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RESEARCH ARTICLE

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A predictive spatial model for roasted coffee using oxygen isotopes of α-cellulose

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Rationale: Fraudulent region-of-origin labeling is a concern for high-value, globally traded commodities such as coffee. The oxygen isotope ratio of cellulose is a useful geographic tracer, as it integrates climate and source water signals. A predictive spatial model ("isoscape") of the $\delta^{18}O$ values of coffee bean cellulose is generated to evaluate coffee region-of-origin claims.

Methods: The oxygen isotope ratio of α-cellulose extracted from roasted coffee beans was measured via high-temperature conversion elemental analyzer/isotope ratio mass spectrometry (TC-EA/IRMS) and used to calculate the $\delta^{18}O$ value of coffee bean water. The ^{18}O enrichment of coffee bean water relative to the $\delta^{18}O$ value of local precipitation was modeled as a function of local temperature and humidity. This function was incorporated into a mechanistic model of cellulose $\delta^{18}O$ values to predict the $\delta^{18}O$ values of coffee bean cellulose across coffee-producing regions globally.

Results: The $\delta^{18}O$ values of analyzed coffee bean cellulose ranged from approximately +22‰ to +42‰ (V-SMOW). As expected, coffees grown in the same region tended to have similar isotope ratios, and the $\delta^{18}O$ value of coffee bean cellulose was generally higher than the $\delta^{18}O$ value of modeled stem cellulose for the region. Modeled $\delta^{18}O$ values of coffee cellulose were within ±2.3‰ of the measured $\delta^{18}O$ value of coffee cellulose.

Conclusions: The oxygen isotope ratio of coffee bean cellulose is a useful indicator of region-of-origin and varies predictably in response to climatic factors and precipitation isotope ratios. The isoscape of coffee bean cellulose $\delta^{18}O$ values from this study provides a quantitative tool that can be applied to region-of-origin verification of roasted coffee at the point-of-sale.

1 | INTRODUCTION

Coffee is one of the most valuable globally traded commodities, with total exports valued over $30 billion in 2016 and approximately 168 million 60-kg bags produced in 2018.[1,2] Several factors drive differential valuation of coffee internationally. Region of origin is one determinant of bean quality and price, with certain coffees, such as Kona from the Big Island of Hawaii, Jamaica Blue Mountain from Jamaica, and St Helena from St Helena Island, commanding high price premiums.[3] As the differential in the prices of coffee from these highly sought-after regions increased relative to those of other coffees, fraudulent region-of-origin labeling became increasingly common and threatened to undercut the value of authentic coffee beans.[4] The development of a supply-chain-integrity analytical method for the verification of coffee region of origin would protect growers, distributors, and consumers from counterfeit coffee beans.

Stable isotope ratios have been used to identify adulterated foods and to establish region-of-origin associations for a wide range of animal and plant products, including honey, cotton, beef, wine, fruits, dairy, and oils.[5-8] In addition, trace element abundances have been

applied as diagnostic and quantitative parameters that distinguish food products from different regions and serve as a differentiating trait for products from protected domains.[5,6,9,10] Several studies have associated the chemical and isotopic characteristics of coffee with region of origin; previous approaches include measurements of carbon, nitrogen, boron, strontium, hydrogen, and oxygen stable isotope ratios,[11-16] and elemental concentrations.[9,10] These studies established that isotope ratios and elemental concentrations in coffee vary geospatially, and that coffees grown in the same region tend to have similar isotopic and elemental compositions.[9,11-13,16,17] However, these approaches have been unable to predict spatial patterns in isotope ratios.[11]

With one exception, previous coffee studies have focused on the analysis of bulk tissue of green coffee beans, thereby complicating comparisons with roasted coffee at the point-of-sale for most consumers. Carter et al[17] analyzed bulk tissue of roasted coffee beans and found that the range of $\delta^{18}O$ values observed in roasted coffee was slightly narrower than that of green coffee in previous studies. Roasting times and temperatures vary among coffees, and roasting results in compound-specific volatilization that affects the chemical composition of the roasted bean.[18] These factors introduce uncontrolled variability in the isotope measurements of bulk material because the isotopic composition of biochemical components varies within plant tissues.[19,20]

Coffee bean cellulose does not undergo measurable degradation during roasting[18] and the oxygen in α-cellulose does not undergo subsequent exchange or fractionation after formation.[21,22] Therefore, measurements of coffee bean cellulose would allow for analysis at the point-of-sale, avoiding isotopic variability arising from roasting-related changes in the chemical composition of the coffee bean. In this study, we extracted and analyzed the α-cellulose of roasted coffee. Here we suggest that the oxygen stable isotope ratios of coffee bean cellulose are a useful measurement for assessing the authenticity of coffee region-of-origin claims. An advantage of cellulose oxygen isotope ratio measurements is the strong theoretical and experimental foundation allowing one to model cellulose isotope ratio values based on environmental characteristics.[23,24]

The oxygen isotope ratio of α-cellulose has been shown to be a useful geographic tracer because it varies predictably in response to temperature, humidity, and the $\delta^{18}O$ value of water taken up by the plant during the time of tissue formation. The climatic and physiological parameters driving the oxygen isotopic composition of α-cellulose in wood and leaves are well studied, and the biochemical fractionation events associated with cellulose synthesis have been identified and modeled.[25-28]

