Isotopic analysis

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Radiocarbon measurements

21 individuals were dated at [14]CHRONO Center at Queen's University Belfast. The dates are shown in Table 1 and in Figure 1, along with the dates for MN00015 (Bot15) and MN00017 (Bot17) presented in Malaspinas et al. (2014). The calibrated dates are between 23 BP and 2035 BP for the Botocudo individuals, and 1080 BP and 2035 BP for the Native American and the Sambaqui individual, respectively.

Table 1: Table 1

ID	Material type	Age (BP)	SD	Details
MN00119	tooth, 1st lower right premolar	23	22	Botocudo_Doce
MN00316	tooth, 1st upper right molar, child (broken)	40	30	Botocudo_Aimores
MN00118	tooth, 3rd upper right molar	48	33	Botocudo_Doce
MN00021	tooth, 1st upper right molar	66	50	Botocudo_Mutum
MN00346	tooth, 2nd lower right premolar	76	30	Botocudo_Unknown(Botocudo)
MN00069	tooth, upper right canine	79	39	Botocudo_Mutum
MN00010	tooth, 1st upper right molar	84	22	Botocudo_Itamacuari
MN00039	tooth, 2nd upper right premolar	106	30	Botocudo_Mutum
MN00068	tooth, 2nd upper right molar	121	33	Botocudo_Mutum
MN00023	tooth, 2nd upper left premolar	137	25	Botocudo_Poxixa_Mutum
MN00022	teeth (2), 1st upper right molar(deciduous and permanent)	145	30	Botocudo_Mutum
MN00013	tooth, 1st lower right molar	150	40	Botocudo_Mutum
MN0003	tooth, 1st upper left molar	160	26	Botocudo_Mutum
MN00056	tooth, 3rd lower right molar	169	25	Botocudo_Poté
MN00067	tooth, 2nd upper left molar	170	21	Botocudo_Minas
MN0009	tooth, 2nd upper righ molar	185	29	Botocudo_Mucuri
MN00064	tooth, 2nd upper right premolar	199	41	Botocudo_Doce
MN00045	tooth, 1st upper left molar (child)	249	29	Botocudo_Mutum
MN00016	tooth, 2nd upper right molar	327	24	Botocudo_Itapemirim
Bot15	Bot15	417	25	Bot15
Bot17	Bot17	487	25	Bot17

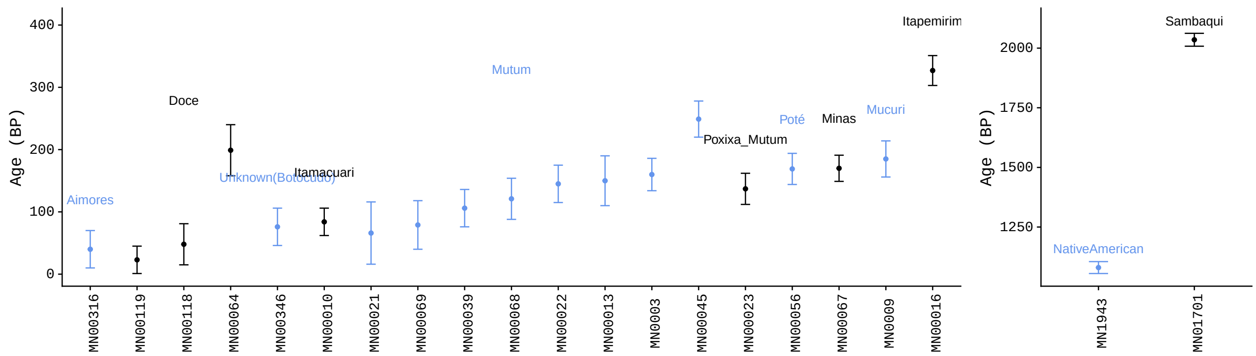


Figure 1: Uncalibrated dates. The bars represent one standard deviation.

ID	Material type	Age (BP)	SD	Details
MN1943	tooth, 3rd lower right molar	1080	25	NativeAmerican_MorrodaBabil
MN01701	tooth, 2nd lower left molar	2035	27	Sambaqui_Cabecuda

δ^{13}C and δ^{15}N measurements

We measured the δ^{13}C and δ^{15}N values for the 21 individuals. These values can give us an insight into the dietary habits for a given organism.

However, we need to be careful when examining children's teeth.

