NOTICE

The quality of this microform is heavily dependent upon the quality of the original thesis submitted for microfilming. Every effort has been made to ensure the highest quality of reproduction possible.

If pages are missing, contact the university which granted the degree.

Some pages may have indistinct print especially if the original pages were typed with a poor typewriter ribbon or if the university sent us an inferior photocopy.

Reproduction in full or in part of this microform is governed by the Canadian Copyright Act, R.S.C. 1970, c. C-30, and subsequent amendments.

AVIS

La qualité de cette microforme dépend grandement de la qualité de la thèse soumise au microfilmage. Nous avons tout fait pour assurer une qualité supérieure de reproduction.

S'il manque des pages, veuillez communiquer avec l'université qui a conféré le grade.

La qualité d'impression de certaines pages peut laisser à désirer, surtout si les pages originales ont été dactylographiées à l'aide d'un ruban usé ou si l'université nous a fait parvenir une photocopie de qualité inférieure.

La reproduction, même partielle, de cette microforme est soumise à la Loi canadienne sur le droit d'auteur, SRC 1970, c. C-30, et ses amendements subséquents.

UNIVERSITY OF ALBERTA

ELEMENTAL ANALYSIS OF SOIL AND HUMAN AND ANIMAL BONE FROM A MATURE PHASE CEMETERY AT HARAPPA, PAKISTAN: IMPLICATIONS FOR PALAEODIET RECONSTRUCTION

BY

© DAVID WALTER LINK

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS

FOR THE DEGREE OF MASTER OF ARTS

DEPARTMENT OF ANTHROPOLOGY

EDMONTON, ALBERTA

FALL, 1990

ISBN 0-315-64812-0

David Link
Box 16, Site 300, RR3
Stony Plain, AB
T0E 2G0

October 28, 1989

W.B. Saunders Co.
1 Goldthorne Ave.
Toronto, Ont.
M8Z 5T9

Dear Madam/Sir:

I would like to request permission to use Fig. 4-1 on page 55 of *Atlas of Histology* (1985) by Leeson, Leeson and Paparo (ISBN 0-7216-1202-4) in my Master of Arts thesis at the University of Alberta. Thank you for your consideration of this.

Yours sincerely,

D. Link

Permission granted by the copyright owner, contingent upon the consent of the author and provided complete credit is given to original source.

Julie Lawley 12/3/89
W. B. Saunders Company Date
Philadelphia, PA

Fig. 2
Pg. 11

David Link
Dept. of Anthropology
University of Alberta
Edmonton, Alberta
Canada T6G 2H4

29.12.1989

Dr. Thomas S. Leeson
Department of Anatomy
University of Alberta

Dear Dr. Leeson:

I would like to request permission to use Fig. 4-1 on page 55 of *Atlas of Histology* (1985) by Leeson, Leeson and Paparo in my Master of Arts thesis at the University of Alberta. Thank you for your consideration of this.

Yours sincerely,

D. Link

Permission granted.

T. Leeson

Jan 8. 1990.

University of Illinois
at Urbana-Champaign

Department of Cell and Structural Biology

506 Morrill Hall
505 South Goodwin Avenue
Urbana, IL 61801

School of Life Sciences

217 333-6118

January 10, 1990

David Link
Box 16, Site 300 R. R. 3
Stony Plain,
Alberta TOE 2GO
CANADA

Dear Mr. Link:

Thank you for your note of 29-12-89.

Permission is granted for you to use Figure 4-1 of the Atlas of Histology (1985).

I trust you will be able to complete your Master of Arts Thesis successfully.

Sincerely,

C. R. Leeson, M.D., Ph.D.
Professor

CRL:djl

CAROLINA BIOLOGICAL SUPPLY COMPANY

P.O. BOX 1059
BURLINGTON, NORTH CAROLINA 27215

1308 Rainey St.
Burlington, N.C. 27215

Phone 919-226-6000
TLX 574-354

16 November 1989

David Link
Box 16, Site 300, RR3
Stony Plain, Alberta
Canada T0E 2G0

Dear Mr. Link:

Thank you for your recent request for the use of Carolina Biological Supply Company's photographs. We are happy to help supply your photo needs.

The photos from 48-2118 Skeletal Tissue Set that you are interested in were produced by us, but do not belong to us. I have listed the co-authors of that series (who own the images) so that you might contact them to obtain permission:

Dr. Randy Kardon
Department of Opthamology
University of Iowa Hospitals
Iowa City, Iowa 52242

Dr. Richard Kessel
Department of Zoology
University of Iowa
Iowa City, Iowa 52242

If you have any questions or if I may be of further assistance, please feel free to call me.

Yours sincerely,

Cindy Vines Bright

Cindy Vines Bright
Manager, Photo Archives

Fig. 3, 4, 5
Figs. 12, 14, 16

The University of Iowa
Iowa City, Iowa 52242

The University of Iowa Hospitals and Clinics
Department of Ophthalmology 319/356-2215
FAX 319/356-0363

Appointments only 319/356-2852
Nights and weekends 319/356-1616

DEPARTMENT HEAD:
Thomas A. Weingeist, M.D., Ph.D.
356-2867

ADMINISTRATION:
Michael W. Bresnahan
356-2866

CATARACT:
Hansjoerg E. Kolder, M.D., Ph.D.
356-4321

COMPREHENSIVE OPHTHALMOLOGY:
Frederick C. Blodi, M.D.
356-2427
Thomas A. Farrell, M.D.
356-0429
Robert Folberg, M.D.
335-7095

CONTACT LENS/LOW VISION:
Colleen A. Howe, O.D.
356-4774

CORNEA/EXTERNAL DISEASE:
Jay H. Krachmer, M.D.
356-2861

ECHOGRAPHY:
Karl C. Ossoinig, M.D.
356-2869

GLAUCOMA:
Service: 356-2878
W.L.M. Alward, M.D.
356-2228

HERITABLE EYE DISEASES:
G. Frank Judisch, M.D.
356-1605

NEURO-OPHTHALMOLOGY:
Service: 356-2548
James J. Corbett, M.D.
356-2571
Randy H. Kardon, M.D., Ph.D.
356-2260
H. Stanley Thompson, M.D.
356-2868

OCULAR PATHOLOGY:
Frederick C. Blodi, M.D.
356-2427
Robert Folberg, M.D.
335-7095