These fractionation and exchange processes associated with cellulose synthesis were integrated into a single model of wood and leaf cellulose isotope ratios by Roden et al[23] that incorporates the influences of temperature, relative humidity, and the oxygen isotope ratio of source water. The use of cellulose $\delta^{18}O$ values as a tracer of coffee origin relies on the known spatial variability in climate and source water isotopes among the coffee-producing regions of the world. *Coffea arabica* requires a narrow range of climatic conditions for optimal growth, including year-round temperatures of 18–21°C, annual rainfall between 1200 and 1800 mm, and a 2–4-month dry season.[29] Despite the specificity of growing conditions, the variations in temperature, relative humidity, and the oxygen isotope ratio of source water among coffee-producing regions are sufficient to produce predictable spatial variations in bulk coffee $\delta^{18}O$ values.[11,13]

The $\delta^{18}O$ value of α-cellulose is a function of the $\delta^{18}O$ value of the sugars used in cellulose synthesis and the $\delta^{18}O$ value of water at the site of cellulose synthesis, as shown in Equation 1. Here, P_{ex} is the proportion of exchangeable oxygen (0.42), $\delta^{18}O_s$ and $\delta^{18}O_l$ are the oxygen isotope ratios of water at the site of cellulose synthesis and the site of evaporation in the leaf, respectively, and ε_o is the enrichment factor associated with isotopic fractionation in cellulose synthesis[21-23]:

$$\delta^{18}O_{cell} = P_{ex} \bullet \left(\delta^{18}O_s\right) + (1 - P_{ex}) \bullet \left(\delta^{18}O_l\right) + \varepsilon_{o.} \quad (1)$$

The $\delta^{18}O$ value of xylem water in suberized stems is generally accepted to be identical to that of the source water,[30] whereas the $\delta^{18}O$ value of leaf water is more positive than that of source water and has been modeled as a function of relative humidity, temperature, the source water $\delta^{18}O$ value, atmospheric pressure, and stomatal and boundary layer conductance.[31,32] We expect the evaporative enrichment of ^{18}O in coffee bean water to be driven by the same evaporative mechanisms that influence leaf water enrichment. Therefore, the coffee bean water will probably be enriched in ^{18}O relative to stem water. We constructed an empirical function analogous to that for leaf water enrichment to describe the $\delta^{18}O$ value of coffee seed water based on temperature, humidity, and source water.

We developed a predictive model of the oxygen isotope ratio of coffee bean α-cellulose by modifying the established model of Roden et al[23] for the $\delta^{18}O$ value of wood cellulose. We calculated the $\delta^{18}O$ value of coffee bean water using the known relationship between water at the site of synthesis and the cellulose $\delta^{18}O$ value and modeled the ^{18}O enrichment of coffee bean water as a function of local relative humidity, temperature, and the source water $\delta^{18}O$ value. We incorporated this bean water enrichment function into a spatially explicit cellulose model to create an isoscape of the likely $\delta^{18}O$ values of coffee bean cellulose for coffee-growing regions around the world. The resultant global map of coffee cellulose $\delta^{18}O$ values will facilitate the prediction of probable regions of origin for a specific coffee sample and thus aid in efforts to reduce fraudulent region-of-origin labeling.

2 | METHODS

2.1 | Sampling

Forty-nine different roasted coffee samples, originating from 21 different countries, were purchased from reputable sources in late 2018 for stable isotope analysis. Forty of these samples were labeled as single-origin with known information on the specific

growth location. The remaining nine samples were labeled only with the country of production and may have been blends of beans from several locations within that country.

2.2 | Cellulose extraction

Samples were ground, sieved to 40–60 mesh, and 0.4-g subsamples were sealed in extraction bags (60-mesh filter bag material; ANKOM Technology, Macedon, NY, USA). Samples were boiled in water for 3 h to remove excess water-soluble compounds prior to the extraction of holocellulose[33] with subsequent purification to α-cellulose[34] according to established protocols. To summarize, lipids were extracted in a Soxhlet apparatus with a 2:1 toluene/ethanol solution for 48 h followed by extraction with ethanol for 24 h. Samples were then bleached in a solution of sodium hypochlorite and acetic acid to produce holocellulose. Hemicellulose was removed using a 17% w/v sodium hydroxide solution followed by an acetic acid rinse. The resultant α-cellulose was dried for a minimum of 24 h at 70°C prior to being loaded into silver capsules for stable isotope analysis. The loaded capsules were stored in a vacuum desiccator until analysis.

