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## Introduction and Background

**Provenance**, a history of ownership, helps provide contextual evidence of an item's origin, or **provenience**. A transdisciplinary collaboration utilized a variety of techniques including archaeological and anthropological assessments, scanning electron microscopy (SEM), energy dispersive x-ray spectrometry (EDS), x-ray fluorescence (XRF), laser ablation inductively coupled plasma quadrupole mass spectrometry (LA-ICP-QMS), stable isotope analysis, radiocarbon dating and DNA analysis to yield information about the skull's background. Researchers hope to obtain enough information in order to repatriate the individual's remains.



Frontal view of the distorted skull



Left side view of skull

The skull is thought to be that of a Native American due to

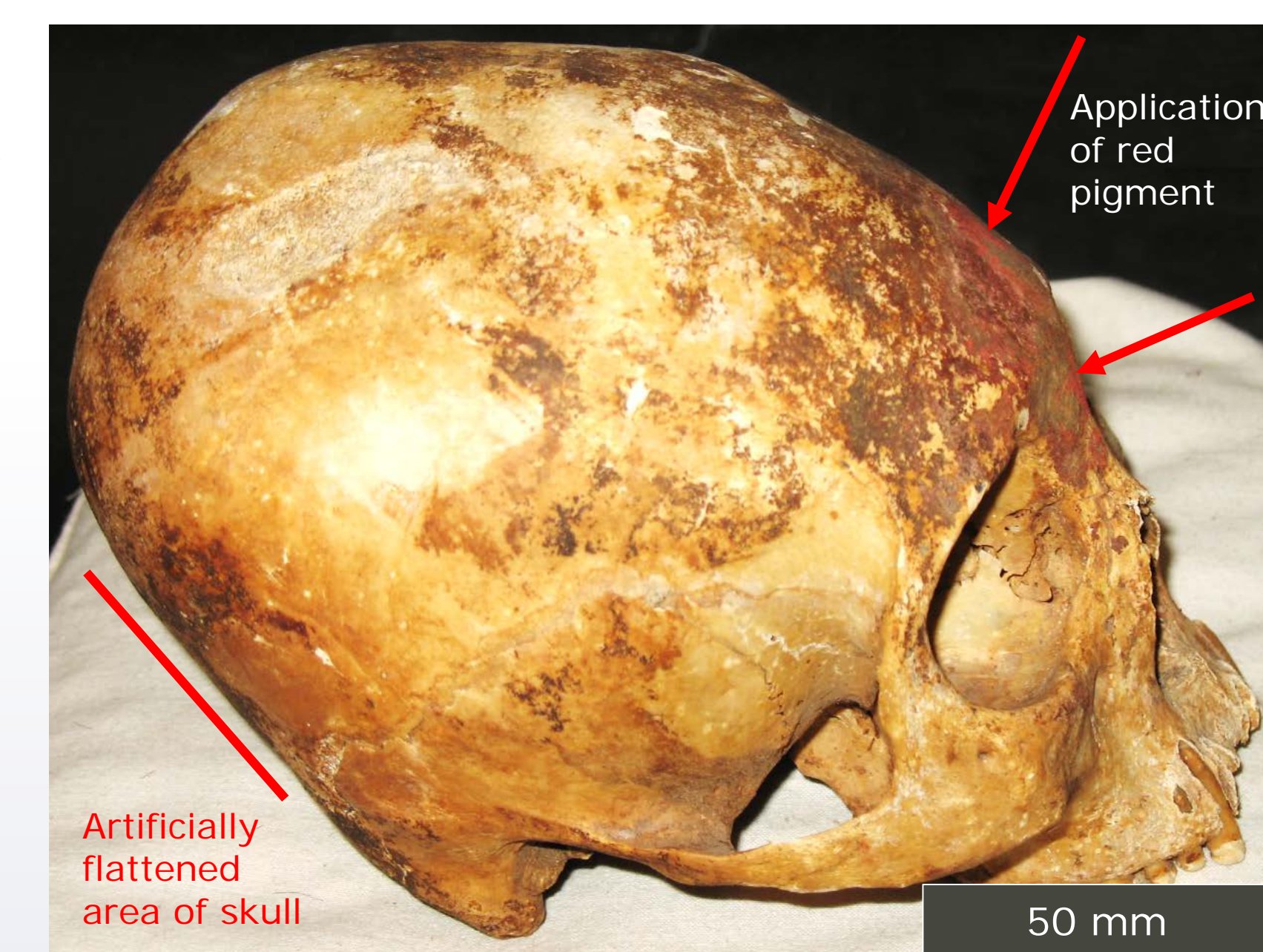
- Post-mortem application of red pigment embedded fibers
- Artificial cranial deformation (flattening at back of skull)