OCULOPLASTICS & ORBIT:
Service: 356-4005
Keith D. Carter, M.D.
356-7997
Jeffrey A. Nerad, M.D.
356-2590

OPHTHALMIC PHOTOGRAPHY:
Service: 356-2339
Paul R. Montague, C.R.A.
356-2880

OPTOMETRY:
Colleen A. Howe, O.D.
356-4774

PEDIATRIC OPHTHALMOLOGY/ STRABISMUS:
Service: 356-2859
G. Frank Judisch, M.D.
356-1605
Ronald V. Keech, M.D.
356-3991
William E. Scott, M.D.
356-2877

VASCULAR DISEASE:
Sohan S. Hayreh, M.D., Ph.D., D.Sc.
356-2921

VITREOUS/RETINA:
Service: 356-2951
Christopher F. Blodi, M.D.
356-3993
James C. Folk, M.D.
356-4338
Alan E. Kimura, M.D.
356-8350
Jose S. Pulido, M.D.
356-1497
Thomas A. Weingeist, M.D., Ph.D.
356-2867

1847

January 3, 1990

David Link
Box 16, Site 300, RR3
Stony Plain, Alberta
CANADA T0E 2G0

Dear Mr. Link:

I would be happy to grant you permission to use images 2, 4 and 6 from part 2 of Carolina Biological Supply's no. 48-2118 Skeletal Tissue Set. I wish you good luck with your Master's thesis and if I could be any further help to you, please do not hesitate to contact me.

Sincerely,

Randy Kardon

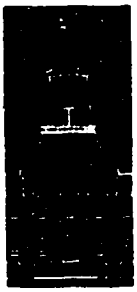
Randy Kardon, M.D., Ph.D.

RK/jgp

The University of Iowa
Iowa City, Iowa 52242 USA

Department of Biology

319/335-1050



1847

April 4, 1990

David Link
Department of Antropology
University of Alberta
Edmonton, Alberta
Canada T6G 2H4

Dear Mr. Link,

You have my permission to reproduce images (figures) 2, 4 and 6 from part 2 of Carolina Biological Supply's No 48-2118 Skeletal Muscle Tissue Set in your MA Thesis at the University of Alberta. You should (a) obtain permission from Carolina Biological Supply and (b) provide complete citation as to source adjacent to the illustration where it is used.

Sincerely,

Richard G. Kessel
Professor

br

GEORG-AUGUST-UNIVERSITÄT GÖTTINGEN
ZENTRUM ANATOMIE
DES FACHBEREICHS MEDIZIN
Abteilung Morphologie

Priv.-Doz. Dr.Dr. Michael Schultz

Zentrum Anatomie · Kreuzbergring 36 · D-3400 Göttingen

Göttingen , 7 - 17 -90

Telefon: 0551/39-
Telex: 96703 unigoe-d

David Link
Dept. of Anthropology
University of Alberta
Edmonton, Alberta
CANADA T6G 2H4

Dear colleague,

thank you very much for your letter. Please, excuse that I answer so late, but I stayed a long time abroad for scientific research. Unfortunately thepapers you asked for are still in press. Thus, I send you another one.

Of course, you may use photographs of my Papaer. Unfortunately I have no photos yet, which I can send you.
As I shall go tomorrow for 2 1/2 months on scientific research in Turkey, I shall be back in Göttingen in autumn. Then, I hope, I shall have a little more time. If you are interested, we should keep contact.

For today my best regard,
yours sincerely

Priv.-Doz. Dr.Dr. M. Schultz

Fig 6, 7, 8
Pgs. 38, 40, 41

UNIVERSITY OF ALBERTA

RELEASE FORM

NAME OF AUTHOR: DAVID WALTER LINK

TITLE OF THESIS: ELEMENTAL ANALYSIS OF SOIL AND HUMAN AND ANIMAL BONE FROM A MATURE PHASE CEMETERY AT HARAPPA, PAKISTAN: IMPLICATIONS FOR PALAEODIET RECONSTRUCTION

DEGREE: MASTER OF ARTS

YEAR THIS DEGREE GRANTED: 1990

DAVID WALTER LINK

14745 51 AVE.
EDMONTON, AB
CANADA
T6H 5E6

DATE: SEPTEMBER 10, 1990

UNIVERSITY OF ALBERTA

FACULTY OF GRADUATE STUDIES AND RESEARCH

THE UNDERSIGNED CERTIFY THAT THEY HAVE READ, AND RECOMMEND TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH FOR ACCEPTANCE, A THESIS ENTITLED

ELEMENTAL ANALYSIS OF SOIL AND HUMAN AND ANIMAL BONE FROM A MATURE PHASE CEMETERY AT HARAPPA, PAKISTAN: IMPLICATIONS FOR PALAEODIET RECONSTRUCTION

SUBMITTED BY DAVID WALTER LINK

IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF ARTS.

NANCY C. LOVELL

OWEN B. BEATTIE

MICHAEL L. WAYMAN

DATE: SEPTEMBER 6, 1990

ABSTRACT

The purpose of this study was to evaluate the suitability for diet reconstruction of the mineral part of human bone samples excavated in a cemetery of the Mature Phase of the Indus civilization. This was done, first, through analysis of soil samples taken from the burial environment with the goal of determining the availability to the bone of ions that could mask or alter any dietary signature. Second, samples of human bone taken in sequence from cross-sections of femora were examined for any gradients of change in elemental content from the periosteal to the endosteal surfaces of the sections. Relationships between the results obtained from analysis of the two materials, in the form of co-occurrence of constituents, were expected to be of assistance when assessing whether elements in the bone could be considered to convey biogenic signals. Third, associated bone samples of an animal of known diet (*Bos*) and of a diet assumed to be similar to that of humans (*Canis*) were analyzed as possible auxiliary indices of postmortem change. Elemental content of samples was obtained in all cases by instrumental neutron activation analysis. Elements of interest were some known indicators of diet (Ba, Na, Sr, Zn) and some known indicators of contamination of bone by soil (Al, Mn).

Evidenced obtained primarily from the trace element signatures of the soil and human bone samples led to rejection of the latter as a source of information for palaeodiet reconstruction. The mechanisms likely involved in the postmortem alteration of the physiological trace element levels of these bone samples are discussed. Discrepancies in results from mature and immature bone from the same burial context are addressed. Several points are raised that warrant attention in future analyses of trace element content of bone mineral.