2.3 | Stable isotope analysis

A total of 166 samples, including at least two replicates of each extracted α-cellulose sample (with the exception of a sample from Kenya, which was only analyzed once), were analyzed for their oxygen isotope ratios using a high-temperature conversion elemental analyzer (TC-EA) coupled to either a ThermoFinnigan MAT 253 or a ThermoFinnigan Delta+ XL isotope ratio mass spectrometer (all instruments from Thermo Scientific, Bremen, Germany). Samples were introduced into the TC-EA via a zero-blank autosampler equipped with an isolation valve (Costech Analytical Technologies, Inc., Valencia, CA, USA). The results are reported in delta notation relative to Vienna Standard Mean Ocean Water (V-SMOW). The data were normalized using laboratory cellulose reference materials (H1 and L6; δ^{18}O = +19.2‰ and +40.6‰, respectively), whose oxygen and hydrogen isotope ratios have been calibrated against international benzoic acid standards IAEA-601 and IAEA-602 (δ^{18}O = +23.3‰ and + 71.4‰, respectively; International Atomic Energy Association, Vienna, Austria). Several quality control materials, including Sigma-Aldrich cellulose ("Cell", δ^{18}O = +28.8‰; Sigma Chemical Co., St Louis, MO, USA) and an in-house coffee cellulose sample (δ^{18}O = +30.1‰), were included in each run to assess measurement precision and monitor long-term stability. Based on the Sigma-Aldrich cellulose standard, the combined standard uncertainty for the measurement of δ^{18}O values of cellulose was ±0.19‰.

2.4 | Data acquisition

Gridded monthly temperature and vapor pressure data were obtained for 2015–2018 from the Climatic Research Unit time-series dataset version 4.03[35] at 0.5° resolution and used to calculate relative humidity. The climate data were resampled to 1° resolution using bilinear interpolation to better match the spatial precision of the available coffee region-of-origin information. The mean temperature and relative humidity over the 2015–2018 period were used as cellulose model inputs.

Gridded data on the annual average δ^{18}O value of precipitation were acquired from the OIPC database (version 3.2),[36] which is a 0.083° resolution interpolation of the GNIP database[37] produced using the methods presented by Bowen and Revenaugh.[38] The precipitation isotope data were resampled to 1° resolution using bilinear interpolation prior to use in the cellulose model. All data extraction and processing were conducted in R[39] (version 3.4.1), using packages 'raster',[40] 'rgdal',[41] `ncdf4`,[42] and 'geosphere'.[43]

Spatial data on areas of coffee harvest were obtained from Sachs et al[44] at 0.083° resolution and resampled to 1° resolution. All data were constrained to grid cells in which a minimum of 10 hectares of coffee were harvested. For the 40 coffee samples labeled with a region of origin, the 1 × 1° grid cell containing the center point of that region was used for the extraction of climate and precipitation data. If a farm address was available, the address was used instead of the region's center point to derive coordinates for the sample.

2.5 | Analysis

The model of leaf and stem cellulose isotopes developed by Roden et al[23] was applied to coffee-growing regions. As the model is relatively robust to changes in stomatal conductance, boundary layer conductance, and barometric pressure within a reasonable range of values, we assumed the stomatal conductance to be equal to 0.3 mol m^{-2} s^{-1}, the boundary layer conductance to be equal to 1 mol m^{-2} s^{-1}, and the barometric pressure to be equal to 90 kPa. Climate and precipitation isotope data were used in the cellulose isotope model to estimate the δ^{18}O values of leaf water, leaf cellulose, and stem cellulose across the coffee-producing regions of the world.

The mean of all replicates for a given sample was used for analysis. For samples with known regions of origin, the δ^{18}O values of coffee seed water were calculated using measured δ^{18}O values of coffee seed cellulose and predicted leaf water δ^{18}O values. In order to spatially model δ^{18}O values of coffee seed water, we used multiple linear regression to derive an equation relating the calculated δ^{18}O value of coffee seed water from known regions with environmental drivers, including relative humidity, temperature, and precipitation δ^{18}O values (Equation 2). This equation for estimating spatially explicit seed water δ^{18}O values was incorporated into the existing cellulose isotope model to produce an isoscape of coffee seed cellulose for coffee-producing areas. All maps were created in R using package 'lattice'.[45]

TABLE 1 Sampled regions, the mean and standard deviation (σ) of the measured $\delta^{18}O$ values of coffee cellulose for the region, the number of distinct farms sampled within the region, and the total number of replicates analyzed from that region. Samples without a known region of origin are designated by country

Country	Region	Mean $\delta^{18}O$	σ $\delta^{18}O$	Farms	Replicates
Brazil	Carmo de Minas	32.09	0.05	1	2
Brazil	Cerrado, Minas Gerais	32.21	0.32	1	2
Brazil	Espirito Santo	33.40	1.38	1	2
Brazil	Mogiana	30.19	0.03	1	2
Brazil	N/A	31.59	0.18	1	2
Burundi	Gakenke	34.79	0.70	1	4
Colombia	N/A	24.39	1.20	2	4
Colombia	Palestina, Huila	28.71	0.45	1	2
Colombia	Vereda Montalvo	26.04	0.66	1	2
Costa Rica	N/A	26.65	0.56	1	2
Costa Rica	Tarrazu	24.12	0.48	2	4
El Salvador	Apaneca	27.48	3.08	2	4
Ethiopia	Gedeb District	35.85	0.31	1	2
Ethiopia	Guji	33.72	0.29	1	2
Ethiopia	Yirgacheffe	33.13	0.88	1	2
Guatemala	Antigua	32.37	0.12	1	2
Guatemala	Huehuetenango	27.43	0.01	1	2
Guatemala	N/A	26.46	0.21	1	2
Honduras	Marcala	26.30	0.06	1	2
Honduras	N/A	29.68	0.21	1	2
Honduras	Santa Barbara	27.62	0.82	1	2
India	Chikmagalur	30.92	0.01	1	2
Indonesia	Kokowagayo	27.67	0.22	1	2
Indonesia	Sumatra	26.00	0.07	1	2
Indonesia	Toraja, Sulawesi	26.76	0.09	1	2
Kenya	Nyeri	36.85	N/A	1	1
Mexico	Chiapas	26.39	0.56	1	2
Nicaragua	Jinotega	27.85	1.20	2	4
Panama	N/A	25.13	0.40	1	2
Panama	Volcan	24.30	0.06	1	2
Papua New Guinea	Kainantu	22.79	0.49	1	13
Papua New Guinea	Kimel estate	22.76	0.22	1	2
Peru	Centrocafe	23.54	0.12	1	2
Peru	N/A	24.93	0.33	1	2
Rwanda	Nyakibanda	36.29	0.06	1	2
Tanzania	Mbeya	34.61	0.56	1	2
USA	Hilo	30.49	0.15	1	2
USA	Ka'anapali, Maui	33.92	0.76	1	2
USA	Ka'u	29.23	0.11	1	2
USA	Kona	30.12	0.52	1	46
USA	Moloka'i	32.63	0.10	1	2
USA	Waialua, Oahu	32.95	0.12	1	2
Vietnam	Central Highlands	23.31	0.11	1	2
Yemen	Bura'a, Western Highlands	39.24	0.10	1	2
Yemen	N/A	41.15	0.52	1	14