Analysis of pigment and fibers was completed using

- Anthropological assessment
- SEM/EDS
- LA-ICP-MS and XRF

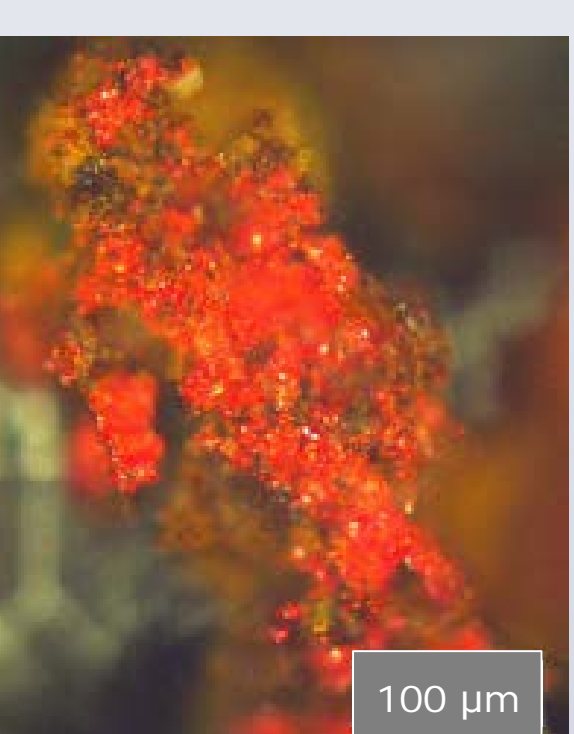
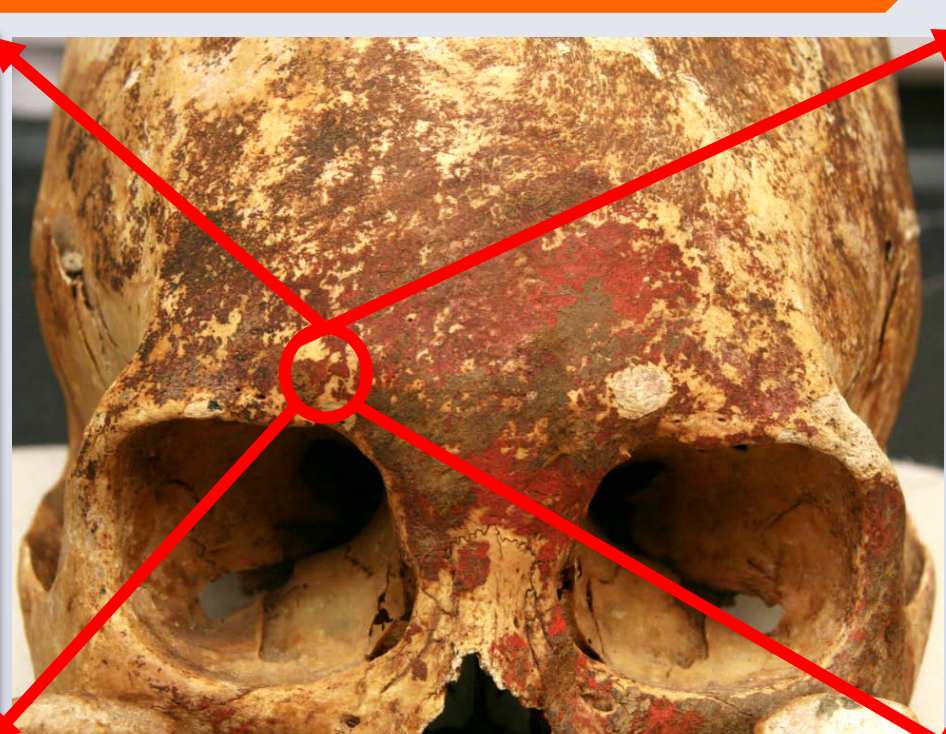
Age, habitat information, and lineage obtained by

- Radiocarbon dating and stable isotope analysis
- Forensic mitochondrial DNA for comparison to known Native American populations



