color figure can be viewed at wileyonlinelibrary.com]*

Drinking Water and Travel History Reconstruction—Variations in the stable isotope profiles of our two individuals throughout the study were consistent with their reported travel history. Specifically that portions of the fingernails produced in Salt Lake City had oxygen isotope ratios that were consistent with resident group mean [$8.9 \pm 1.3‰$ and $-104.1 \pm 5‰$ (24)] and those portions of the fingernails developed in the Galapagos were outside the range of values for the Salt Lake City resident group signal (Fig. 5, oxygen isotope ratio values are only shown as patterns were similar). Oxygen isotope values that were reflective of the Galapagos travel period are significantly different from the resident group mean value ($p < 0.0001$) for both individuals. Inter-individual differences were present between participant 5 and participant 6, with participant 6 tending to have samples more ^{18}O enriched. This consistent offset between the two individuals could be related to individual metabolic differences that relate to sex, body mass index, exercise, water intake, and perspiration (23,43). However, further research is needed to examine within-population variance in the isotopic composition and relationship of body water and keratin proteins to determine the cause of this offset (23).

As we previously mentioned, the reported travel histories of our two individuals were consistent with the oxygen isotope ratio profiles that were constructed from their fingernail clippings. In particular, participant 5 remained on the Galapagos Island throughout the 4.5-month period, which allowed for a consistent drinking water source associated with the island and thus, they were able to completely incorporate the isotope signature of the new location into their tissues (manifested in the plateau in Fig. 5) before leaving. Unlike participant 5, participant 6 did not remain on the island throughout the 4.5 months period, but instead traveled to Quito, Ecuador for 2 weeks, half-way through their time in the Galapagos. This movement away from the island can be seen as the sharp decline in the oxygen isotope values (Fig. 5), which is consistent with the integration of drinking waters from higher elevation (being depleted in the heavy isotope values) (17,19,44). Their fingernail tissues became ^{18}O enriched upon return to the island, but never fully reached their pre-Quito values before the individuals returned to Salt Lake City. It is remarkable that short-term travels of participant 6 were detectable. The oxygen isotope ratios of this individual's fingernails revealed the brief Galapagos-Quito travel event, highlighting the ability of the fingernail stable isotope ratio record to capture brief trips or movements.

Lastly, when we compared our observed data with that of the reaction progress variable modeled data (Fig. 6), a significant linear relationship was found ($\delta^{18}O_{obs} = 1.07 \times \delta^{18}O_{pred} - 0.876‰$, $r^2 = 0.7288$, $p < 0.0001$). Overall, the observed data fit the modeled expectation well (slope of the line was similar to the slope if data had fit perfectly on the 1:1 slope). The modeled data underestimated the oxygen isotope ratio values of the fingernails mostly during the transition to the local Galapagos signature. The trip to Quito by participant 6 could add to this under-representation in the model.

A Traveler Returns—Repeated Travel to the Galapagos, Ecuador

Repeated Travel History—Participant 6 made two trips to the Galapagos Islands from Salt Lake City in 2015 and again in 2016. The 2015 trip, which was described in the previous section, was repeated in 2016. Only participant 6 made this trip, and left Salt Lake City in January 2016 and resided in the Galapagos for a period of 3.5 months before returning at the end of April 2016. During this trip, participant 6 collected fingernail clippings from May 2016 until January 2017, which would reflect fingernails synthesized from February 2016 to October 2016. We would miss the immediate isotope departure from the resident value (nails clipped in April 2016), but our sampling dates would capture their time on the island and the return to Salt Lake City.

Isotope analysis of the return trip—Dietary isotopes were measured from fingernail clippings during the repeated trip to the Galapagos, and differences in mean carbon and nitrogen stable isotope ratios were indistinguishable from those measured during the first sampling period (Fig. 5). This is not surprising as the individual did not report major changes to their diet over the two-year sampling period. Similar to the previous sampling period, isotope ratio differences associated with travels to the Galapagos and back did not display significant changes in their dietary inputs.