3 | RESULTS

3.1 | Cellulose homogeneity

Three α-cellulose samples extracted from different coffees were analyzed repeatedly to assess the variability of isotopic measurements within a given sample; the standard deviation of the oxygen isotope ratio for each of the three samples was ~0.5‰ (n = 46, 14, and 13). The variability of isotopic measurements across extraction batches was assessed by extracting a selected sample in three separate batches. The standard deviation of the mean δ^{18}O value among batches (σ = 0.11) was within the measurement uncertainty, indicating that the extraction procedure was consistent over time.

3.2 | Oxygen isotope ratios of coffee bean α-cellulose

The δ^{18}O values of analyzed coffee bean cellulose ranged from approximately +22‰ to +42‰ (Table 1). Coffees grown within the same country or region tended to have similar oxygen isotope ratios (Figure 1). In particular, African coffees were highly enriched in ^{18}O relative to coffees from other parts of the world. The range of values between farms approached 7‰ for samples grown in the USA (Hawaii), El Salvador, Guatemala, and Colombia, indicating that fairly large regional differences in coffee cellulose δ^{18}O values may exist within some countries.

3.3 | Modeled oxygen isotope ratios of coffee stem and leaf α-cellulose

Stem and leaf cellulose δ^{18}O values were modeled using relative humidity, temperature, and source water δ^{18}O values for each sampled region. When aggregated by country, the measured seed cellulose δ^{18}O values tended to fall between the modeled stem and leaf cellulose values (Figure 2). The mean δ^{18}O values of samples from Peru, Papua New Guinea, and Mexico were slightly lower than those of the modeled stem cellulose, but were within 1σ of the country's modeled stem values. A single sample from Papua New Guinea had δ^{18}O values more than 1σ lower than the modeled stem values. Under climate conditions that do not strongly drive evaporative enrichment of coffee bean water, the δ^{18}O value of seed cellulose would be close to that of stem cellulose. The differences between the measured seed cellulose δ^{18}O values and the modeled stem cellulose δ^{18}O values ranged from approximately −9‰ (Colombia) to +3‰ (Peru), with a mean difference of −3‰. Half of the samples with a known region of origin had a modeled mean leaf oxygen isotope ratio, indicating that the leaf that was less enriched in ^{18}O than the seed tissue, and nine seed samples were ^{18}O-enriched by more than 1σ over the modeled leaf value.

3.4 | Model of isotopic enrichment in coffee bean water

We calculated the δ^{18}O values of coffee bean water for the analyzed samples using the established relationship between cellulose δ^{18}O values and the δ^{18}O values of water at the site of cellulose synthesis