Artificial cranial deformation evident at back of skull where flattening is seen

## Experimental Techniques



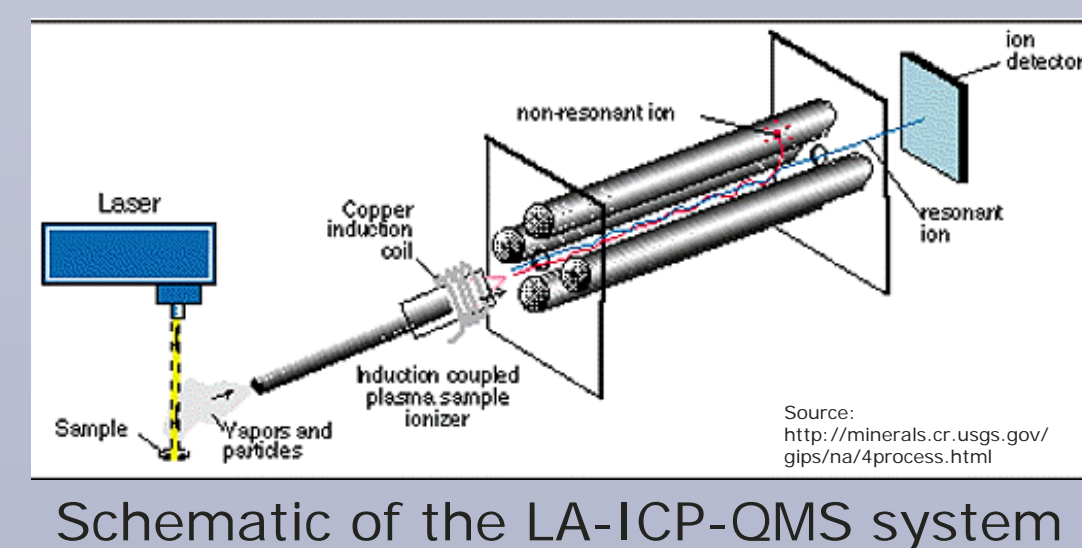
EDS electron image of elemental analysis locations. Inset: optical photo of same location

Close-up of skull where pigment samples were taken from

Optical photograph of red pigment

### Laser Ablation Inductively Coupled Plasma Quadrupole Mass Spectrometry (LA-ICP-QMS)

- Sample is vaporized and ionized in the plasma
- The quadrupole rod charge determines which ions will be carried to the detector for counting
- Samples were analyzed using a ThermoElectron X-Series II and New Wave Research UP-213 ND:YAG UV (213 nm) laser ablation system

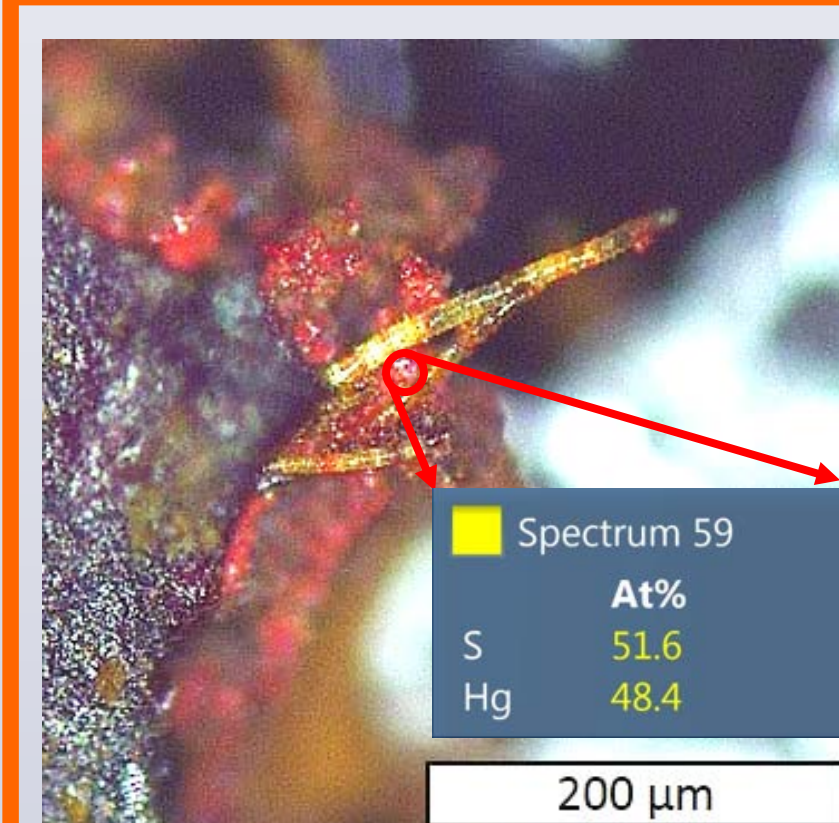


Schematic of the LA-ICP-QMS system

### Optical Microscopy, Scanning Electron Microscopy (SEM) and Energy Dispersive X-ray Spectroscopy (EDS)

- Similar to the beam of light used in an optical microscope, the SEM uses a beam of electrons to image the pigment
- EDS analysis was used to identify the elemental composition of the pigment