ACKNOWLEDGEMENTS

I wish to thank the members of my committee for their involvement in this project. In particular, thanks are due to my supervisor, Dr. Nancy C. Lovell, who allowed access to the materials used in the analyses, and to Professor Michael Wayman, who allowed access to laboratories in the Department of Mining, Metallurgical and Petroleum Engineering. I am also indebted to several individuals for their time and technical expertise: Gordon Haverland, SLOWPOKE reactor research associate; Christina Barker, Robert Konzuk, and Shiraz Merali, technicians, Department of Mining, Metallurgical and Petroleum Engineering; and Professors James Robertson and Marvin Dudas, Department of Soil Science.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

1. INTRODUCTION

Analysis of the trace element content of bone has been a popular tool in palaeodietary studies over the past ten to 15 years. Operating from the premise that the mineral content of bone can reflect quantities of certain essential, nonessential, and toxic substances ingested or absorbed during life, researchers have attempted to identify broad categories of food sources by the presence, absence, or relative amounts of certain elements in the skeletal remains of humans and animals (Price *et al.* 1985).

Research of this nature must involve the acceptance, whether stated or not, of certain assumptions: the element of interest is a consistent factor in a food source, the intrapopulation geochemical environment is unvarying, the presence of the element in bone is correlated with its presence in the diet, the element is not obscured by mortuary practices (*e.g.*, embalming), and the element has not been affected by postmortem change in bone resulting from interment. Moreover, possible elemental variation due both to physical location of the sample (such as mortuary site, burial, skeleton, bone, and bone portion) and to demographic and physiological factors (such as age, sex, reproductive status, and

social standing) may or may not be viewed as a problem in sampling (Buikstra *et al.* 1989; Price 1989). The inability to control for one or more of these factors could lead to spurious results.

An awareness of postdepositional alteration in the chemical content of bone has, more than any other of the noted factors, led to concern about the accuracy and precision of techniques of palaeodiet reconstruction involving trace elements. Such postmortem change in elemental content may be the result of movement of materials either from bone to its burial environment, normally soil, or vice versa (Grupe 1987). It may also appear as diagenesis, a process of chemical reconstitution that can occur in conjunction with or independent of contamination (Lambert *et al.* 1989).[1]

Attempts have been made to identify and subsequently control for net loss or gain of elements of interest through various means. These include comparison of the elemental content of cortical and cancellous bone, with the latter assumed to be more prone to alteration (Grupe 1988a; Lambert *et al.* 1982); investigation of

---

1. The term "diagenesis" has been misconstrued to mean contamination of bone by soil (*e.g.*, Aufderheide 1989), any postmortem change in chemical composition (*e.g.*, Katzenberg 1984; Klepinger *et al.* 1986), or even gross morphological alteration caused by microfloral invasion (Bell 1990). Its definition here is limited to the postmortem reconstitution of chemical compounds in bone.

calcium/phosphorus ratios, which normally increase with diagenesis (Sillen 1989); measurement of bone mineral crystallinity, which tends to increase with the length of time the bone is interred and is indicative of change in the stoichiometric relationships in the mineral (Hassan *et al.* 1977; Sillen 1989); identification of shifts in isotopic ratios of strontium ($^{87}Sr/^{86}Sr$) as indicators of diagenetic recrystallization (Nelson *et al.* 1983); comparison of human bones with those of animals with known diet from the same archaeological site (Francalacci 1989; Sillen 1981); electron microprobe analysis of bone cross-sections with the intent of searching for gradients of change (Lambert *et al.* 1983; Sagne 1978); and analysis of burial context, usually soil (El-Kammar *et al.* 1989; Pate and Hutton 1988). From a survey of the literature, the final three methods currently appear to be the most successful.

Using these three techniques, this study evaluates the suitability of the inorganic part of interred bone for dietary reconstruction. This is done, first, through instrumental analysis of soil samples taken from the burial environment with the goal of determining the availability to the bone of ions that could mask or alter any dietary signature. Second, cross-sections of femoral bone, some with known association to particular soil

samples, are examined for gradients of change in elemental content across the section. Relationships between the results obtained from analysis of the two materials in the form of co-occurrence of constituents are expected to be of assistance when assessing whether any elements present in the bone can be considered to convey biogenic signals. Third, associated bone samples of animals of known diet (*Bos*) and of a diet assumed to be similar to that of humans (*Canis*) are instrumentally analyzed. This step may assist in clarifying whether or not postmortem alteration, which must assumed to be similar for the human and faunal samples, has occurred to a degree that renders any signs of dietary origin unreliable.

The samples used in this study are from mortuary site excavations undertaken in 1987 and 1988 at Harappa, Pakistan, by researchers from the University of California at Berkeley and the University of Wisconsin at Madison (Dales and Kenoyer 1988). One of the two most important sites for the study of the Indus civilization, the settlement centre of Harappa includes five known cemeteries, of which the excavations of three have not yet been published (Allchin and Allchin 1982). The material used for analysis here was collected from the mature phase cemetery R37, first investigated in the

period 1937-1941.

The sites of Harappa and Mohenjo-daro (Fig. 1) appear best to represent the culmination of a cultural development in the Indus river valley system that is identifiable as early as the Neolithic (Allchin and Allchin 1982; Jansen 1989). The spread of settlements and the convergence of material culture traits suggest a trend toward cultural unification by the so-called Early Indus Phase, believed to span the time period from the second half of the fourth to the middle of the third millenium B.C. By this stage, exploitation of the Indus alluvium, with much cereal production and animal husbandry, resulted in incipient urbanism and a general build-up of population throughout the Indus plains. The apparent establishment of a socio-economic interaction sphere appears to have been the consequence of this development and the associated growth of technology and agricultural expertise.