The decidious teeth ('baby teeth') develop during the embryonic stage of development, and the isotopic measurements reflect the dietary habits of the child's mother.

Moreover, the crown of the first molars are formed during the early years/breastfeeding period. In some instances, a child's first molar can have enriched ^{15}N values due to breastfeeding, indicating that a significant proportion of the dentine was formed at that time (Schroeder et al. 2009). In order to know more about an individual's dietary habits, it is better to date second and third molars.

In Figure 2, we show whether the samples analyzed belonged to a child, and whether the tooth was a first molar. Among the samples, we observe two children's first molars (from MN00022 and MN00045) that could have shifted δ^{15}N values due to breastfeeding. We have to keep in mind this information for the next steps, in which we will infer dietary proportions.

Mixing models for stable isotopic data

We used SIAR (Stable Isotope Analysis in R, (Parnell and Jackson 2013)) in order **to infer dietary proportions** for the individuals.

Model

SIAR fits a model via Markov chain Monte Carlo. The parameters to fit are the **dietary proportions**.

Keypoints of the model:

- a consumer (e.g., a Native American) integrates isotopes from one or more sources (e.g., marine mammals, marine fish, terrestrial fauna, etc.)
- we have known values of the trophic enrichment factors (TEF), which indicate the difference in the isotopic ratio between a consumer and their diet
- a consumer incorporates an isotope proportionally to the dietary proportion of a given source, its TEF and its isotope value
- for a consumer, the observed isotope value is explained by the sum of the dietary proportions

An advantages of SIAR package is that it has a robust estimation of the parameters even when the variation within groups is large.

We used the default Dirichlet prior distribution for the parameters. This distribution allows us to treat source data independently; e.g., the estimated dietary proportion for source X does not depend on the estimated proportion for source Y. All inferred dietary proportions must sum to one.

We fitted a model per individual. As suggested in the manual, we set 500,000 iterations and discarded the first 50,000 iterations.