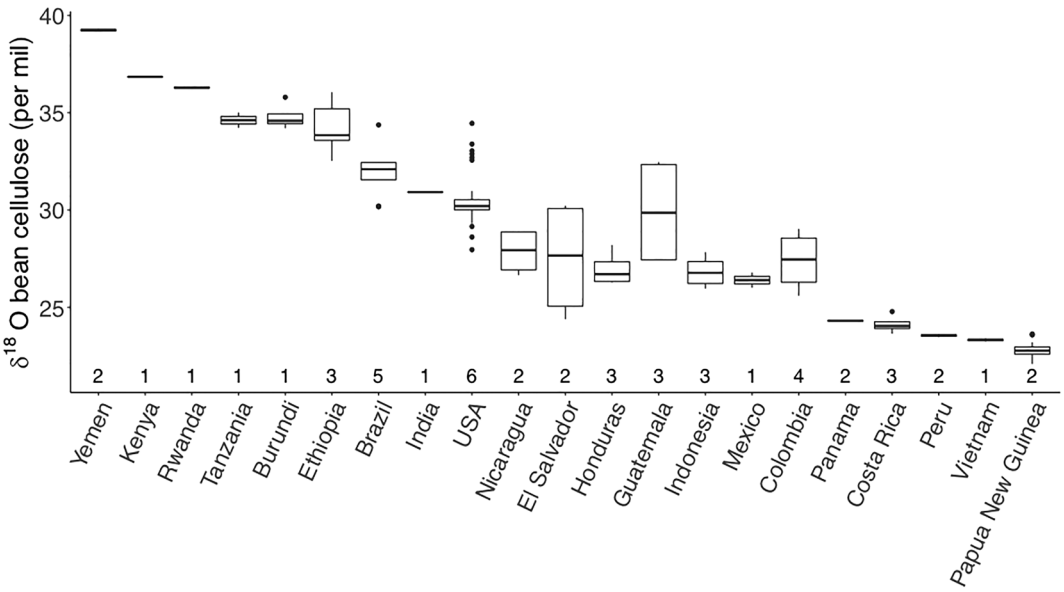

FIGURE 1 Measured δ^{18}O values of coffee α-cellulose vary by country of origin, ranging from approximately +22‰ to +42‰. Numbers along the x-axis indicate the number of farms or cooperatives sampled in each country (see Table 1 for the number of replicates from each region). Box edges represent the 1st and 3rd quartiles, center lines represent the median values, and whiskers represent ±1.5 IQR from the 1st and 3rd quartiles

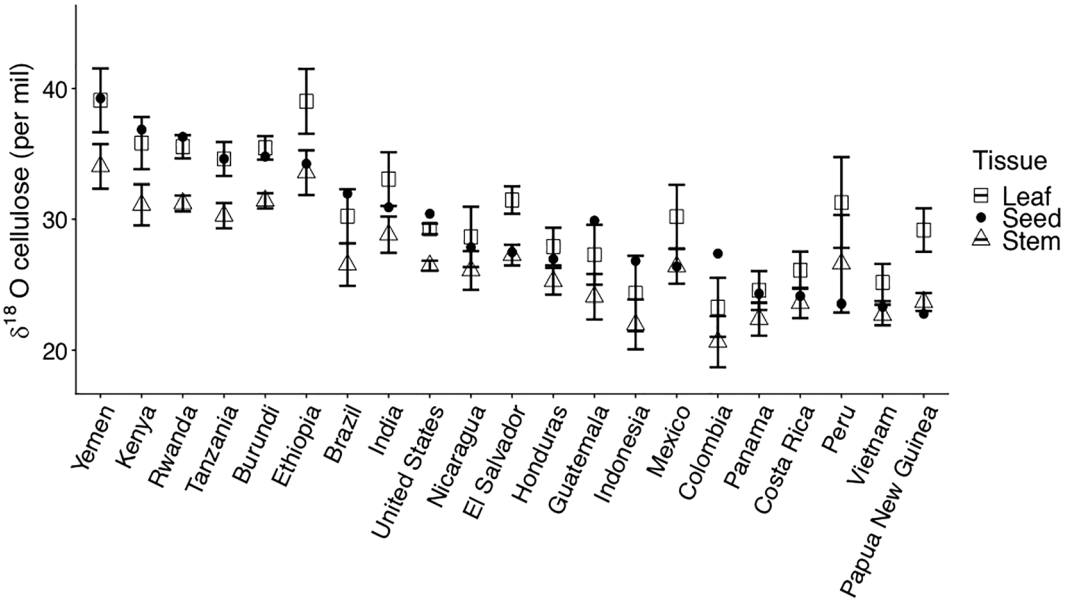

FIGURE 2 The δ^{18}O values of leaf and stem α-cellulose were modeled at a 1° resolution for coffee-growing regions using local climate and precipitation δ^{18}O values, and the mean and standard deviations of the modeled values were calculated for each country. Mean measured values of coffee bean α-cellulose δ^{18}O (filled circles) are presented alongside the mean modeled values of leaf and stem α-cellulose δ^{18}O for each country (squares and triangles, respectively). Error bars show ±1σ from the modeled values for each country

(Equation 1). We empirically modeled the calculated δ^{18}O values of coffee bean water ($\delta^{18}O_{sw}$) as a function of the δ^{18}O values of precipitation ($\delta^{18}O_s$; $p < 0.01$), relative humidity (rh; $p < 0.01$), and temperature (t; $p = 0.014$) using multiple linear regression (Equation 2, adj. $R^2 = 0.37$).

$$\delta^{18}O_{sw} = 1.15 \bullet \delta^{18}O_s + 0.56rh - 0.99t - 11.76 \quad (2)$$

Surprisingly, we found a negative correlation between the δ^{18}O value of coffee bean water and temperature (Pearson's $r = -0.39$, $p = 0.014$). This relationship is the opposite of the expected pattern but could be explained in part by correlation between some of the independent variables in the regression model. Among the samples that we analyzed, the δ^{18}O value of precipitation is negatively correlated with regional temperature (Pearson's $r = -0.37$, $p = 0.02$). This relationship is different from global trends and appears to be driven largely by coffee-growing regions of Yemen, Ethiopia, Burundi, Tanzania, and Kenya, which have high δ^{18}O values of precipitation but low temperatures relative to other coffee-growing regions. While multicollinearity complicates interpretation of the slope values, the observed multicollinearity in the regression equation is not severe enough to warrant the removal of independent variables according to analysis of variance inflation factors (VIF)[46] (VIF = 1.25 for $\delta^{18}O_s$ values, 1.18 for *t*, and 1.21 for *rh*).

