# Paleoecology Lab Standard Analysis

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## Introduction

Dr. Suzanne Birch is an anthropology professor and paleoecology lab director at the University of Georgia. Her lab has been studying the tooth enamel of various animal fossils since 2016 to gain a better understanding of these animals. Tooth enamel samples have been collected from archaeological dig sites around the world and analyzed for delta carbon 13 (δ13C) and delta oxygen 18 (δ18O) values, which are measurements of the ratio of the stable isotopes of each element. Lab standards, which are control samples from archaeological cow (AC), modern cow (MC), and archaeological elephant (AE) fossils, are run regularly to ensure the machines are running correctly and the lab protocols are being properly followed. The standards are treated with a bleach and acid solution. Dr. Birch assigned the task of analyzing her lab standards to answer three questions:

**1) Are the δ13C and δ18O values of the lab standards different from year to year?**

**2) Is there a relationship between the type of machine used (Gas Bench, PreCon) and δ13C and δ18O values?**

**3) Are the δ13C and δ18O values of the untreated standards significantly different from the treated standards?**

## δ13C/δ18O Values versus Year

The dot plots below visualize δ13C and δ18O values for each lab standard type against levels of year. The plots revealed significantly different variances for δ13C and δ18O values across level of year for each lab standard type, confirmed by the Levene test, violating the assumption of homoscedasticity. Hence, Welch's ANOVA test was used to check for differences in means between groups with unequal or unknown variances. Results showed that for five of the six combinations of lab standard type and δ13C/δ18O value, the mean value of δ13C or δ18O was not equal for at least one level of year. Tukey's HSD test was applied and the resulting p-values suggested 2018 was significantly different than most other years for almost every combination of δ13C/δ18O and lab standard type.

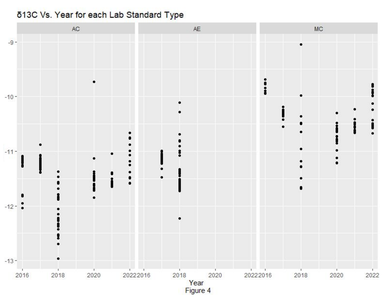


Figure 4

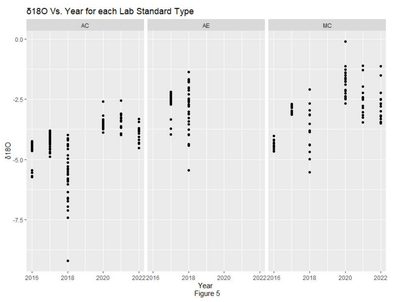


Figure 5

## δ13C/δ18O Values versus Year (Cont.)

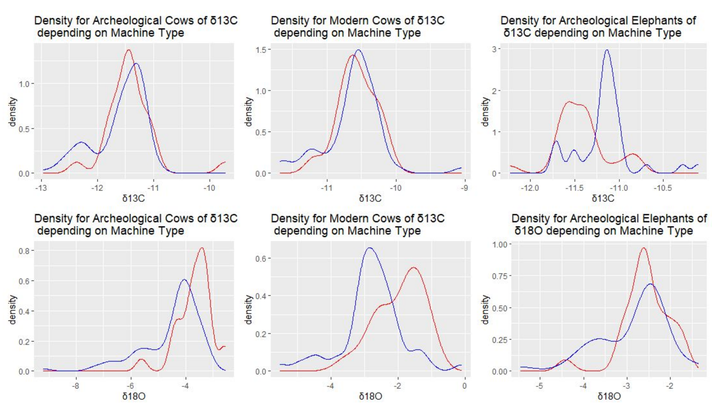
Dr. Birch was surprised by the differences in δ13C and δ18O values over the years and suspected that the change in lab and machines used in 2018 might have something to do with it. She requested an analysis of the 'machine' standards to verify if they followed the same pattern as her lab standards. We again ran Welch's ANOVA test on the difference in mean δ13C and δ18O values between year for the 'Fisher' and 'A1296' machine standards. The results of the tests showed that there was a significant difference in mean δ13C values between levels of year for both machine standard types, but not for δ18O values. This suggests an interaction between year and δ13C value, but not δ18O value.

Welch's ANOVA results for Year vs. δ13C and δ18O

| | AC | MC | AE | Fisher | A1296 |
|---|---|---|---|---|---|
| $\delta^{13}C$; F-value | 33.027 | 94.763 | 7.5607 | 3.6399 | 4.1003 |
| $\delta^{13}C$; p-value | <0.001 | <0.001 | 0.0089 | 0.0073 | 0.0038 |
| $\delta^{18}O$; F-value | 62.364 | 94.763 | 1.984 | 0.1301 | 0.1085 |
| $\delta^{18}O$; p-value | <0.001 | <0.001 | 0.1644 | 0.9847 | 0.9899 |

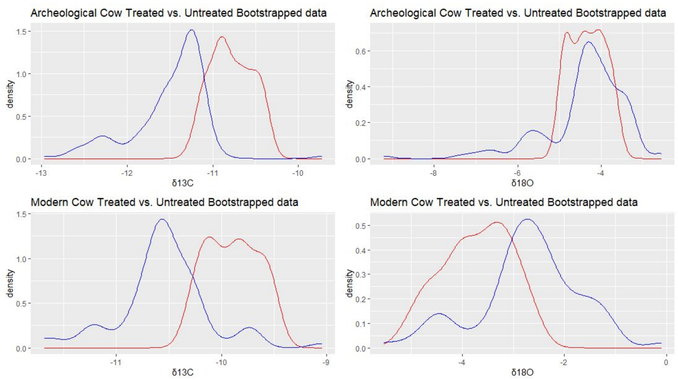
## δ13C/δ18O Values versus Machine Type

Welch's t-test was run on mean δ13C and δ18O values from the Gas Bench and PreCon machines across each lab standard type. Results showed that only certain combinations of lab standard types and δ13C or δ18O values had significant differences in mean values between machine types. Therefore, it is inconclusive whether machine type affects δ13C and δ18O values.




## δ13C/δ18O Values for Treated vs Untreated

Dr. Birch ran a batch of untreated lab standards for us to test if the treatment method affected δ13C or δ18O values. Bootstrapping was applied to the sample of untreated standards, as the n was relatively small. We used Welch's t-test to compare the means between the treated and untreated lab standards for each combination of lab standard, δ13C, and δ18O. The resulting p-values suggested a difference in mean δ13C /δ18O values between treated and untreated for every combination except AC and δ18O.




## Conclusions

Based on the analysis of Dr. Suzanne Birch's lab standards, we were able to answer three key questions that have implications for her larger study on stable isotopes in animal fossils.

First, we found that there is a significant difference in the δ13C and δ18O values of the lab standards from year to year. The year 2018 stands out as having significantly different δ13C and δ18O values than other years for almost every lab standard type. These results potentially undermine the assumed consistency and reliability of the lab protocols.

Second, we found there may be a potential relationship between the type of machine used and the δ13C and δ18O values, though the results varied based on combination of lab standard type and δ13C or δ18O. These results possibly suggest that she should only use one type of machine for her study.

Finally, we determined that there is a significant difference between the δ13C and δ18O values of treated and untreated standards, suggesting that the treatment process is likely validated in her study.