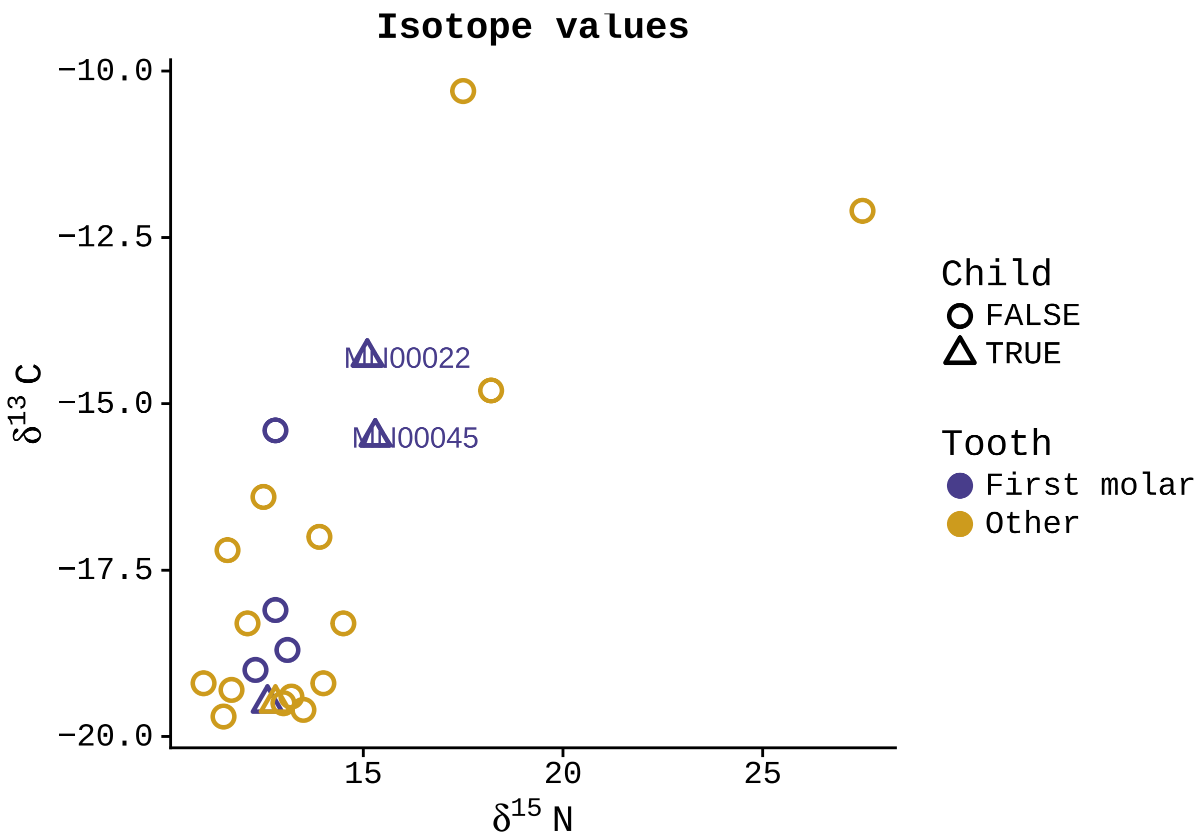


Figure 2: Isotope values. The triangles represent children and the circles represent adults. The color indicates whether we examined a first molar (purple) or any other type of tooth (golden).

Results

Raw data

The squares represent the mean isotope values for a consumer whose diet was based on a specific source (terrestrial fauna, marine fish or marine mammals).

The coordinates of the squares were obtained as follows: The TEF values represent the expected difference between the consumer and source isotope values. Thus, we summed the mean TEF values and the mean isotope values of a source in order to get the mean isotope values for a (theoretical) consumer whose diet consisted on the source.

The dashed lines represent two standard errors of the mean isotope values.