3.5 | Isoscape of δ^{18}O values of coffee bean α-cellulose

The calculated regression equation for predicted coffee bean water was incorporated into the cellulose isotope model developed by Roden et al[23] and used in conjunction with climate and precipitation δ^{18}O data to model likely oxygen isotope ratios of bean cellulose across coffee growing regions (Figure 3). The modeled values of coffee bean cellulose δ^{18}O ranged from +21.6‰ to +39.9‰, which is consistent with the range of measured values.

To evaluate the potential impact of interannual climate variability on the modeled results, we predicted the δ^{18}O values of coffee cellulose for each year from 2011 to 2018 using annual average climate data. We then subtracted the minimum δ^{18}O value from the maximum δ^{18}O value in this time range for each location, producing a map of the magnitude of the impact of interannual climate variability on the modeled δ^{18}O values of coffee cellulose (Figure 4). We found that the difference between the highest and the lowest predicted δ^{18}O values in a given location ranged from <0.1‰ to 0.7‰, with an average of 0.3‰.

3.6 | Model evaluation

To assess the accuracy of the model, we compared the measured δ^{18}O value of each analyzed coffee cellulose sample with the mean modeled value for its region of origin (Figure 5; y = 1.06x −1.98; adj. $r^2 = 0.74$, $p < 0.01$). For samples without a more specific known region of origin, the mean value for the country of origin was used. The model predicted the δ^{18}O values of all coffee cellulose samples within ±4‰; the mean absolute error was ±2.0‰, and the standard deviation of the residuals was 2.3‰.

4 | DISCUSSION

The oxygen isotope ratio of coffee bean α-cellulose integrates local climate and precipitation δ^{18}O signals and can therefore be used as a predictable indicator of the origin of roasted coffee beans. By

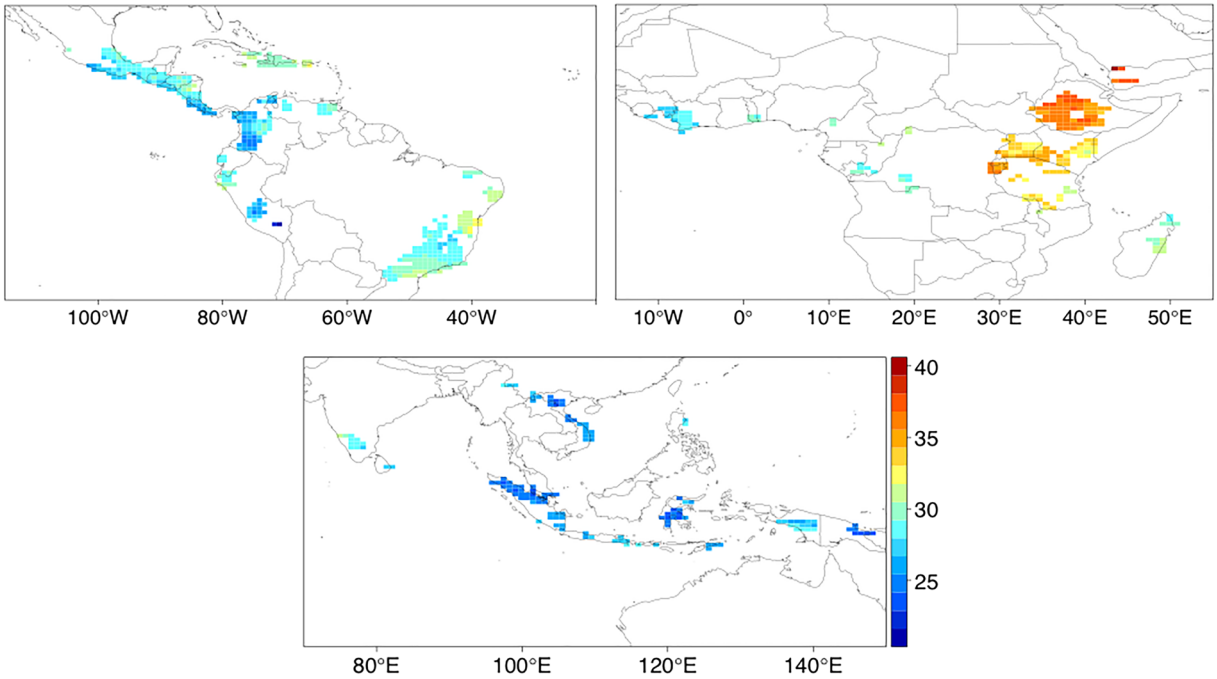

FIGURE 3 Map of predicted $\delta^{18}O$ (‰) values of coffee bean cellulose for coffee-producing regions. Values were calculated using local climate and precipitation $\delta^{18}O$ data, the derived model of the $\delta^{18}O$ values of coffee bean water, and the known fractionations associated with cellulose synthesis[23] [Color figure can be viewed at wileyonlinelibrary.com]