The oxygen isotope ratio profiles of the two trips are similar to each other in their incorporation of the Galapagos waters into the fingernail keratin proteins (Fig. 7). During both trips, the peak oxygen isotope ratios differed by ~0.3‰, which is within the precision of the isotope ratio measurement. The turnover period in the second trip to the Galapagos was much shorter, ~30 days shorter than the original, but there were similarities in the patterns and measured isotope values on their return to Salt Lake City. The consistency of the isotope ratio profiles over the two trips is remarkable, the isotope ratio turnover of the oxygen isotope ratios as the participant moved from the Galapagos to Salt Lake City are within 5 days of each other (45-51 days). The consistencies in the isotope ratio profiles of the repeated travel history suggest that isotope incorporation is consistent within an individual for a given location and that fingernail clippings record this information well.

Conclusion

Ehleringer et al. (18), Bowen et al. (21), and O'Grady et al. (23) developed the theoretical framework for understanding the oxygen and hydrogen isotope ratio inputs (drinking and food waters) that contribute to the isotope ratio values of human scalp hair. In addition to Bowen et al. (19), Hobson et al. (45), Wassenaar and Hobson (46) established that drinking waters could be used to understand movement and migration patterns, which were subsequently applied to butterfly and bird migratory studies. Their work was grounded on the spatial differences of precipitation over geographic landscapes and subsequently has been applied to human studies. Furthermore, work from Cerling et al. (22) has focused on using a different modeling technique for isotope incorporation into different tissues, that suggests that there are multiple pools within the body that have different turnover rates and thus, impact the time it takes for a tissue to fully integrate a new input (location, diet, etc.). This work has allowed us to predict when isotope changes and stabilization within a tissue could be expected.

The examination and interpretation of our case studies have pulled from the theoretical groundwork that has come before to highlight the forensic and anthropologic applications that can take advantage of these relationships and natural isotope patterns that exist. Our case studies of travel history reconstruction and movement patterns support the reaction progress variable model related to the timing of isotope ratio changes and incorporation of those values associated with changes in geospatial locations. Additionally, the turnover of the oxygen isotope ratios in human fingernails is consistent with prior studies examining keratin proteins (18,20,24). The rapid turnover time for oxygen isotope

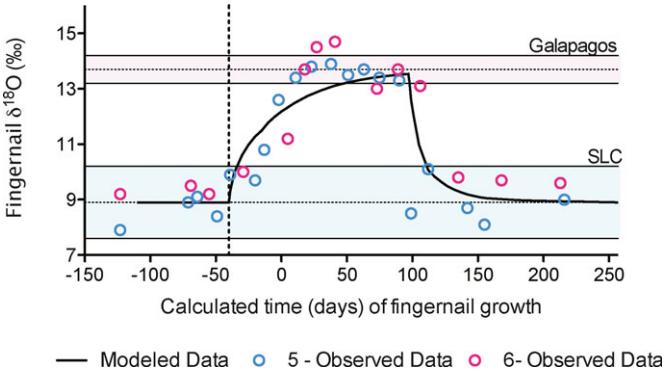

FIG. 6—*Oxygen isotope ratios of reaction progress model (black line) compared to observed data from participants 5 and 6. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24) and pink box reflects the predicted oxygen isotope value of scalp hair (18) using calculated precipitation isotope values from the OPIC (29) for the travelers given region. [Color figure can be viewed at wileyonlinelibrary.com]*

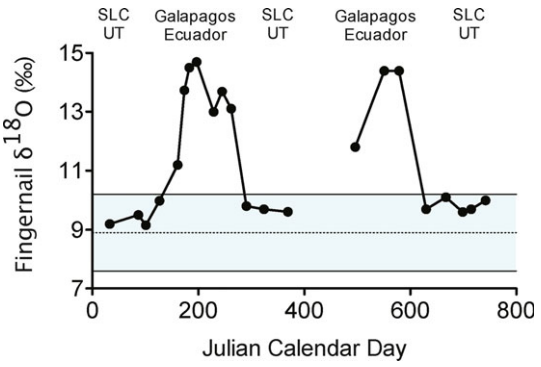

FIG. 7—*Oxygen isotope profile of participant 6, who made two trips to the Galapagos, Ecuador from Salt Lake City in 2015 and 2016. Blue box reflects the Salt Lake City resident group mean ± SD oxygen isotope ratio (24). Dotted vertical lines reflect time when fingernails were developed in the Galapagos. [Color figure can be viewed at wileyonlinelibrary.com]*