## Results and Discussion



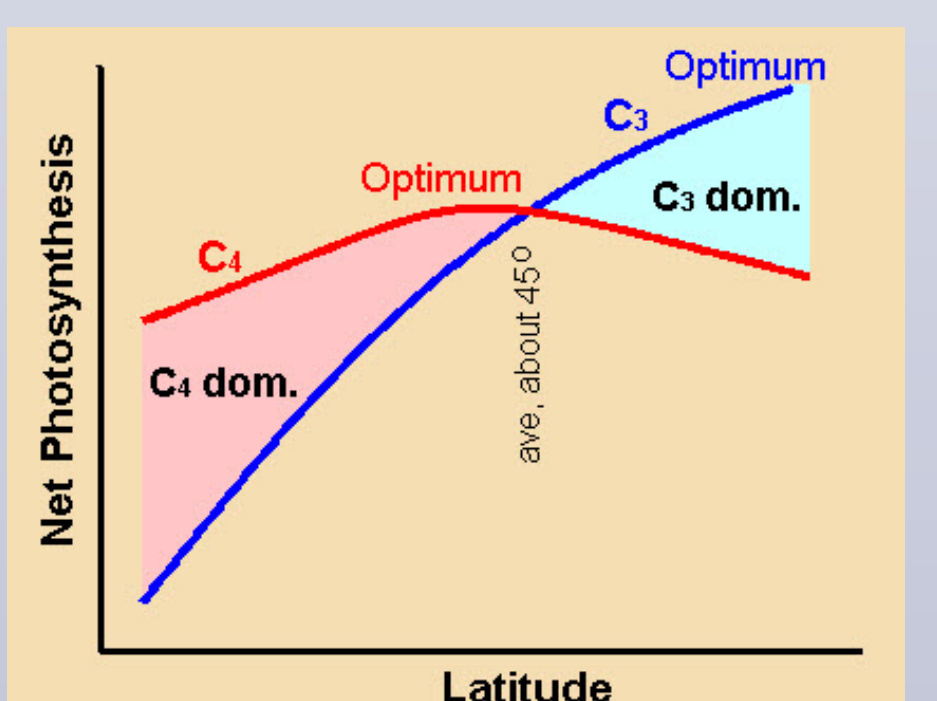
Optical image of red pigment with EDS inset showing elemental atomic %

Red pigment identified as mercury sulfide (HgS)

- Expected ratio of Hg to S would be 1:1
- Average ratio for all spectra was 1.12:1, possibly indicative of loss of sulfur due to microbial activity

Major Trace Elements (mass %)	Minor Trace Elements (ppm)
Silicon 12.7	Phosphorus 8300
Calcium 9.7	Titanium 6000
Iron 3.3	Zirconium 5400
Potassium 2.2	Bromine 1400
Dysprosium 1.1	Nickel 1300
Aluminum 0.9	Manganese 1200
	Europium 1200
	Neodymium 800

- XRF identified major and minor trace elements also present in the HgS
- Suggests the cinnabar is of a silica/calcium or silicate mineral origin and not a refined mineral as would be found in artworks



Net photosynthesis vs. Latitude for C<sub>3</sub> and C<sub>4</sub> plants. C<sub>3</sub> plants dominate at higher latitudes. [1]