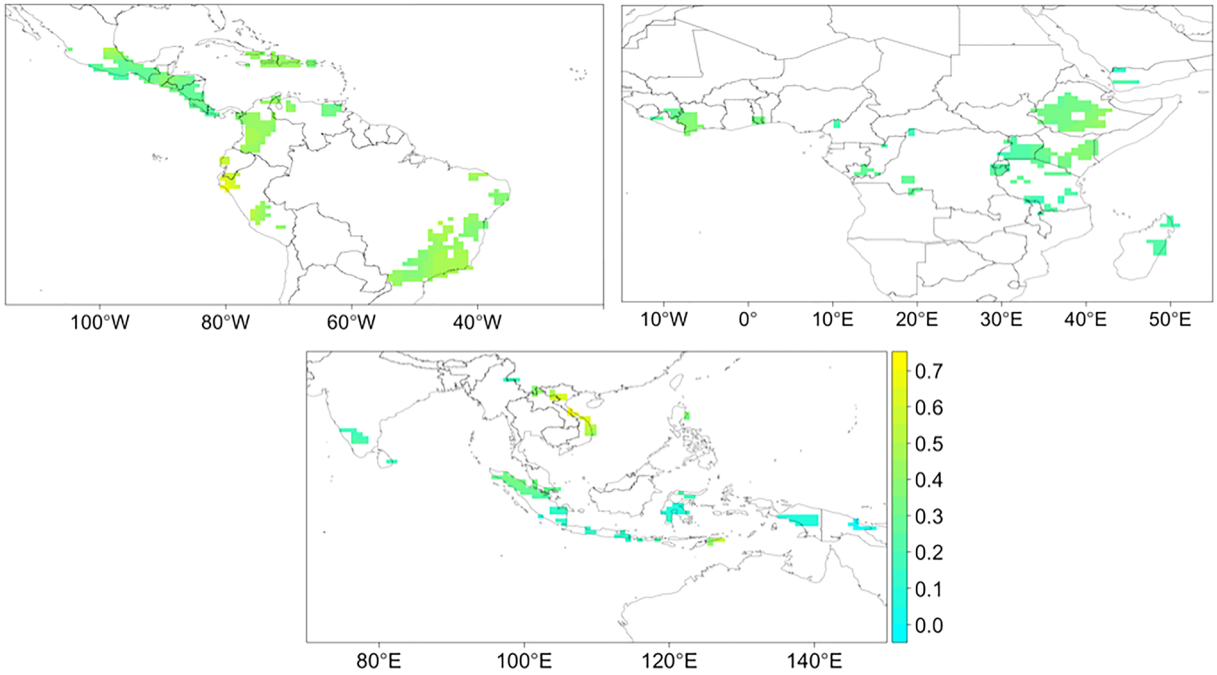

FIGURE 4 Map of interannual variability in predicted $\delta^{18}O$ values of coffee bean cellulose due to interannual climatic variation. Predicted $\delta^{18}O$ values of coffee bean cellulose were calculated using annual average climate data for each year from 2011 to 2018. For each grid cell, the minimum predicted $\delta^{18}O$ value was subtracted from the maximum $\delta^{18}O$ value [Color figure can be viewed at wileyonlinelibrary.com]

analyzing cellulose extracted from roasted beans rather than bulk tissue from green beans, we circumvent limitations of previous studies that examined spatial trends in the $\delta^{18}O$ values of green coffee beans.[11-13] Previous studies using isotopes as an indicator of coffee origin have focused primarily on the analysis of green coffee beans, although the vast majority of coffee sold to consumers is roasted. Santato et al[16] reported $\delta^{18}O$ values of bulk green coffee ranging from 19‰ to 37‰ and found generally lower values in

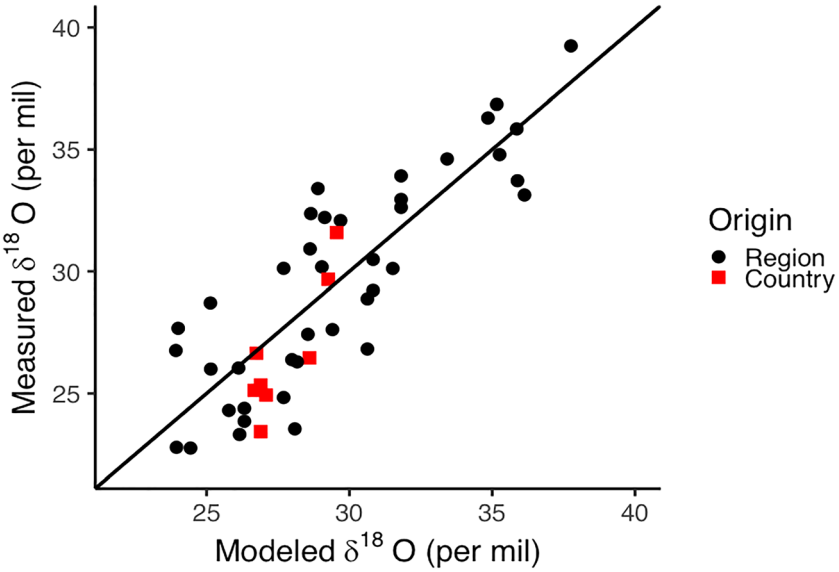

FIGURE 5 Measured δ^{18}O values of coffee bean α-cellulose for all samples compared with the mean modeled δ^{18}O value for the sample's region of origin (y = 1.06x − 1.98; the slope was not significantly different from 1 and the intercept not significantly different from 0). Values of samples with only a known country of origin (red squares) are compared with the mean modeled value for the country; these samples were not used to fit the regression model of seed water δ^{18}O values (Equation 2). Samples with a known region of origin (black circles) were used to fit the regression model. The solid black line represents a 1:1 relationship [Color figure can be viewed at wileyonlinelibrary.com]