The mature phase of the Indus civilization, now believed to date from around 2300 to 1750 B.C., is characterized by cultural uniformity throughout the region, for example in the form of town planning, brick sizes, and tool types (Agrawal 1982). Lacking the time depth of the major Neolithic, Chalcolithic, Early Indus and Mature Indus site of Mehrgarh, Harappa and Mohenjo-

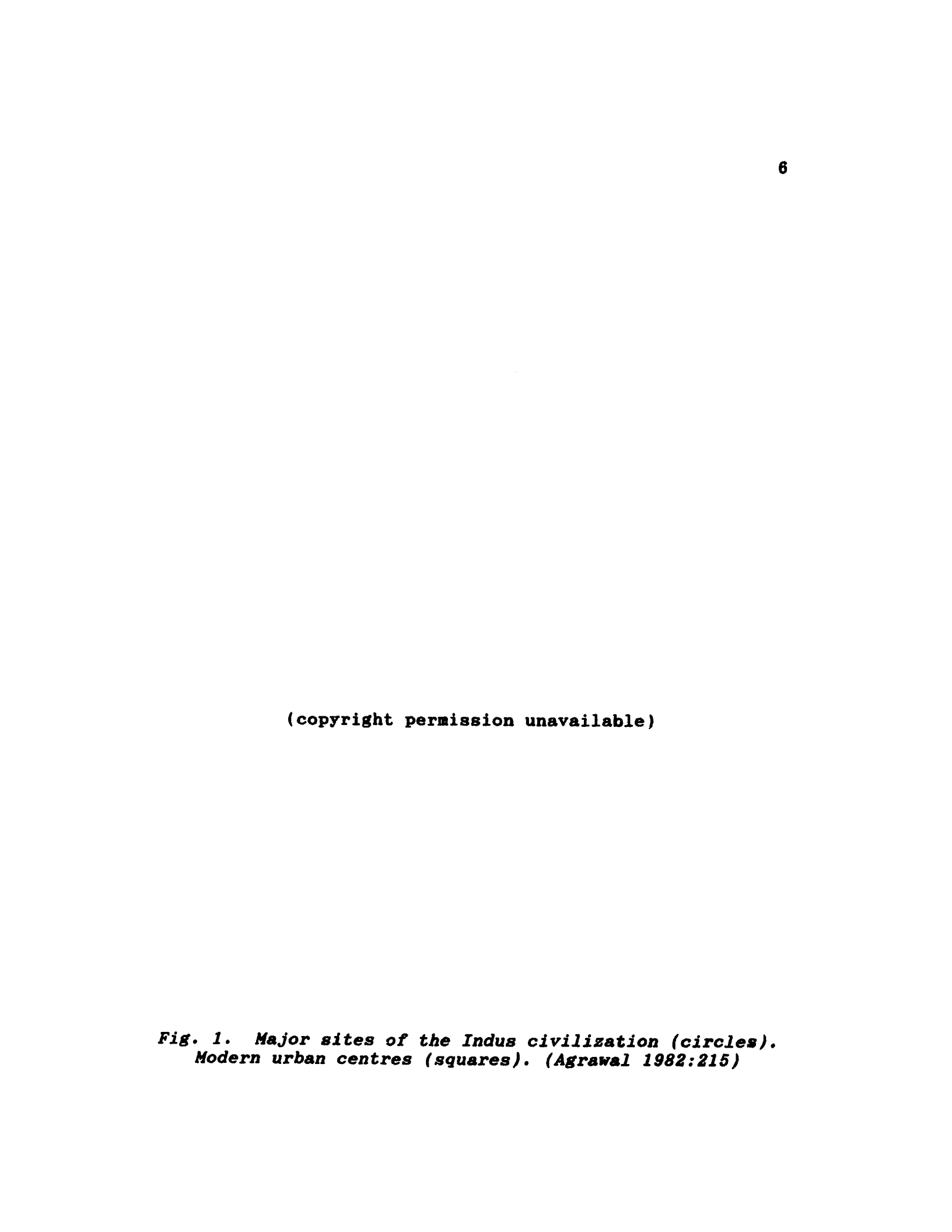


Fig. 1. Major sites of the Indus civilization (circles). Modern urban centres (squares). (Agrawal 1982:215)

daro do not appear to have existed before the beginning to the middle of the Early period. This may be attributable to the increase in settlement number during this time (Allchin and Allchin 1982).

The catalyst for the rather rapid demise of the Indus valley civilization after approximately 1750 B.C. has been a point of contention (Dales 1989). Earlier scholars favoured an invasion theory involving speakers of an Indo-European language, but more recent work has shown that a general system collapse probably occurred (Renfrew 1989). This may have resulted from the rapidly increasing aridity in the region, as evidenced by palynological studies (Agrawal 1982). Cemetery R37 does not appear to have been in use after the decline of the Mature Indus phase. It is characterized by Mature Indus pottery and radiocarbon dates consistent with that time period (N. Lovell, pers. comm.). Consequently, it may be assumed that the bone samples analyzed here date from the same known time period.

Previous attempts to document diet or change therein in the Indus civilization by means of biochemical bone assay have been thwarted by problems of postdepositional alteration (Radosevich 1989). At Harappa specifically, it was hoped that such assay would provide information regarding the possible presence of different socio-

economic divisions in the population (Dales 1989). An examination of the interaction between soil and bone would shed light on the nature of the alteration mechanisms involved, both at the level of this particular site and, more broadly, in archaeological context in general.

In order to place the subsequent discussions and analyses in a clear context, the following chapter provides a description of bone histology and bone mineral chemistry. Chapter 3 presents a consideration of complications in trace element analysis related to antemortem (intrinsic) and postmortem (extrinsic) conditions in bone mineral composition as a background to the discussion of methodology and results in chapters 4 to 6.

2. THE STRUCTURE AND COMPOSITION OF FORMED BONE

A. Introduction

An examination of problems associated with the elemental analysis of bone requires consideration of both micromorphological and biochemical aspects of this complex material. This chapter presents a review of basic bone histology and bone mineral chemistry with the aim of providing sufficient information for the discussion of *in vivo* and postmortem processes of elemental behaviour in chapter 3.

B. Bone Histology

Bone is a rigid connective tissue that consists of three parts: an organic component, an inorganic component, and water (Ortner and Putschar 1985). These components interact in a complex manner, with the organic element (cells and matrix) forming a framework on which the inorganic mineral part of bone is established. Bone matrix consists mainly of fibres of collagen that, in fully formed bone, are impregnated with chemical compounds, chiefly phases of calcium phosphate (Neuman 1980). It is these compounds that impart firmness to the structure.