ratios in these tissues is linked to the full exchange of oxygen isotopes in protein sources with body water sources (2,23). Lastly, the link between local precipitation, drinking water sources, and isotope ratios in fingernail proteins also support previous studies. We have shown that changes in drinking water inputs relate to changes in isotope ratios in our case studies that can be linked to local precipitation and modeled to predict expected local keratin (scalp hair) isotope values.

Here, we have highlighted the use of stable isotope ratio biomarkers in human fingernail tissues to examine travel histories. Our case studies have specifically looked at fingernail clippings collected from individuals over a period of 9–24 months to examine travel history patterns. While this long-term collection of fingernail clippings is reasonable within living populations, travel history reconstruction from human remains may only reflect a period of 5–6 months. This time limitation is a potential shortcoming to applying fingernail tissues for these purposes; however, we have shown through our case studies that change in location can be measured and differentiated from a resident population over a 5–6 month period. Fingernail tissues from human remains may not be able to reflect multiple trips to a given location (e.g., case study 3), but we can see changes related to individual trips (e.g., case studies 1 and 2) within the length of a fingernail. Fingernail tissues are useful for understanding human movement and travel reconstruction that could aid anthropological and forensic studies in determining travel histories of human remains.

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References

1. O'Connell TC, Hedges REM, Healey MA, Simpson AHRW. Isotopic comparison of hair, nail and bone: modern analyses. J Archaeol Sci 2001;28(11):1247–55.
2. Thompson AH, Chesson LA, Podlesak DW, Bowen GJ, Cerling TE, Ehleringer JR. Stable isotope analysis of modern human hair collected from Asia (China, India, Mongolia, and Pakistan). Am J Phys Anthropol 2010;141(3):440–51.
3. Nardoto GB, Silva S, Kendall C, Ehleringer JR, Chesson LA, Ferraz ESB, et al. Geographical patterns of human diet derived from stable-isotope analysis of fingernails. Am J Phys Anthropol 2006;131(1):137–46.
4. Knudson KJ, Pestle WJ, Torres Rouff C, Pimentel G. Assessing the life history of an andean traveller through biogeochemistry: stable and radiogenic isotope analyses of archaeological human remains from Northern Chile. Int J Osteoarchaeol 2012;22(4):435–51.
5. Gregoricka LA, Scott AB, Betsinger TK, Polcyn M. Deviant burials and social identity in a postmedieval Polish cemetery: an analysis of stable oxygen and carbon isotopes from the "vampires" of Drawsko. Am J Phys Anthropol 2017;163(4):741–58.
6. Meier-Augenstein W, Fraser I. Forensic isotope analysis leads to identification of a mutilated murder victim. Sci Justice 2008;48(3):153–9.
7. Cerling TE, Wittemyer G, Ehleringer JR, Remien CH, Douglas-Hamilton I. History of animals using isotope records (HAIR): a 6-year dietary history of one family of African elephants. Proc Natl Acad Sci USA 2009;106(20):8093–100.
8. DeNiro MJ, Epstein S. Influence of diet on the distribution of carbon isotopes in animals. Geochim Cosmochim Acta 1978;42(5):495–506.
9. Peterson BJ, Fry B. Stable isotopes in ecosystem studies. Annu Rev Ecol Syst 1987;18(1):293–320.
10. Gragnani JG, Garavello MEPE, Silva RJ, Nardoto GB, Martinelli LA. Can stable isotope analysis reveal dietary differences among groups with distinct income levels in the city of Piracicaba (southeast region, Brazil)? J Hum Nutr Diet 2014;27(3):270–9.
11. Kusaka S, Ishimaru E, Hyodo F, Gakuhari T, Yoneda M, Yumoto T, et al. Homogeneous diet of contemporary Japanese inferred from stable isotope ratios of hair. Sci Rep 2016;6:33122.
