Characterization of the elemental deposits in fossils from the Cradle of Humankind in South Africa and modern bones from the same geological area

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A dissertation submitted to the faculty of science, University of Witwatersrand, in fulfilment of the requirements for the degree of Master of Science.

Johannesburg 2010

Declaration

I declare that this dissertation is my own, unaided work. It is being submitted for the degree of Master of Science in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination in any other University.

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(Signature of Candidate)

______________________________ Day of ______________________________ 2010

ABSTRACT

Bones are complex, composite tissues consisting of inorganic calcium phosphate crystallites precipitated in an organized organic collagen matrix. However, diagenetic processes alter the original chemical composition and structure of the mineral and the organic components of bones during the burial period through leaching, decomposition and exposure to ground water. These activities serve to enrich, deplete and/or substitute the original elements in the bone through partial or complete dissolution, erosion, precipitation, recrystallization, ion uptake by sorption and diffusion, hydrolysis, crystal growth, and repolymerization processes. Thus exogenous elements from groundwater and soil may become incorporated into bone structure in a number of ways, and may reside in pores, voids or microcracks in the bone matrix. They can also form complexes with the organic component and adsorb onto the surface of hydroxylapatite matrix via ionic exchange.

Because of all these processes, the state of bone preservation varies greatly and depends on the physical and chemical characteristics of the burial environment, such as ground water and sediment composition, soil hydrology and pH, redox potential and temperature, soil solution fluoride and carbonate concentration, mechanical pressure, microbial activity, duration of interment and particle transport.

The study of the diagenesis process and the correlation of the morphological, organic and inorganic changes in varying geochemical environments, i.e. attaining adequate reliable data for modelling (predictive) purposes remains one of the challenges in the study of the diagenesis process.

Therefore, a study to characterize the morphological, mineralogical, and chemical features of fossils from Gladysvale Cave, South Africa, was done to investigate the mechanisms by which bone chemical compositions and mineralogical alterations occur during the burial period.

To achieve this, several analytical techniques were employed for the analysis of the burial soils and for the characterization of both modern and fossil bones. The following methods were employed: scanning electron microscopy (SEM) for morphological studies; X-Ray Fluorescence (XRF) mapping for the identification of elemental distribution within the bone apatite matrix; Powder X-Ray Diffraction (PXRD) for mineral determination; Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP AES) for elemental determination; bone porosity and density measurements; Carbon, Hydrogen, Nitrogen and Sulphur (CHNS) for detection of light elements, particularly nitrogen, whose concentration is correlated to the collagen content of the bone; and the redox and pH measurements of the soil in which the fossil bones were buried. These analytical techniques were used to determine qualitatively and quantitatively the sample's post-mortem elemental enrichment and/or leaching, even at trace level, and examine the elemental distribution across bone transverse sections.

Analysis of the soil where fossil bones were buried indicated relatively high amounts of Al, Fe, Mn, Ba, K, Si, V and Ti than in both the fossil and modern bones. Elemental enrichment was found to be more pronounced in fossil bones showing an increase in Fe, V, Ba, Cr, Zn, Al, Cu, Ti and Mn relative to modern bones due to the incorporation of authigenic minerals from the burial environment during the fossilisation process. This pattern also confirmed how porosity affects post-mortem elemental uptake and loss, thus porous tissue of fossil bones functions as a pathway of least resistance for postmortem elemental exchange with the depositional environment. Clay particle inclusions account for the high levels of Al in fossil bones relative to modern bones.

Numerous minerals were identified in all fossil specimens with carbonates, sulphides and phosphates being the most abundant. In contrast, few minerals were identified in modern specimens, with hydroxylapatite being present in all samples. Though all modern specimens were microscopically well-preserved, they were found to be poorly crystalline. The amorphous nature of these modern bones is correlated with high amounts of organic matter. In contrast, fossil bones are crystalline and this increase in crystallinity is correlated or attributed to loss of organic matter, especially the carbon contained in the carbonate hydroxylapatite mineral.

The morphological and physical (i.e. porosity, density and water absorption measurements) studies of these bones at both macro- and microscopic levels revealed that all the fossil specimens had suffered severe microbial attack due to their high porosity relative to modern bones. Using the nitrogen and carbon contents as proxies for bone preservation, the values are much lower in fossil bones relative to modern bones. Modern bones have retained much of the original fragments of original bio-molecules, for instance, collagen (as revealed by high %N content) as compared to fossil bones, which have lost more than 90% of their original bio-molecules. The well preserved specimens i.e. modern bones have a high collagen content, high bulk density and low porosity values. In contrast to this, degraded fossil bones displayed a low collagen content, low bulk density, large increases in porosity. This is an indication of poor preservation state of fossil bones relative to modern bones.