The solid circles represent the pair of isotope values reported by the CHRONO Centre per individual. The triangles correspond to the isotope values for Bot15 and Bot17 (from Malaspinas et al., 2014), dated at the AMS 14C Centre at Aarhus University

We observe that for the Cabeçuda individual (or Sambaqui) the isotope values correspond to a marine diet, in agreement with the origin of the individual.

The Native American from Morro da Babilonia shows an unusual high value for d15N. We need to contact the ^{14}CHRONO Centre in order to get more information.

Regarding the Botocudo individuals, most of the samples group close to the expected values for a diet based on terrestrial fauna, with some of them deviating from those values.

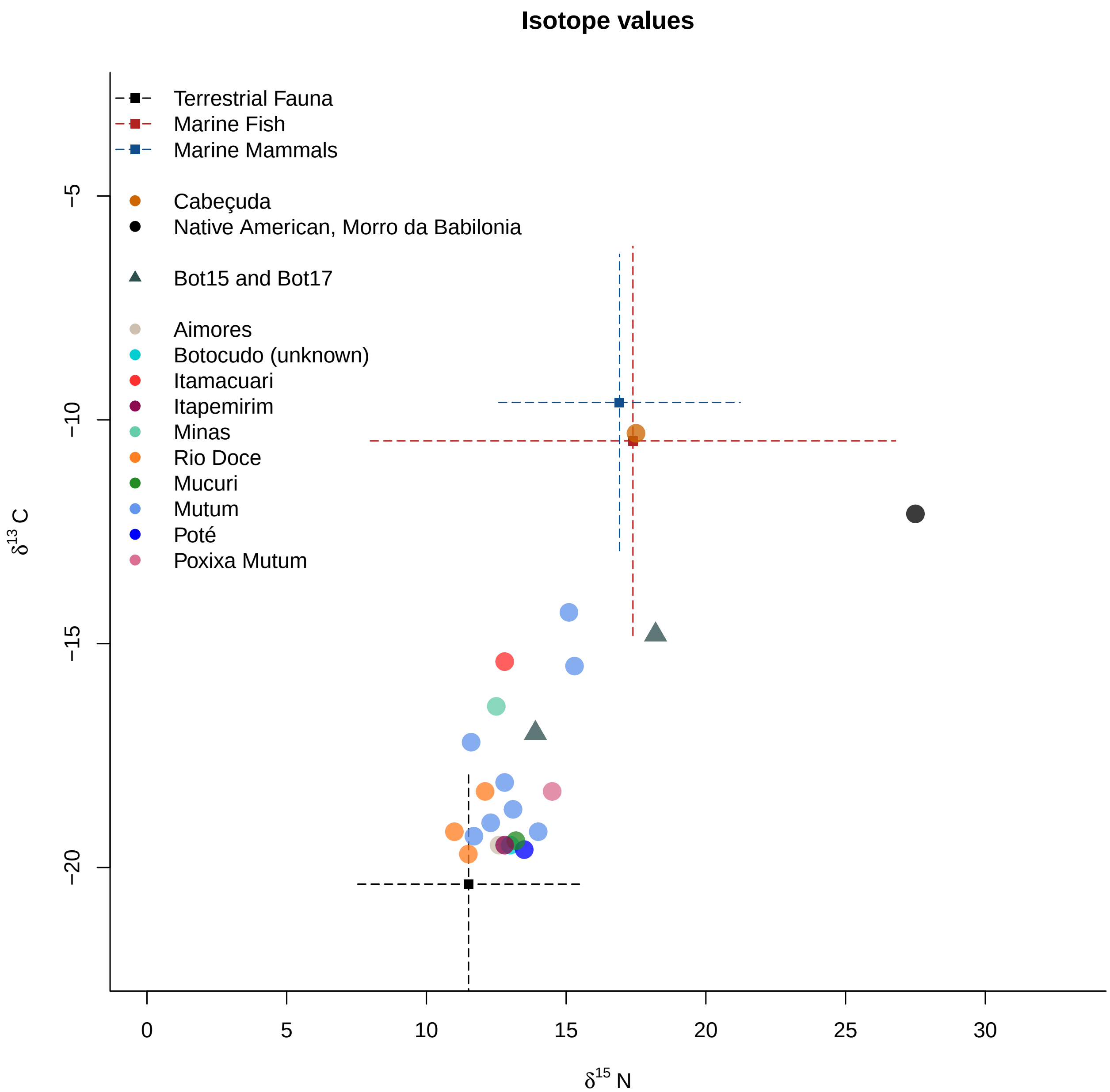
Interestingly, the Polynesian individuals (Bot15 and Bot17) fall outside of the expected isotope values for a marine diet (delimited by the ellipse).