12. McCullagh JSO, Tripp JA, Hedges REM. Carbon isotope analysis of bulk keratin and single amino acids from British and North American hair. Rapid Commun Mass Spectrom 2005;19(22):3227–31.
13. Valenzuela LO, O'Grady SP, Enright LE, Murtaugh M, Sweeney C, Ehleringer JR. Evaluation of childhood nutrition by dietary survey and stable isotope analyses of hair and breath. Am J Hum Biol 2018;30(3): e23103.
14. Nakamura K, Schoeller DA, Winkler FJ, Schmidt HL. Geographical variations in the carbon isotope composition of the diet and hair in contemporary man. Biomed Mass Spectrom 1982;9(9):390–4.
15. Farquhar GD, Ehleringer JR, Hubick KT. Carbon isotope discrimination and photosynthesis. Annu Rev Plant Physiol Plant Mol Biol 1989;40 (1):503–37.
16. DeNiro MJ, Epstein S. Influence of diet on the distribution of nitrogen isotopes in animals. Geochim Cosmochim Acta 1981;45(3):341–51.
17. Michener RH, Lajtha K. Stable isotopes in ecology and environmental science. Malden, MA: Blackwell Publishing, 2007.
18. Ehleringer JR, Bowen GJ, Chesson LA, West AG, Podlesak DW, Cerling TE. Hydrogen and oxygen isotope ratios in human hair are related to geography. Proc Natl Acad Sci USA 2008;105(8):2788–93.
19. Bowen GJ, Ehleringer JR, Chesson LA, Stange E, Cerling TE. Stable isotope ratios of tap water in the contiguous United States. Water Resour Res 2007;43(3):W03419.
20. O'Brien DM, Wooller MJ. Tracking human travel using stable oxygen and hydrogen isotope analyses of hair and urine. Rapid Commun Mass Spectrom 2007;21(15):2422–30.
21. Bowen GJ, Ehleringer JR, Chesson LA, Thompson AH, Podlesak DW, Cerling TE. Dietary and physiological controls on the hydrogen and oxygen isotope ratios of hair from mid-20th century indigenous populations. Am J Phys Anthropol 2009;139(4):494–504.
22. Cerling TE, Ayliffe LK, Dearing MD, Ehleringer JR, Passey BH, Podlesak DW, et al. Determining biological tissue turnover using stable isotopes: the reaction progress variable. Oecologia 2007;151(2):175–89.
23. O'Grady SP, Valenzuela LO, Remien CH, Enright LE, Jorgensen MJ, Kaplan JR, et al. Hydrogen and oxygen isotope ratios in body water and hair: modeling isotope dynamics in nonhuman primates. Am J Primatol 2012;74(7):651–60.
24. Mancuso CJ, Ehleringer JR. Resident and non-resident fingernail isotopes reveal diet and travel patterns. J Forensic Sci In press; http://doi.org/10.1111/1556-4029.13856.
25. De Berker D, Baran R. Science of the nail apparatus. In: Baran R, De Berker D, Holzberg M, Thomas L, editors. Baran & Dawber's diseases of the nails and their management, 4th edn. Oxford, U.K.: Wiley-Blackwell, 2012;1–50.
26. Dawber RPR, Baran R. Nail growth. Cutis 1987;39:99–103.
27. Bowen GJ, Chesson L, Nielson K, Cerling TE, Ehleringer JR. Treatment methods for the determination of delta2H and delta18O of hair keratin by continuous-flow isotope-ratio mass spectrometry. Rapid Commun Mass Spectrom 2005;19(17):2371–8.
28. Chesson LA, Podlesak DW, Cerling TE, Ehleringer JR. Evaluating uncertainty in the calculation of non-exchangeable hydrogen fractions within organic materials. Rapid Commun Mass Spectrom 2009;23 (9):1275–80.
29. Bowen GJ. The online Isotopes in precipitation calculator. Version OIPC 3.1, 2018; http://www.waterisotopes.org (accessed March 1, 2018).
30. Bowen GJ, Revenaugh J. Interpolating the isotopic composition of modern meteoric precipitation. Water Resour Res 2003;39(10):1299.
31. IAEA/WMO. Global network of isotopes in precipitation, 2015; https://nucleus.iaea.org/wiser (accessed June 12, 2018).
32. Tipple BJ, Valenzuela LO, Ehleringer JR. Strontium isotope ratios of human hair record intra-city variations in tap water source. Sci Rep 2018;8(1):3334.
33. Ehleringer JR, Barnette JE, Jameel Y, Tipple BJ, Bowen GJ. Urban water–a new frontier in isotope hydrology. Isotopes Environ Health Stud 2016;52(4–5):477–86.
34. Good SP, Kennedy CD, Stalker JC, Chesson LA, Valenzuela LO, Beasley MM, et al. Patterns of local and nonlocal water resource use across