Depending on the amount of solid matter and the arrangement of spaces in this matter, bone is classified as either spongy (cancellous) or compact (dense) (Leeson *et al.* 1985). Histologically, both types contain the same cells and matrix, but while the avascular trabeculae of spongy bone form a lattice that accomodates bone marrow within its structure, the matrix in compact bone is characteristically arranged in layers (lamellae). The appearance of these layers is determined both by the original, unremodelled structure of the bone (periosteal and endosteal lamellae) and by the distribution of blood vessels that subsequently occur in channels called Haversian canals (concentric lamellae) (Fig. 2). While the Haversian canals run longitudinally, connecting transverse (Volkmann's) canals entering through the periosteum and endosteum form additional supply routes for nutrients. Situated between the lamellae are lacunae that, in live bone, contain osteocytes, cells probably responsible for final mineralization of adjacent matrix and mineral exchange with the rest of the body (Ortner and Putschar 1985). Very fine channels (canaliculi) radiate outward from the osteocytes and connect with canaliculi from adjacent lacunae and directly or indirectly with the corresponding Haversian canal (Fig. 3). The entire arrangement of the Haversian canal,

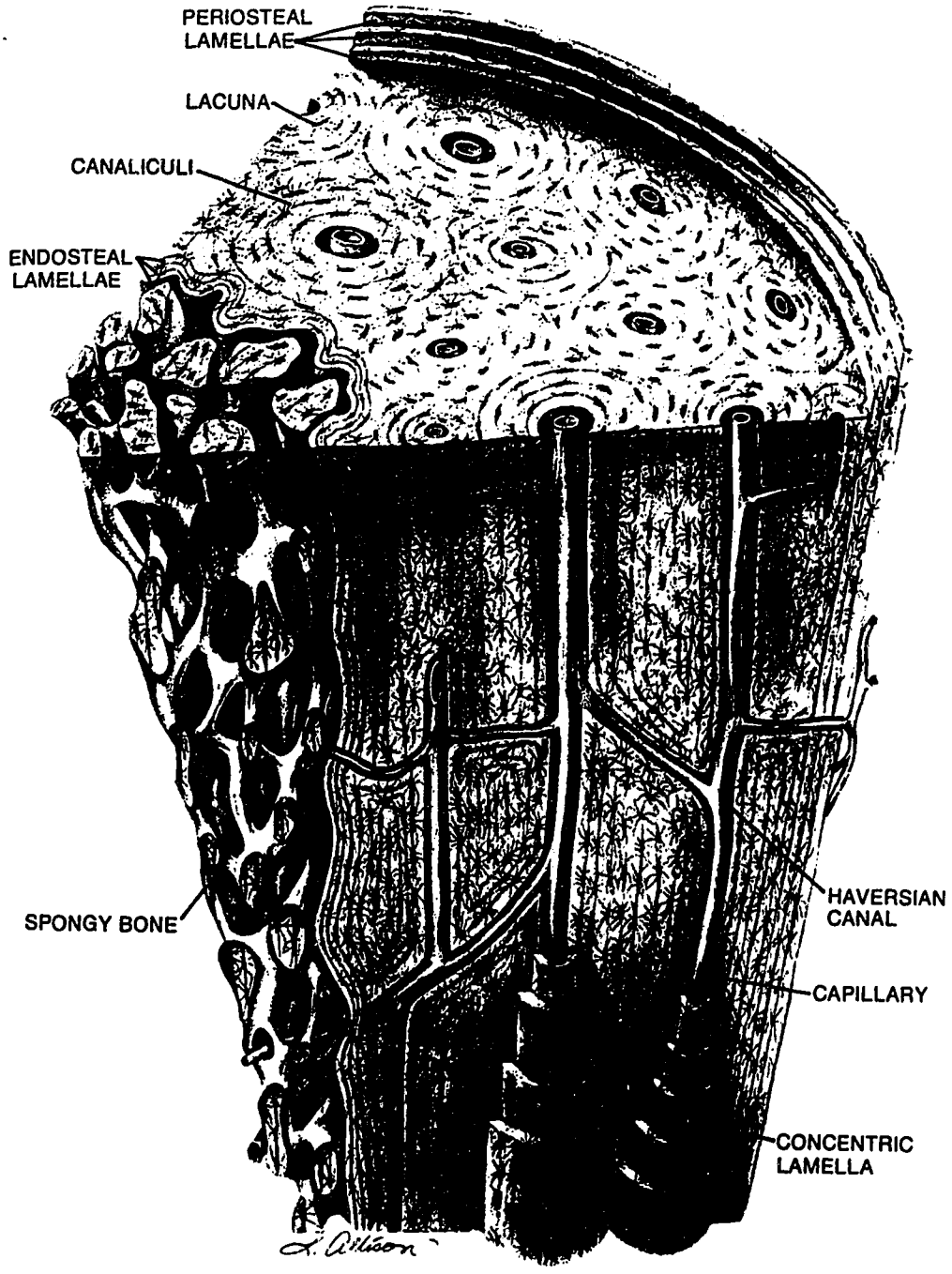


Fig. 2. Diagrammatic representation of a small portion of compact bone. (Leeson et al. 1985:56)