Inferred dietary proportions

Densities

Diagnostic matrix plot

High correlations indicate that the program has problems to distinguish between the dietary proportions of two sources. Thus, as expected, the draws for marine fish and marine mammals tend to be highly correlated, as their isotope values are similar.



Isotope values
Terrestrial Fauna
Marine Fish
Marine Mammals
Cabeçuda
Native American, Morro da Babilonia
Bot15 and Bot17
Aimores
Botocudo (unknown)
Itamacuari
Itapemirim
Minas
Rio Doce
Mucuri
Mutum
Poté
Poxixa Mutum
−5
−10
−15
−20
δ13 C
0
5
10
15
20
25
30
δ15 N

Figure 3:

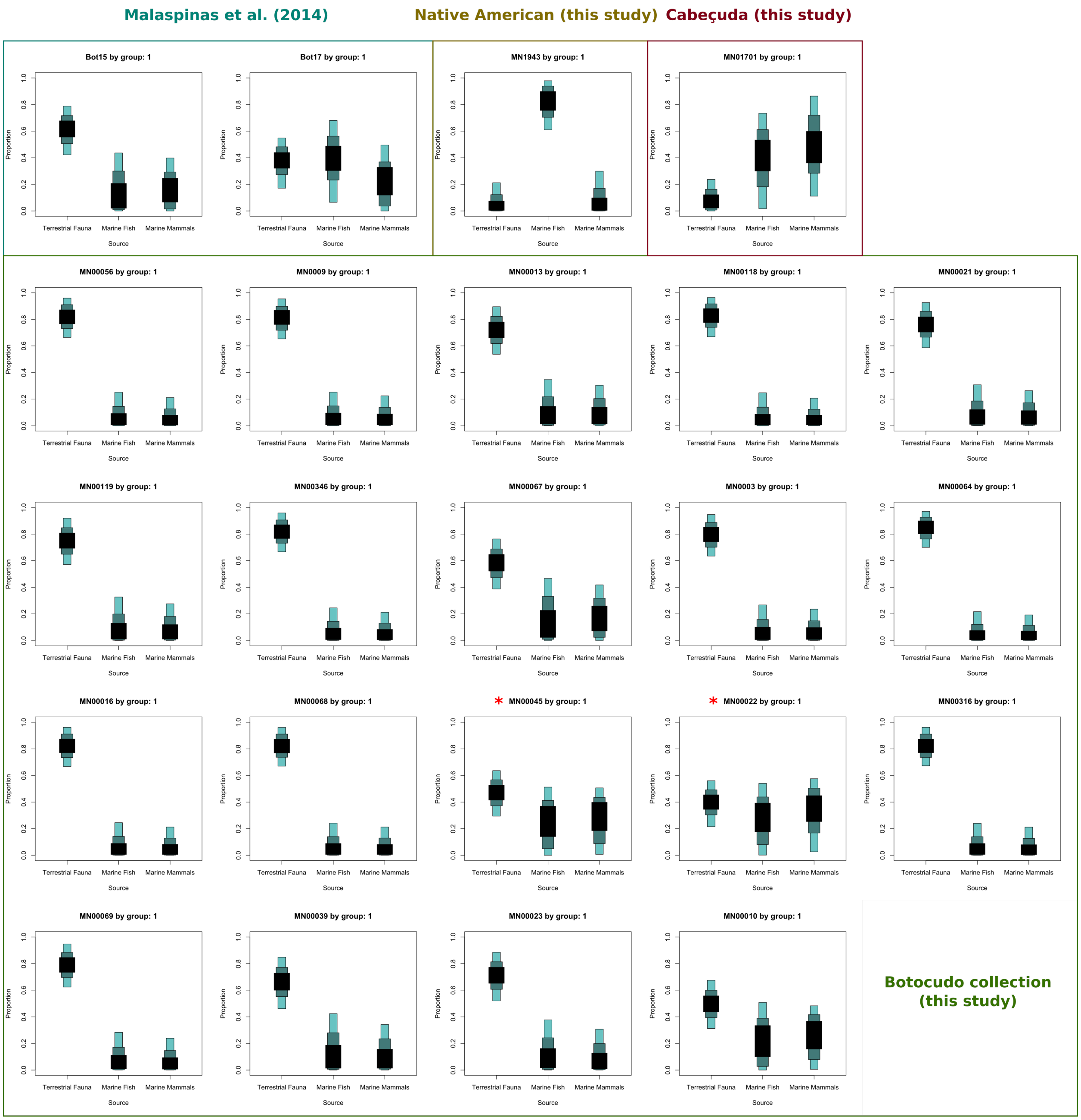


Figure 4: Dietary proportion (boxplots)