the western U.S. determined via stable isotope intercomparisons. Water Resour Res 2014;50(10):8034–49.
35. Drapé J-L, Wolfram-Gabel R, Idy-Peretti I, Baran R, Goettmann S, Sick H, et al. The lunula: a magnetic resonance imaging approach to the subnail matrix area. J Invest Dermatol 1996;106(5):1081–5.
36. Mützel E, Lehn C, Peschel O, Hölzl S, Roßmann A. Assignment of unknown persons to their geographical origin by determination of stable isotopes in hair samples. Int J Legal Med 2009;123(1):35–40.
37. Michener RH, Kaufman L. Stable isotope ratios as tracers in marine food webs: an update. In: Michener R, Lajtha K, editors. Stable isotopes in ecology and environmental science. Malden, MA: Blackwell Publishing, 2007.
38. Chesson LA, Podlesak DW, Thompson AH, Cerling TE, Ehleringer JR. Variation of hydrogen, carbon, nitrogen, and oxygen stable isotope ratios in an American diet: fast food meals. J Agric Food Chem 2008;56 (11):4084–91.
39. Petzke KJ, Boeing H, Klaus S, Metges CC. Carbon and nitrogen stable isotopic composition of hair protein and amino acids can be used as biomarkers for animal-derived dietary protein intake in humans. J Nutr 2005;135(6):1515–20.
40. Jahren AH, Kraft RA. Carbon and nitrogen stable isotopes in fast food: signatures of corn and confinement. Proc Natl Acad Sci USA 2008;105 (46):17855–60.
41. Fry B. Stable isotope ecology. New York, NY: Springer, 2006.
42. Cree A, Lyon GL, Cartland-Shaw L, Tyrrell C. Stable carbon isotope ratios as indicators of marine versus terrestrial inputs to the diets of wild and captive tuatara (*Sphenodon punctatus*). New Zeal J Zool 1999;26 (3):243–53.
43. Fraser I, Meier-Augenstein W, Kalin RM. The role of stable isotopes in human identification: a longitudinal study into the variability of isotopic signals in human hair and nails. Rapid Commun Mass Spectrom 2006;20 (7):1109–16.
44. Bowen GJ, Wassenaar LI, Hobson KA. Global application of stable hydrogen and oxygen isotopes to wildlife forensics. Oecologia 2005;143 (3):337–48.
45. Hobson KA, Atwell L, Wassenaar LI. Influence of drinking water and diet on the stable-hydrogen isotope ratios of animal tissues. Proc Natl Acad Sci USA 1999;96(14):8003–6.
46. Wassenaar LI, Hobson KA. Natal origins of migratory monarch butterflies at wintering colonies in Mexico: new isotopic evidence. Proc Natl Acad Sci USA 1998;95(26):15436–9.

Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Figure S1. Oxygen isotope ratios of traveler fingernails and location where the fingernails were developed.

Figure S2. Fingernail oxygen isotope ratios of reaction progress modeled data (solid line) and observed traveler data (open blue circle) (A) participant 15 (B) participant 9 over time.