Central America and higher values in Africa. Rodrigues et al[11] reported δ^{18}O values between 18.7‰ (from Papua New Guinea) and 34.8‰ (from Ethiopia). We observed a very similar ranking of countries from which coffees with relatively low and high δ^{18}O values originate. The δ^{18}O values of roasted coffee cellulose measured in this study were slightly higher than those of bulk coffees reported in previous studies. This is in part due to the inclusion of a sample from Yemen with a very high δ^{18}O value, and also partly due to cellulose typically having a higher δ^{18}O value than other constituents of plant tissue.[20]

Unlike the analysis of bulk green coffee beans, the analysis of roasted coffee bean cellulose allows for verification of the product at the point-of-sale based on a mechanistic model of spatial variability. If analysis of the product at the point-of-sale is of paramount importance and if oxygen isotopes are used to verify supply chain integrity, we must recognize that roasting introduces an uncontrolled source of variability in bulk coffee bean δ^{18}O values by changing the chemical composition of the bean (e.g., volatilization of sugars, etc.). Carter et al[17] analyzed bulk tissue of roasted coffee and reported a similar range of δ^{18}O values to those found in studies of green beans (20.6 to 36.7‰). That study assumed that all coffees were heated to the same temperature, and therefore were similarly affected by roasting. Cellulose is unaffected by the roasting process and thus the δ^{18}O values of cellulose are robust to differential roasting practices.

Prior research has established that, for certain geographic subsets of coffee, the δ^{18}O value of bulk green coffee is variously related to altitude, temperature, mean annual precipitation, distance from the coast, latitude, and the δ^{18}O value of precipitation.[11-13] However, patterns of δ^{18}O values in bulk coffee tissue on a global scale have been difficult to identify.[11] Recognizing the relationships between the oxygen isotope ratios of cellulose and climate drivers, we were able to successfully predict the δ^{18}O values of coffee seed cellulose across all coffee-producing regions. A typical stable isotope based approach to authenticate the origin of food products depends on comparing an unknown sample with a reference set of known origin.[6] While this is an effective approach, it is limited to comparisons with samples of known origin and may not include samples from all possible growing regions. The approach presented here reduces the need for an extensive reference dataset by predicting expected coffee cellulose δ^{18}O values based on global climate and precipitation data.

There are a number of sources of variability that are not included in the model and could be constrained by further research. Potential sources of model uncertainty include intra- and interannual variability in climate, source water, and atmospheric water vapor isotope ratios, and the potential contributions of coffee bean photosynthate, as opposed to leaf photosynthate, to cellulose synthesis. While the model was developed using average temperature and relative humidity over the 2015–2018 period for relatively large regions, it neglects the influences of spatial and seasonal variability within a region. However, we found that the impact of interannual climate variability within most regions is relatively small (0.3‰ on average) over the 2011–2018 period, suggesting that the model is relatively robust to typical year-to-year changes in climate and therefore is useful even if the growing year of the sample is unknown. A further complication in parameterizing microclimate is that some high-value coffees are shade-grown,[47] significantly altering growth conditions relative to the regional average. Carbon isotope ratios may be helpful in determining whether a sample was shade- or sun-grown,[48] and climatic model inputs could be adjusted for shade-grown plants. We also assumed that the δ^{18}O value of the precipitation was identical to that of the plant source water, but plants may access surface or ground water in addition to precipitation, particularly on irrigated farms. Finally, the δ^{18}O value of atmospheric water vapor in humid environments has a large effect on modeled cellulose δ^{18}O values, and the oxygen isotopic composition of atmospheric water vapor can deviate substantially from equilibrium over the short term.[49] Reassessing the model of coffee bean water ^{18}O enrichment under controlled

conditions may provide a much clearer picture of the relationship between coffee bean water oxygen isotope ratios and climate. In addition, hydrogen isotopes of wood cellulose have been modeled as a function of local climate and source water $\delta^{18}O$ values,[23] and adaptation of the cellulose hydrogen isotope model for application to coffee beans may provide an additional predictive tool for identifying coffee origin. Despite these remaining sources of uncertainty, the model of coffee bean water enrichment can be used to estimate the oxygen isotope composition of coffee bean α-cellulose to within approximately ±2.3‰ based on local climate and the ^{18}O composition of precipitation.

A number of measurements, including those of oxygen, hydrogen, carbon, nitrogen, and strontium isotopes and trace element concentrations, can serve as useful discriminators of coffee origins. Rather than considering oxygen isotope ratios of cellulose to be the sole indicator of coffee origin, we suggest that our model of cellulose oxygen isotopes can be used to limit the scope of possible origins prior to the measurement of additional isotopic or trace element parameters. The oxygen isotope ratios of coffee bean α-cellulose can effectively discriminate between different regions of origin and could be used to assess the authenticity of coffees that may be fraudulently labeled. Using our model of cellulose oxygen isotopes in conjunction with other parameters that are indicative of origin will provide the most precise, accurate, and robust determination of coffee origin.

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