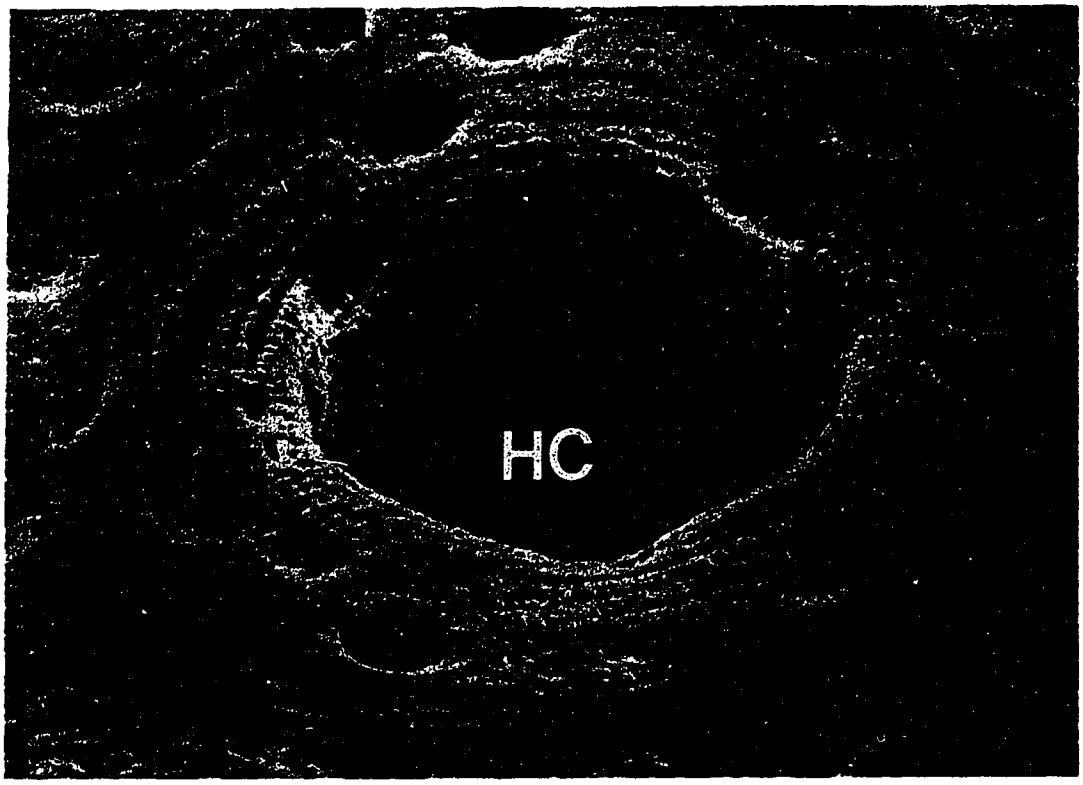


Fig. 3. Haversian canal (HC) with openings of canaliculi (Ca). (Carolina Biological Supply)

concentric lamellae, lacunae, osteocytes, and canaliculi is termed the Haversian system or osteon. Each osteon, which has a diameter of approximately 300 microns, is outlined by a cement shell consisting of modified matrix. Anastomoses that connect the central canals of two osteons are often seen (di Fiore 1989).

Figure 4 shows an area of compact bone that contains a small group of interstitial lamellae. These are the remnants of Haversian systems partly destroyed during internal bone reconstruction, known as osteon remodelling (Leeson *et al.* 1985). This occurs throughout the life of bone and is necessary because bone cannot grow and mature by interstitial expansion and change like other connective tissues (Ortner and Putschar 1985). During osteon remodelling, a linearly advancing group of osteoclasts (bone-resorbing cells) produces a vascularized tunnel which is refilled by osteoblasts (bone-forming cells) differentiating to osteocytes. Thus, resorption and formation occur simultaneously, but at different localities in the tunnel.

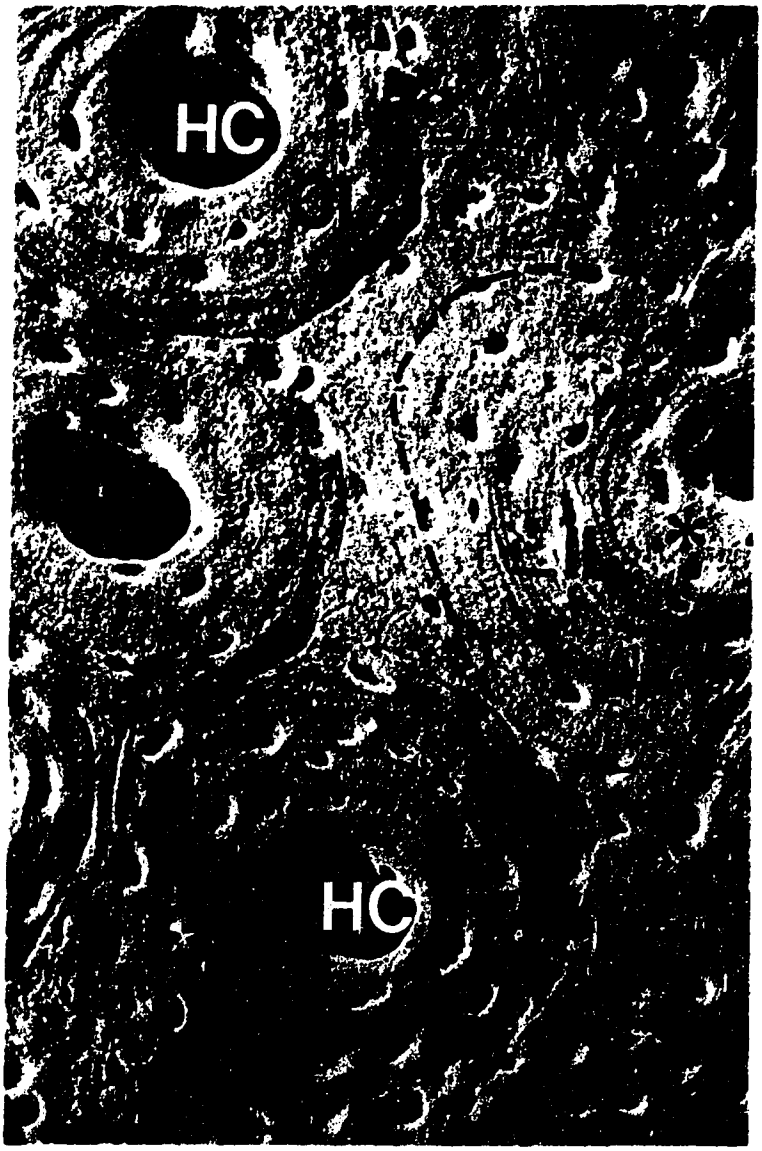


Fig. 4. Interstitial lamellae (IL) with first generation Haversian system (broken line) and second generation Haversian system (*). Cement lines (CL) and osteocyte lacunae (La). (Carolina Biological Supply)

C. Bone Mineral

The complexity of the mineralized matrix of bone is not completely understood (Ortner and Putschar 1985; Posner 1969; Vaughan 1981). The mineral consists of two broadly defined calcium phosphate (apatite) pools: a noncrystalline, amorphous phase and several crystalline phases. These are precipitated in the organic (chiefly collagen) matrix (Fig. 5). Not only can the overall composition of the mineral vary with the age of the individual and the type of bone, but different forms of calcium phosphate can be found at different locations within a single bone (Pate and Hutton 1988). Table 1 reveals the various phases of the calcium phosphate compounds in order of decreasing acidity and solubility and indicates their molar calcium/phosphorus ratios (Neuman 1980). Hydroxyapatite forms the major ingredient and is typically the subject of bone mineral studies (Posner *et al.* 1979). The more soluble (immature) phases are found chiefly at the margins of compact bone, including the surfaces of Haversian canals. After death, the stability of the amorphous phase is weakened and the relative crystallinity of the bone increases.