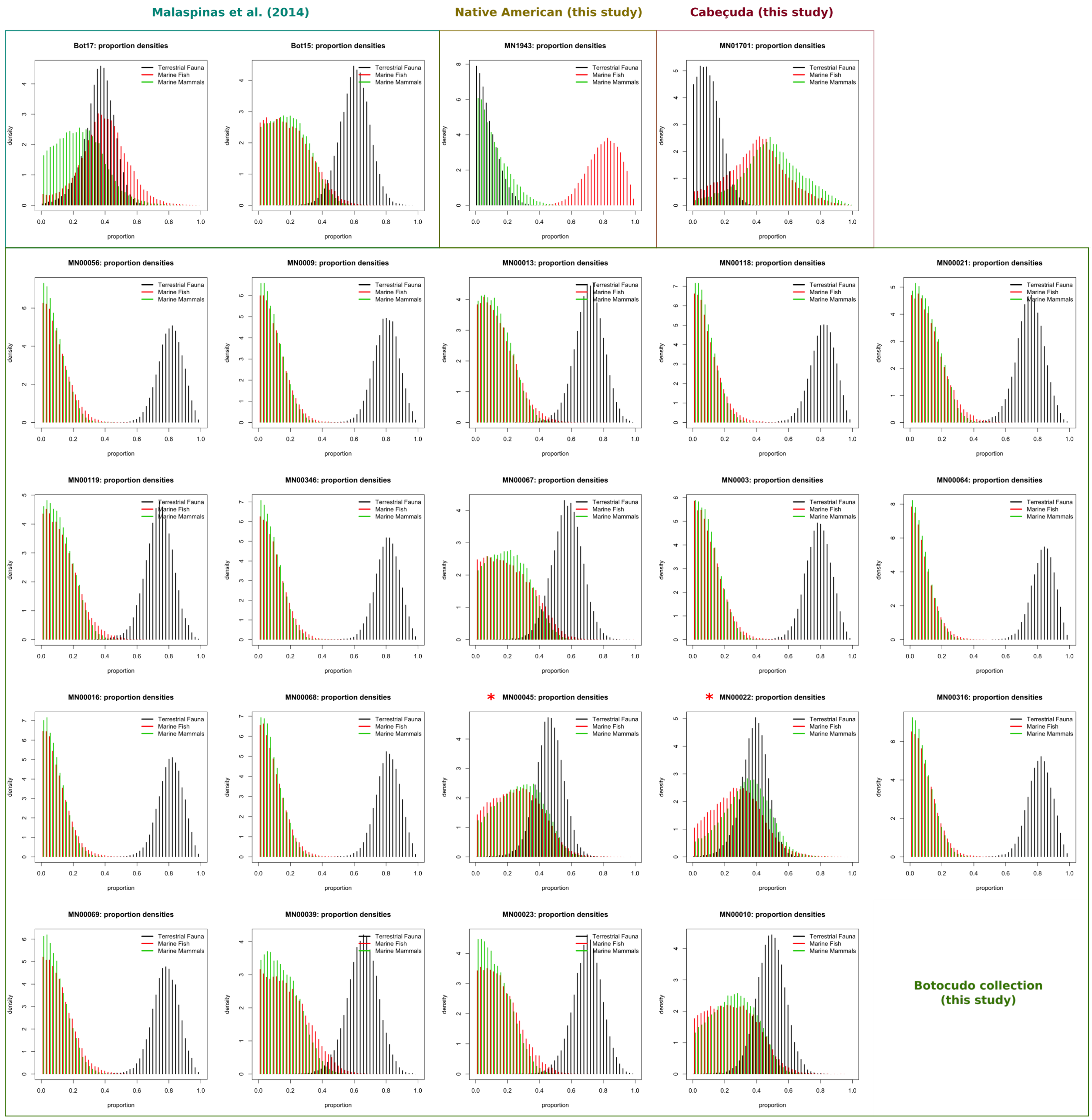


Figure 5: Dietary proportion (densities)