### Stable Isotope Analysis

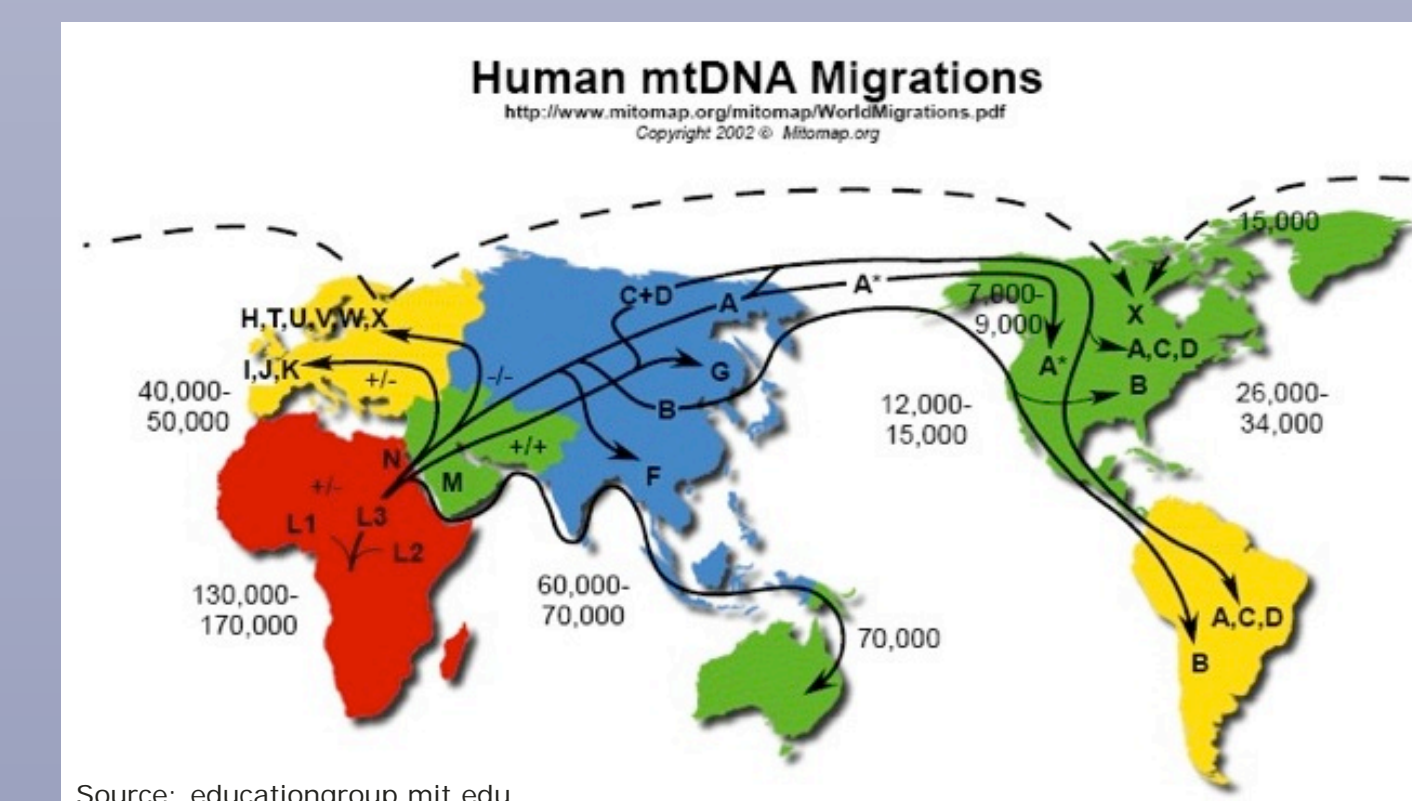
- Ratio of C<sub>3</sub> to C<sub>4</sub> in plants varies in different geographical locations
- This ratio, when measured in an animal, can identify the type of climate the animal lived in based on the kinds of plants it ate

### X-Ray Fluorescence (XRF)

- Uses emission of secondary x-rays from samples excited with high energy x-ray or gamma rays to identify elemental composition
- Samples analyzed using a Horiba XGT-7200 XRF Spectrometer

### Forensic mtDNA Analysis

ISU's Ancient DNA Extraction Lab and Molecular Research Core Facility specialize in extraction, amplification, sequencing and analysis of mitochondrial and genomic DNA from old, degraded, or fragmented samples.