In reality, the composition of each phase deviates from the stoichiometrically derived values due to ionic interaction with materials present in cellular fluid

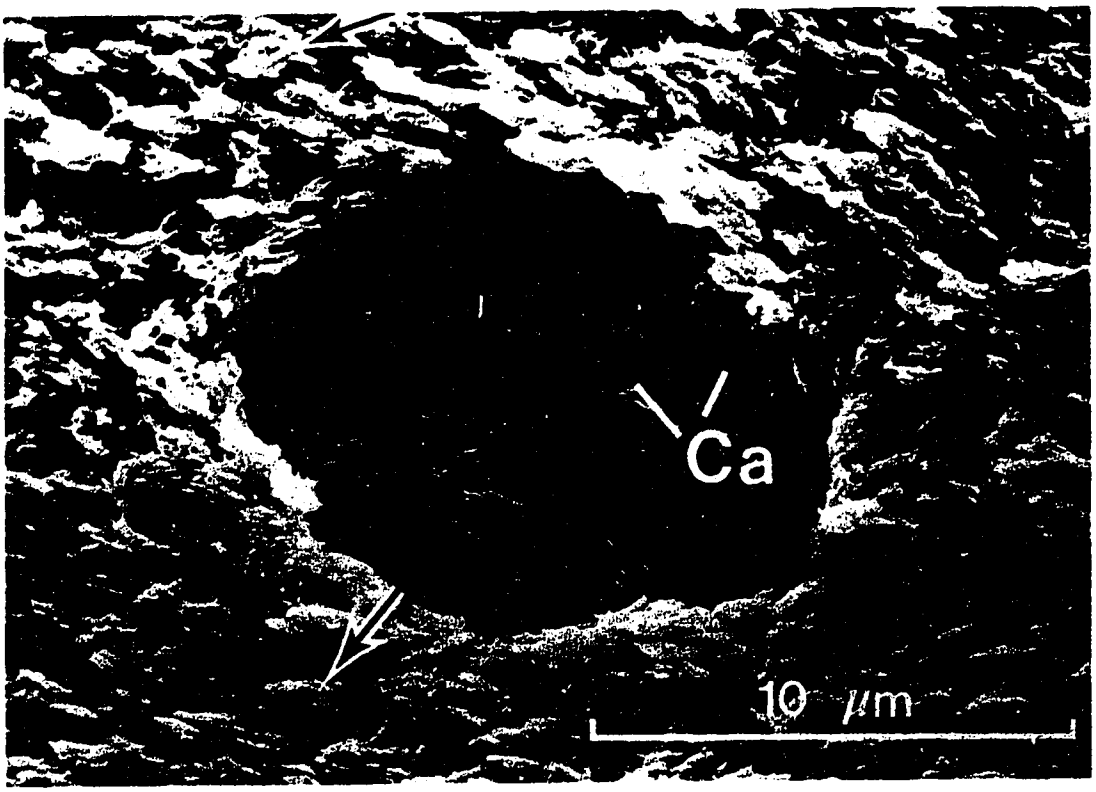


Fig. 5. Osteocyte lacuna with pattern of calcium phosphate crystal formation (arrow). Canaliculi (Ca). (Carolina Biological Supply)

Table 1. Calcium phosphate phases of bone mineral in order of decreasing acidity and solubility, with molar Ca/P ratios. (from data in Neuman 1980)

Dicalcium phosphate dihydrate	$Ca(HPO_4)2H_2O$	1.00
Octacalcium phosphate	$Ca_4H(PO_4)_3$	1.33
Amorphous calcium phosphate	$Ca_9(PO_4)_6$ var.	1.3-1.5
Tricalcium phosphate	$Ca_3(PO_4)_2$	1.50
Hydroxyapatite	$Ca_5(PO_4)_3OH$	1.67

during initial bone mineralization (Neuman 1980) and with those subsequently ingested while the individual is alive (Grupe 1987; Price *et al.* 1985). These materials include carbonate, citrate, and minor and trace elements.[1] Bone acts as a mineral bank (Glimcher 1981; Triffitt 1985), and the most soluble phases of calcium phosphate, which are immediately adjacent to the vasculature, are apparently most prone to this ionic interaction after incipient calcification. During life, however, discrimination can exist for or against the uptake and storage of certain elements, and the composition of bone mineral does not necessarily reflect elemental concentrations in the human circulatory and physiological exchange system as a whole (Grupe 1987; Runia 1988). Thus, not only can the chemical composition of bone mineral vary inter- and intraosseously according to past and current physiological, nutritional, and environmental factors, but the rate of ionic change may differ in various parts of bone at any given time.

The nature of elemental binding and substitution in the crystalline calcium phosphate phases varies according

1. The term "trace element" has no precise definition. In biomedical research, it normally refers to those components present in tissue in concentrations less than 0.01% (anon. 1974; Armelagos *et al.* 1989) or typically measured in milligrams per kilogram (Aufderheide 1989). Trace elements have been categorized as essential, nonessential, or toxic. These designations are subject to change as knowledge of the elements' roles increases.

to the involvement of three different crystal zones: the crystal interior, the crystal surface, and the hydration shell (Vaughan 1981). The last is a layer of water that is believed to be bound to the crystal surface. It is through this layer that transfer of ions between the crystals and other matter in or adjacent to bone occurs. The surface area in question is enormous: 440,000 to 550,000 m^2 per individual (Howell 1971). The presence of this large specific surface, combined with an apparent high surface reactivity rate, results in bone mineral being metabolically very active (Posner and Beebe 1975; Posner *et al.* 1979).

The state of knowledge regarding the exact binding and substitution processes of certain ions has changed little in the last 30 years. In 1958, Neuman and Neuman published a major work dealing with the biochemistry of bone mineral. Part of this work addressed the issues of adsorption and substitution and how various ions otherwise interact with the hydroxyapatite crystal. They differentiated four classes of ion interaction (1958:82-83):

1. Diffusion of ions into the hydration shell occurs, a process which is readily reversible. A steady state is attained in a few hours or less. The ion concentration in the hydration shell is directly

proportional to that in the surrounding solution.

