The Reconstruction of Ancient Diets and Environments

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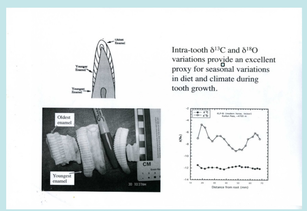
PURPOSE:

The purpose of the laboratory research is to determine the reconstruction of ancient diets and environments using stable carbon and oxygen isotopes in fossil teeth.

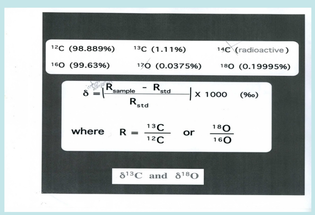
Analysis of the fossil tissues of ancients animals using carbon isotopes has enabled scientists to determine the food ancient animals consumed based upon their environment and time period. Since most living organisms spent a substantial amount of time trying to replenish lost energy, determining how this energy was obtained is crucial. This data enables us to discover the evolutionary changes that may have occurred due to dietary differences.

Morphologically, teeth are shaped to process the type of food the organism eats. Scientists now know that foods rich in hard fruits or grasses leave microscopic traces on teeth as well as distinct damage patterns on enamel surfaces. Yet, not all teeth adaptations lead to the actual behavior of the animal.

So scientists turned to another investigative approach probing for the information locked in the crystal structure of fossil tooth enamel. Tooth enamel is a crystalline mineral made up mostly of calcium and phosphate along with other small amounts of other ions, including carbonate , are present. These ions are then preserved in the tooth when it was forming. In addition, these ions provide development inside the enamel that have specific carbon and oxygen isotopes. These isotopes are the elements which we will be investigating to determine environmental and historical conditions of particular species.



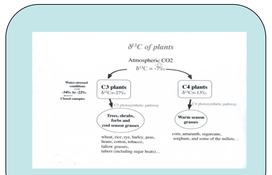
Enamel coated shown in white and stripes.



Formula used calculate the 12C/13C ration.

INTRODUCTION:

The ratios of the two stable forms of carbon (distinguished only by their atomic mass, ^{13}C and ^{12}C), provide a natural tracer tool for the chemical and biochemical reactions of the carbon cycle. In our research, the teeth were derived from the Mojave desert which produced both C3 and C4 plants from photosynthetic pathways. Some plant examples for C3 plants are trees, shrubs, forbs, and cool season grasses. The plant example for C4 plants are warm season grasses such as corn, amaranth, sugarcane, sorghum, and some of the millets. Rainfall environments, trees, shrubs and herbs follow a photosynthesis pathway. "The result is that the two groups of plants have very distinct $^{13}C/^{12}C$ ratios. Animals incorporate the plant carbon they eat into their tissues, which then directly reflect proportions of C_4 grasses and C_3 plants eaten." (Lee-Thorpe) In order to adequately retrieve the isotope ratios of our enamel samples, a gas spectrometer must be used to separate each isotope variation. The way this is done is by first positively ionizing the elements that are run through the mass spectrometer. Then the spectrometer increases the speed of the elements so that each variation is at the same speed. The mass spectrometer then deflects the moving elements by magnetism. Each element is separated into categories based upon their mass. Once the elements and their isotopes are in place, a beam of ions passes through each of them and is then recorded electronically. (Jim Clark)



Categorization of isotopic variation of C3 and C4 plants

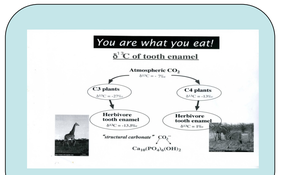
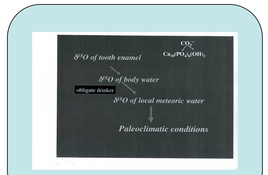


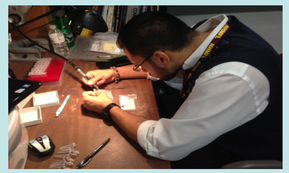
Diagram of diet variation for tooth enamel of various species



Categorized animals based on their food intake

EXPERIMENTAL PROCEDURES

1. Drill the serial sample teeth by taking a light-duty tissue wiper and folding it in half to collect the enamel powder, once cleared of debris found on the teeth.
2. Once 24 samples have been collected, **add 1 mL of 5% sodium hypochlorite solution to oxidize organic material** in each sample, mix them, and let it sit to react over night.
3. Rinse each sample with distilled water after pipetting the previous solution out by placing the samples in a centrifuge to separate the sample and the solution, and then adding the 1 mL of distilled water to each sample. Repeat this process 3 times.