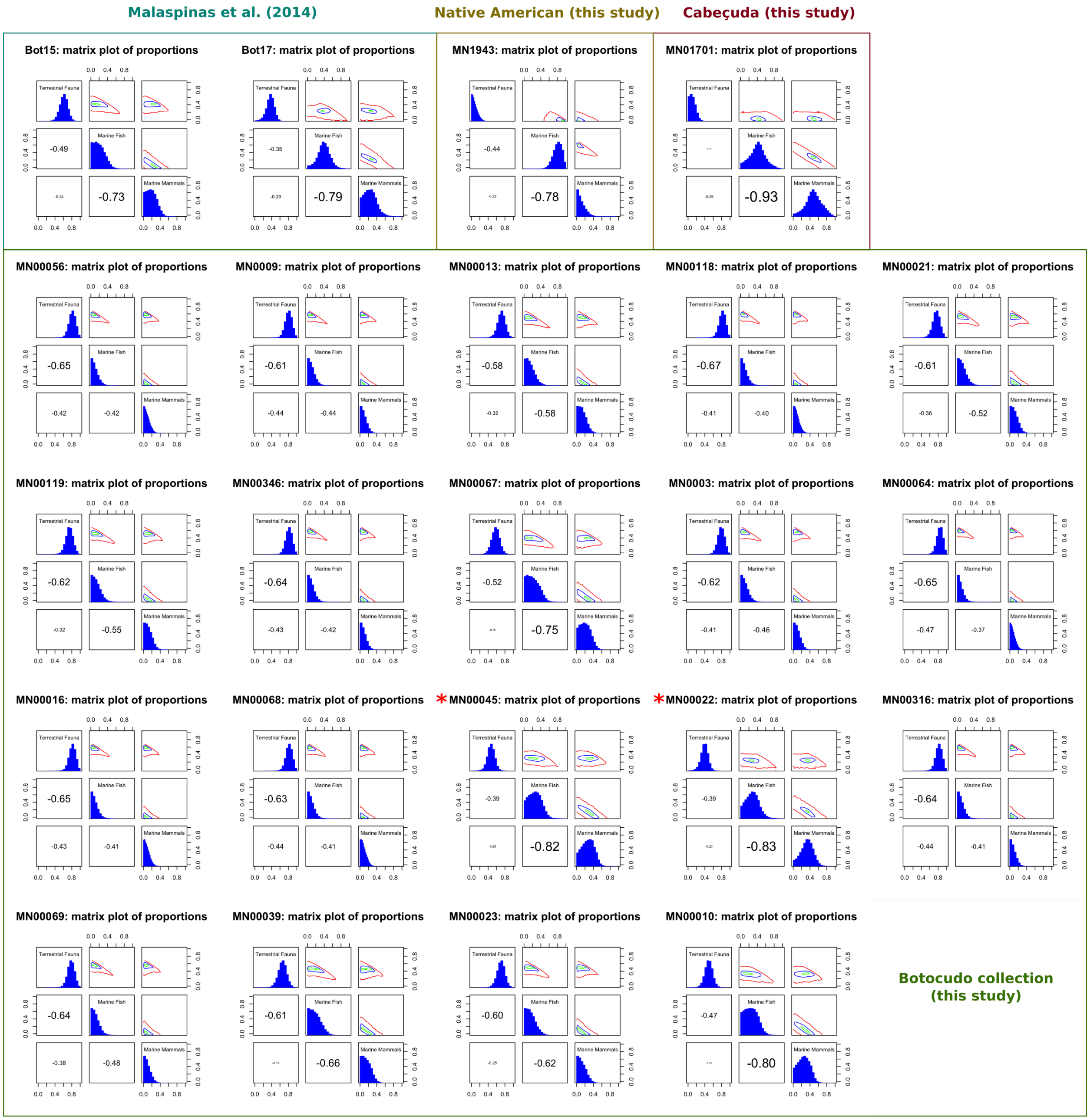


Figure 6: Dietary proportions (histograms and correlations). For every individual, we have a matrix with nine entries: The histograms of the estimated proportions are located on the diagonal; on the upper entries, the contour plots depict whether the posterior distributions are correlated; the lower entries contain the values corresponding to the correlation coefficient for pairs of distribution.

Traces

We saved the sampled values for 30,000 out of 500,000 draws for every estimated dietary proportion per individ-

ual.

Calibrated dates

Finally, we calibrated the radiocarbon results using a Southern Hemisphere calibration curve (ShCal13, (Reimer et al. 2013)) for all individuals.

Additionally, for Bot15, Bot17 and MN01701 (Cabeçuda from Sambaqui), we corrected for a marine reservoir effect, according to the estimated carbon uptake from a marine diet. The estimated values are presented in Table 2, along with the parameters reported in Malaspinas et al. (2014) for Bot15 and Bot17.

Table 2: Table 2. Calibration parameters for individuals with possible marine diets.

	Malaspinas et al. (2014)	This study
Marine carbon protein (Bot15)	30±16%	39±18%
Marine carbon protein (Bot17)	60±16%	63±18%
Marine carbon protein (MN01701)	NA	89±14%
Atmospheric calibration curve	ShCal04 (Board 2004)	ShCal13 (Reimer et al. 2013)
Marine calibration curve	Marine09 (Reimer et al. 2009)	Marine13 (Reimer et al. 2013)
ΔR offset	0	33±24

We used the OxCal programme, (version 4.3, (Bronk Ramsey 2009)) to calibrate the radiocarbon results. The figures below represent the posterior density for the ^{14}C dates. We show in parentheses the date and standard deviation reported by ^{14}CHRONO Centre, as well as the agreement indices (A) and convergence integral (C) returned by OxCal.

The agreement indices are a measure of the agreement between the model (prior) and the observational data (likelihood); this value should be over 60%.

The convergence integral is a test of the effectiveness of the MCMC algorithm; this value should be above 95%.

The horizontal brackets denote the regions associated to the 96% highest posterior density. The circles indicate the estimated mean and the bars represent one standard deviation.

ID	Terrestrial Fauna	Marine Fish	Marine Mammals	Total marine
MN00316	87±15%	10±14%	9±12%	13±17%
MN00118	82±15%	10±15%	9±12%	18±15%
MN00056	81±15%	10±15%	9±12%	19±15%%
MN00346	81±15%	10±15%	9±12%	19±15%
MN0009	80±15%	10±15%	9±13%	20±15%
MN0003	79±16%	11±16%	10±14%	21±16%
MN00069	78±17%	12±16%	10±14%	22±17%
MN00021	76±17%	13±18%	12±14%	24±17%
MN00119	74±17%	14±19%	12±16%	26±17%
MN00064	84±14%	9±13%	8±11%	26±14%
MN00013	72±18%	15±20%	14±16%	28±18%
MN00016	81±15%	10±14%	9±12%	29±15%
MN00068	81±15%	10±14%	9±12	29±15%
MN00023	70±19%	16±22%	13±18%	30±19%
MN00039	66±20%	19±23%	16±18%	34±20%
Bot15	61±20%	20±24%	19±21%	39±20%
MN00067	58±18%	22±25%	21±21%	42±18%
MN00010	49±19%	25±16%	25±25%	51±19%
MN00045	47±18%	26±25%	27±24%	53±18%
MN00022	39±17%	28±24%	33±25%	61±17%
Bot17	37±18%	39±32%	24±26%	63±18%
MN01701	11±13%	40±39%	49±38%	89±13%
MN1943	8±13%	80±20%	12±18%	92±13%