Redox and pH studies revealed that the fossil bones were buried on the surface under highly oxidising conditions. Such an oxygen-rich environment during dry periods is likely to have resulted in a rapid degradation of the bone's organic matter and also favoured the activity of micro-organisms, which is visible on scanning electron microscopy results.

Therefore fossil specimens show marked and complex alterations relative to their modern counterparts. These chemical and preservation disparities between the modern and fossil bones is attributed to significant diagenetic alteration, through the introduction of exogenous material to the existing matrix, and chemical alteration of the original bone matrix during the fossilisation processes.

Black-coloured fossil bones are also a common sight in most South Africa caves. Questions normally arise as to whether changes in bone colour were a result of burning or surface coating from minerals. In the event that the bones were burnt, then a challenging question would be regarding the cause of the fire, i.e. whether natural or human-controlled fire. In this study, a relatively simple, non-destructive chemical procedure is applied to distinguish between burned bones and mineral-coated bones by removing the oxides from soil/mineral contaminated fossil bones, followed by characterisation of elemental composition by CHNS and ICP-AES. Any coated versus burnt fossil bones disparities were ascribed to burning effects. The methods mentioned above were used for differentiating between black fossil bones that are burned and unstained; burned and stained; and stained but not burned. The results following the cleaning process have shown that the majority of the bones were indeed burned, of which a few were burned and stained. These observations were based on comparison of the cleaning results of burnt bones against coated bones. The self-ignition temperatures of organic matter found in the vicinity of burnt bones were experimentally obtained and were assumed to have caused a spontaneous ignition of the organic matter (bat guano), resulting in burning of the bones. Results of the CHNS analysis have showed insignificant amounts of free carbon in unburned bones. The quantity of char is directly correlated to the observed blackening. Blackened bones (those which were burnt between 300^{o}C and 400^{o}C) contained the most char. A relatively low percentage of free carbon was found in greyish-brown bones (heated at 500^{o}C to 600^{o}C) due to oxidation of the carbon in the organic molecule to carbon dioxide. Little char was observed in light-coloured specimens (those heated at 700^{o}C-800^{o}C). The bone is chalky-white and extremely light and brittle i.e. almost complete combustion has occurred. From both CHNS and ICP-AES analysis, the burnt fossil bone contained less amounts of elements compared to their coated counterparts simply due to major alterations in bone mineralogy which occurred when bones were heated above 700^{o}C.

Dedication

To my parents, Mr and Mrs T. Nhauro and the whole family

Acknowledgements

My heart-felt appreciation and sincere gratitude goes to my principal supervisor, Prof. Ewa M. Cukrowska, for her guidance, support, sound advice, boundless patience, enthusiasm, inspiration and great effort to explain things clearly throughout this work. To Dr. L. Backwell, my co-supervisor, I owe you a debt of gratitude for your valuable assistance, support, guidance and informative discussions from the beginning of this work, through field work up to the final write up.

Special thanks to the following organizations for their financial support during my studies; The National Research Foundation (NRF) Grantholder Bursary through Prof Ewa M. Cukrowska, University of the Witwatersrand Postgraduate Merit Award and Paleontological Scientific Trust (PAST) through Dr. L. Backwell.

I would like to extend my gratitude to the following individuals for having been of help during my research:

My parents, brothers and sisters for their unfailing love, support and encouragement.

Prof D. Billing, for his help with powder X-ray diffraction. Mr Mvuyisi Ngqola and Mr T. Mabaso for the outstanding courtesy and promptness in technical assistance.

My fellow colleagues and members of department in the Environmental Analytical Chemistry research Group (2008-2009), who contributed to my understanding of this vast field.

It is impossible to mention you all, and I apologise to those I have inadvertently left out.

Finally, I would like to thank The Almighty God for raising me up to more than I can be, giving me strength and wisdom to put this piece of work together. I just want to say "I love you Lord."

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ABBREVIATIONS

CHNS: carbon, hydrogen, nitrogen and sulphur

EDPA: Electron diffraction pattern analysis

EPMA: Electron probe micro analyzer

EPR: Electron paramagnetic resonance

ESCA: Electron Spectroscopy for chemical Analysis

FBS: Fossil bone sample

ICP AES: Inductively coupled plasma atomic emission spectrometry

MBS: modern bone sample

PIXE: Proton induced X-ray emission

PXRD: Powder X-ray diffraction

PY-GC-MS: Pyrolysis gas chromatography–mass spectrometry

RSD: Relative standard deviation

SEM: Scanning electron microscope

TOF-SIMS: Time-of-flight secondary ion mass spectrometry

TXRF: Total reflection X-ray fluorescence

XPS: X-ray photoelectric spectroscopy

XRF: X-ray fluorescence

Syn: Synthetic