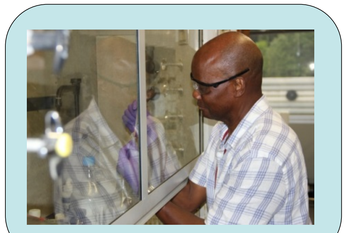


The process of cleaning and grinding the enamel off a teeth.

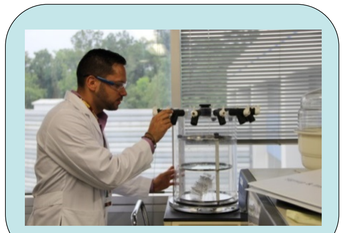
Centrifuge is used to rid the sample of impurities

4. Rinse each sample with acetic acid to extract any non-structural carbonates by adding 1 mL of the acid, shaking it, and centrifuging it to remove the reacted to acetic acid. Then an additional 1 mL of acidic acid is added to each sample for it to sit over night.
5. Repeat the process for **rinsing each sample with distilled wate**r to remove the solution in sample, as demonstrated in step 3.

6. Freeze-dry each sample by covering each sample with aluminum foil and poking a hole in it. Then place the samples in the freezer to sit overnight, so those frozen samples can be taking into the freeze dryer to remove any moisture.



Pipetting acetic acid to the enamel powder in a test tube.



Placing samples in the freeze dryer.

7. Bake each sample overnight to remove and additional moisture.
8. Flush each sample and standard with Helium to remove each sample of air and any non-structural carbonates that exist in the air.
9. **Add 8-10 drops of 100% phosphoric acid** to each sample and let it sit over night to be able to extract the structural carbonate when the mass spectrometer runs through the sample.
10. **Run each sample through the mass spectrometer** to collect the isotopic variation data of oxygen and hydrogen.
11. Weight each samples and carbonate standards *that correspond to the samples on a microbalance. This will provide a basis of comparison to the flushed samples in the future use of the mass spectrometer.*

Pipetting 100% phosphoric acid to the samples

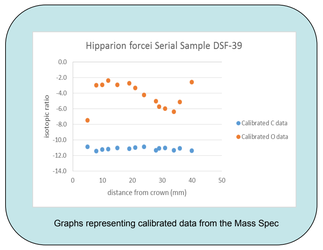
Observing and analyzing the results of the samples.

RESULTS:

DSF-39 DATA

Remarks	cali d13C	cal d18O	SS avg dC13	SS avg dO18	
5 mm from crown	-10.9	-7.5	-11.1	-4.2	5
8 mm	-11.4	-3.0			8
10 mm	-11.3	-2.9			10
12 mm	-11.2	-2.4			12
15 mm	-11.0	-3.0			15
19 mm	-11.1	-2.7			19
21 mm	-11.0	-3.4			21
24 mm	-10.9	-4.3			24
28 mm	-11.3	-5.1			28
29 mm	-11.1	-5.8			29
31 mm	-11.0	-6.0			31

Raw Data of Sample DSF-39



Graphs representing calibrated data from the Mass Spec

Based on the data, the *Hipparion forcei* Serial Sample DSF-39 was a species that consumed C-3 plants, as the isotopic variation result in the lower range then with C-4 plants. In addition, the oxygen isotope variation is correlated with the standard seasonal variation. The oxygen that was consumed by the water intake by the species.

CONCLUSION

The *Hipparion forcei* was a species that existed during the Dove Spring Formation in the Mojave Desert. A serial sample was taken from the enamel of the species to determine its isotopic variation of carbon and oxygen intake during development. The data confirmed that the species did consume C-3 plants and experienced a full seasonal cycle during it's development which enables us to be informed of the migration, location and identification of this species.

This is the region of the Mojave Desert where the fossils were collected

A further view of the fossils collection site

The Hipparion forcei.

ACKNOWLEDGMENTS

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