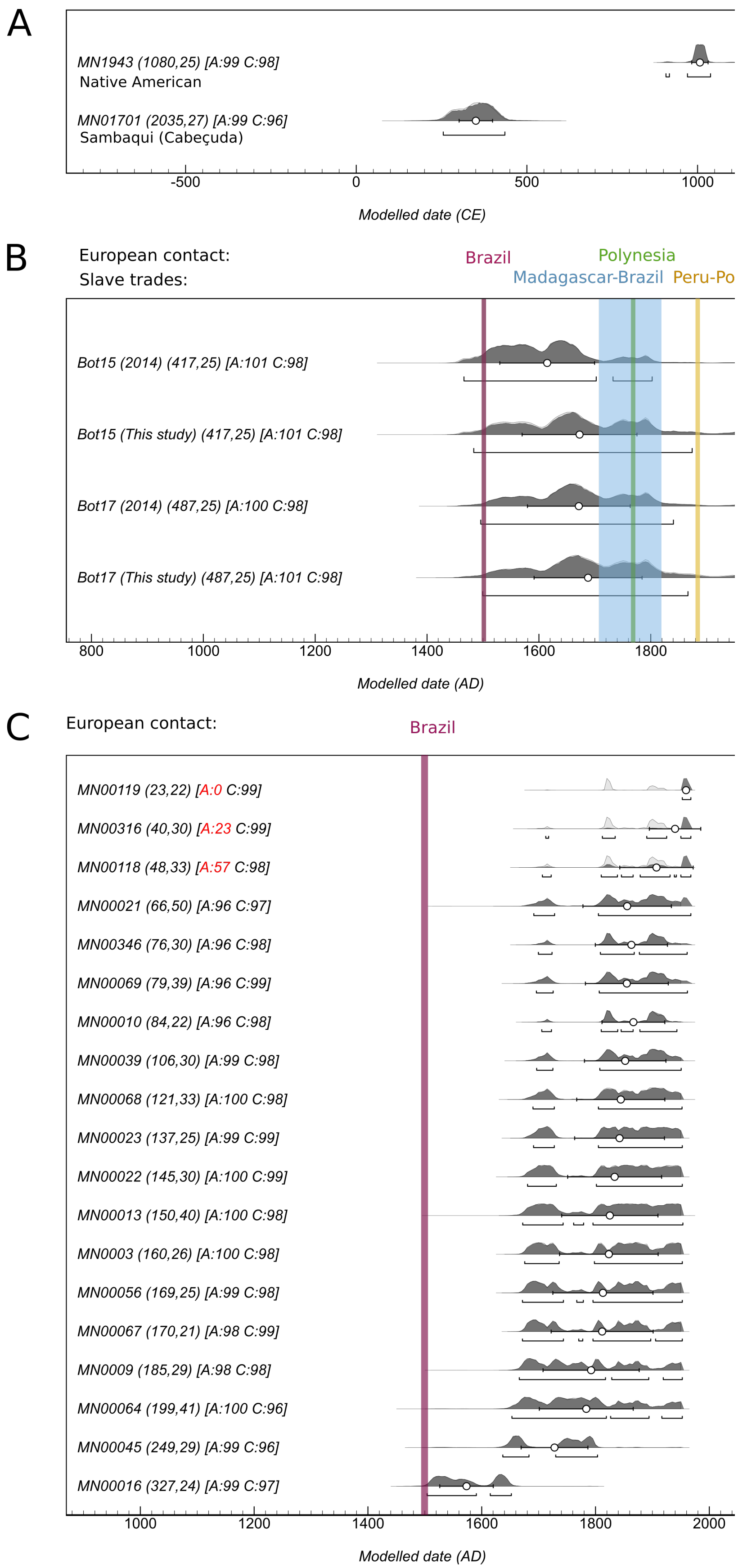


Figure 7: Calibrated dates for (A) MN1943 and MN01701, (B) Bot15 and Bot17 from Malaspinas et al. (2014) and from this study and (C) the Botocudo collection (this study only).

References

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