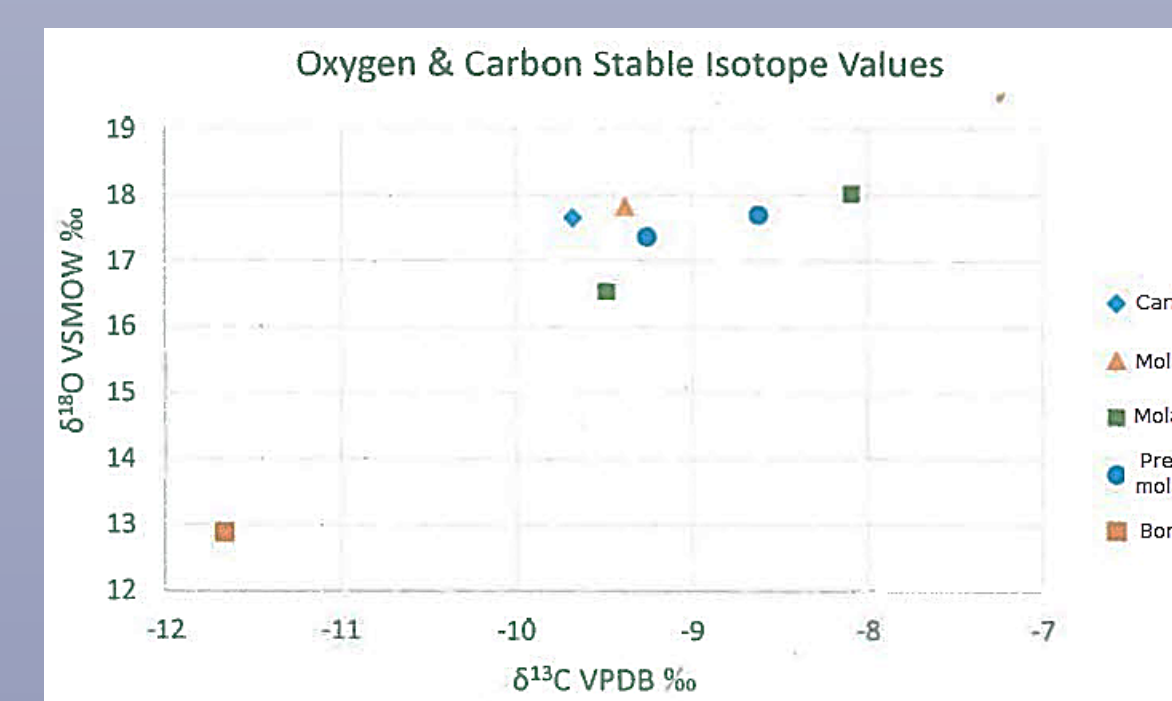


Source: educationgroup.mit.edu

LA-ICP-QMS Red Pigment Trace Elements (ppb)	
Phosphorus 189,000	Neodymium 180
Titanium 57,000	Lanthanum 90
Potassium 47,000	Dysprosium 75
Cerium 330	Europium 5

Stable Isotope analysis identified different values for teeth and bone

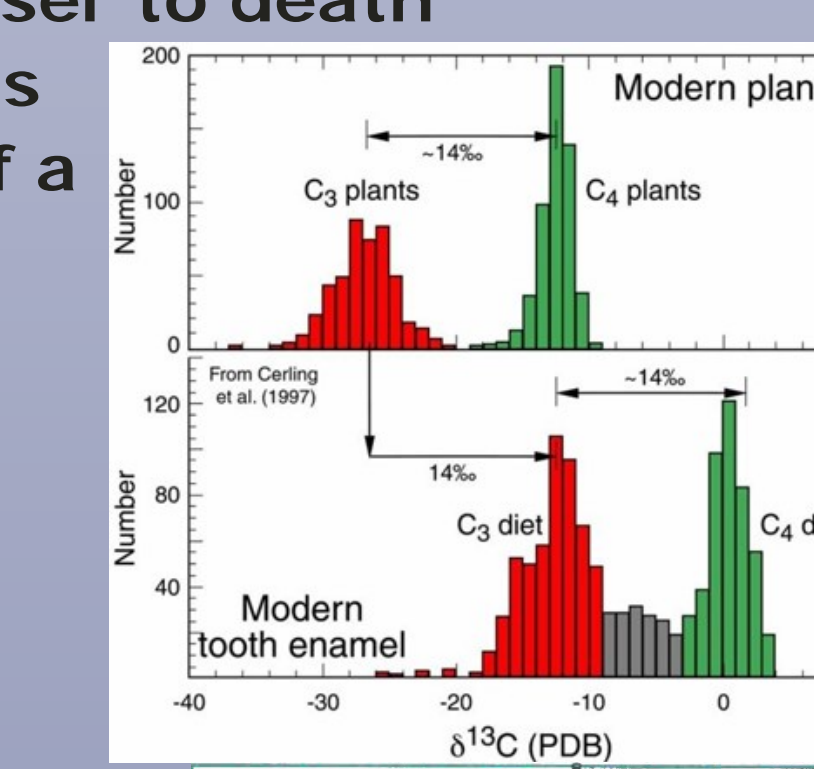
- Indicates individual lived in one area during youth and another area closer to death
- Bone value suggests C<sub>3</sub> type plants while tooth enamel is indicative of a mixed C<sub>3</sub>/C<sub>4</sub> environment



O and C stable isotope values from carbonate component of bone and teeth

LA-ICP-QMS is a more sensitive technique than XRF

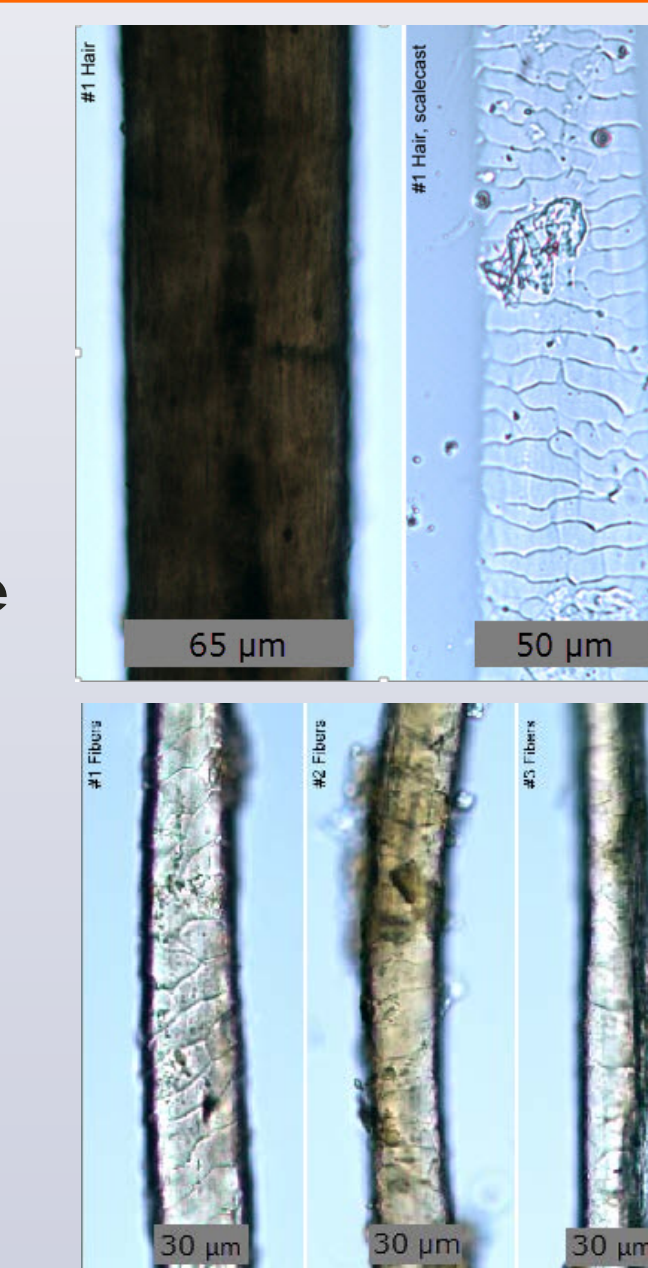
- Identified additional trace elements in red pigment
- Data can potentially be used to identify a geological cinnabar source



Relationship between carbon isotope values of C<sub>3</sub> and C<sub>4</sub> in modern plants and enamel

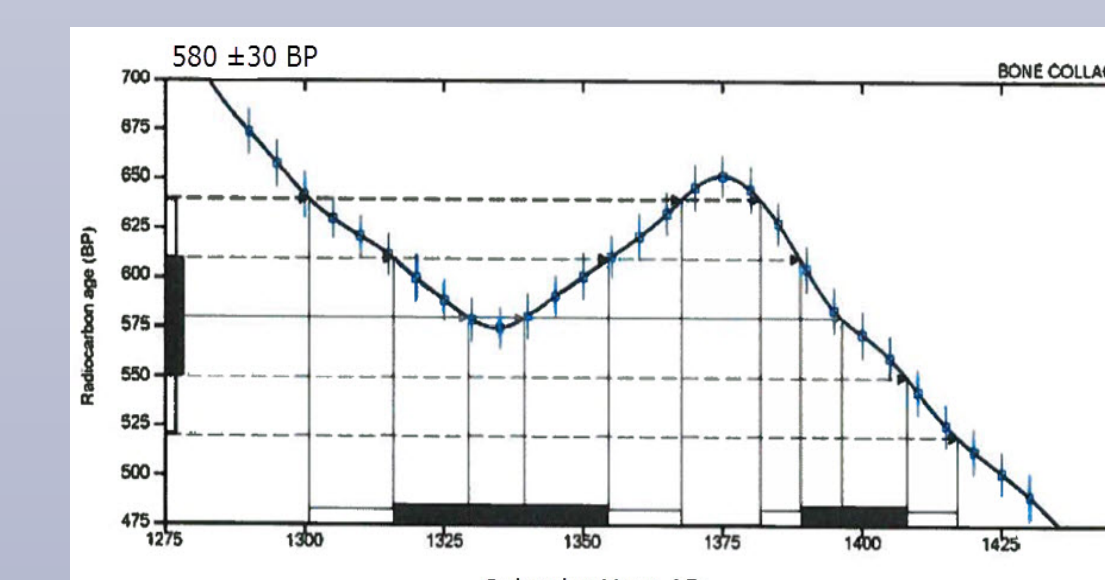
Hair and fiber samples sent for anthropological analysis to the University of Delaware Department of Art Conservation

- Hair samples (top) match the consistency of human hair
- Animal fibers (bottom) most likely sheep's wool or fine strands of cattle, deer, dog, goat, marmot or musk ox



### Radiocarbon Dating

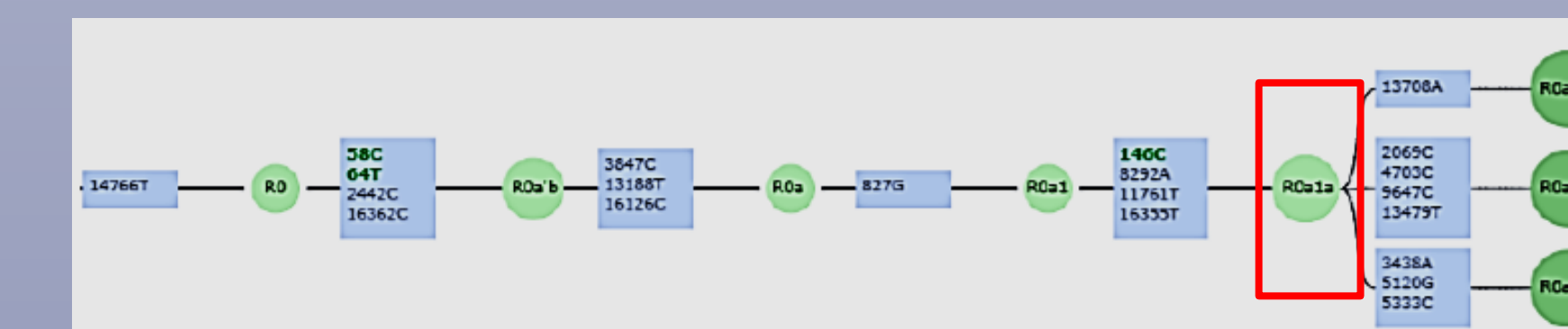
- Skull is ~550-620 years old
- Pre-Columbian (i.e. pre-European contact)
- Possibly of Chinook or Fremont culture, late pre-historic agricultural groups that extended into other states



Calibration of radiocarbon age to calendar years from Beta Analytic (Beta-393782) dating the skull to 1330-1400 AD

### Forensic Mitochondrial DNA

- Haplogroup R0a1a identified, although it is not associated with known Native American populations
- Area where Haplogroup R0a1a originated is from the Arabian Peninsula or western Eurasia



Haplogroup sequence identifying the skull's lineage from a maternal contribution as being from the Arabian Peninsula or western Eurasia

## Conclusions and Future Work

- Skull possibly of Chinook or Fremont culture
  - Cranial deformation and post-mortem application of HgS with adhered animal hair
- Trace elements in the HgS may potentially identify a geological source for the cinnabar
- Radiocarbon dated age ~ 550-620 years old
- Stable isotope analysis suggests habitation in different locations as a child and as an adult; locale characterized as cooler than would have been found in Idaho

- Forensic mitochondrial DNA identified haplogroup R0a1a which originated on the Arabian Peninsula; this is not consistent with the rest of the data in the study
- Future work will include
  - Additional trace element analysis from the bone to provide constraints to the duration of burial independent of C-14 dating
  - A secondary ancient DNA analysis to confirm or disprove the initial findings

## Acknowledgments

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  - Special thanks to Ken Reid, Idaho State Historical Preservation Officer, Idaho State Historic Preservation Office, Boise, ID.
  - The researchers are immensely grateful to Vicki Cassman and Elizabeth Shaeffer, University of Delaware, Department of Art Conservation, for their analysis on the hair and fiber samples.
- [1] "Patterns of Distribution." *Patterns of Distribution*. Web. 6 Apr. 2015. <<http://www.geo.arizona.edu/Antevs/ecol438/lect04.html>>.