2. Ions that have entered the hydration shell participate in the neutralization of surface charge asymmetry. These anions and cations become part of a bound ion complex. Again, the system is reversible, and a steady state is attained in a few hours. The ion concentration in the hydrated layer is a function of that in the surrounding solution and is relative to the concentration of the displaced ion (calcium, phosphate or hydroxyl).

3. Penetration of ions into the hydration shell and replacement of ions on the crystal surface (adsorption) takes place. Participation in the neutralization of surface charge asymmetry may or may not occur. The system is reversible, and a steady state is reached in a few hours. The adsorbate concentration in the hydrated solid is a function of that in the surrounding solution and is relative to the concentration of the displaced ion of the adsorbent.

4. Displacement of lattice ions both on the crystal surface and within the crystal occurs (substitution). This appears to be irreversible. A steady state cannot be demonstrated over a period of several months. The substituent concentration in the solid phase of the crystal is a function of that in the surrounding solution

and is relative to the concentration of the displaced ion and the duration of the equilibration.

Table 2 summarizes Neuman and Neuman's (1958) data regarding the probable *in vivo* involvement of major ions in the three hydroxyapatite crystal zones of modern bone, *i.e.*, of bone alive in the atomic age. A review of contributions on the chemistry of bone mineral (Dallemagne and Richelle 1973; Davies *et al.* 1979; Eanes and Posner 1970; Engstrom 1972; Hancox 1972; McConnell 1973; Neuman 1980; Posner 1969; Posner *et al.* 1979; Triffitt 1985; Urist 1976; Vaughan 1981; Zipkin 1970) reveals that this issue has not been at the forefront of more recent research. This may be related to the decrease in atomic fallout studies in the period since the 1950s.

Nevertheless, it may be stated with certainty that the specificity of an ion adjacent to the hydration layer determines the variable involvement of the three crystal zones and, consequently, whether surface adhesion, or surface or internal interchange, will result (Neuman and Neuman 1958; Vaughan 1981). Newesely (1988) defines this specificity in terms of physical space, factors of crystallographic symmetry, polarization behaviour, and the lattice space oxidation state of the crystal. An awareness of these factors can be important when

Table 2. Probable ion participation in hydroxyapatite crystal interaction. (from data in Neuman and Neuman 1958)

Ion	Hydration shell		Crystal surface	Crystal interior	Ion displaced
	Water only	Bound ion layer			
K^+	+	-	-	-	-
Na^+	+	-	+	-	Ca^{2+}
Mg^{2+}	+	?	?	?	(Ca^{2+})
UO_2^{2+}	+	+	?	-	Ca^{2+}
Sr^{2+}	+	+	+	+	Ca^{2+}
Ra^{2+}	+	+	+	+	Ca^{2+}
$^{45}Ca^{2+}$	+	+	+	+	Ca^{2+}
Cl^-	+	-	-	-	-
$Citrate^{3-}$	+	?	?	-	(PO_4^{3-})
CO_3^{2-}	+	+	+	-	PO_4^{3-}
$^{32}PO_4^{3-}$	+	+	+	+	PO_4^{3-}
F^-	+	?	+	+	OH^-

considering the potential interaction with bone mineral of ions that have not been studied as intensively as those in Table 2. While any ion will presumably enter the hydration shell, substituents must generally satisfy the requirements of specificity just mentioned, especially oxidation state equivalence. However, in the case of coupled substitutions, entering cations or anions need only to satisfy overall oxidation state parity (*e.g.*, $2Ca^{2+}$ can be exchanged for Na^{+} and Y^{3+}, or $2PO_4^{3-}$ for SiO_4^{4-} and SO_4^{2-} [Newesely 1988]). For all practical purposes, when replacement occurs, the restraints of specificity result in ordered replacement of lattice ions as follows: cations displace calcium, multivalent ions displace phosphate, and anions, typically F^- or Cl^-, displace hydroxyl (Herrmann and Newesely 1982; Newesely 1980; Posner 1969).

The preceding discussion of ion involvement with bone mineral has dealt exclusively with the main ingredient of the inorganic part of formed bone, hydroxyapatite. The nature of reactions of materials with the amorphous phase of calcium phosphate has not been explored, for the spatial organization of ions in this substance is unknown (Vaughan 1981). Indeed, the very existence of this phase has been questioned recently (Bonar *et al.* 1985; Grynpas *et al.* 1981). Nor have

distinct observations been made of ion interchange with the other crystalline phases of apatite.

Using this discussion as background, the following chapter will examine the problems and considerations associated with the attempt to identify biogenic signals in the minor and trace element content of bone.

3. PROBLEMS AND CONSIDERATIONS IN TRACE ELEMENT ANALYSIS

A. Introduction

Although the examination of the elemental content of human skeletal remains has been of value in palaeopathological research (Grupe 1988b; Hisanaga *et al.* 1988; Rogers and Waldron 1985), in problems involving separation of individuals (Aufderheide *et al.* 1988; Brätter *et al.* 1980), and in the identification of visually ambiguous tissue residues (Bethell and Smith 1989; Henderson *et al.* 1987), it is in the reconstruction of diet that the exercise has seen the greatest application. Characterization of diet by this means has also led to corroboration of postulated social ranking in historic and prehistoric societies (Aufderheide *et al.* 1985; Schoeninger 1979) and the ascertainment of reproductive status or, correspondingly, age of weaning (Blakely 1989; Grupe 1986a; Sillen and Smith 1984). While artifactual, faunal, and botanical evidence from an archaeological site can indicate which foodstuffs were available, determination of the presence, absence, or relative amounts of certain minor and trace elements in bone can, within limitations, point to what was actually consumed. The major trio of these substances currently consists of strontium, barium, and zinc, although sodium

may also be of importance (Price *et al.* 1989). While strontium and barium decrease with rising trophic level, zinc and possibly sodium increase in quantity.

Trace element analysis, with stable isotope assay, has greatly enhanced the possibilities of discovering the diet and, less often, the nutritional status of individuals as well as populations, a field formerly limited to examinations of certain pathological conditions or, rarely, fecal matter. The technique, however, is not without problems.

B. Intrinsic Complications

The search for certain trace elements in human bone, no matter what the goal, takes place against a background of bone mineral composition that is vastly more complex than the expected formula for hydroxyapatite. As was discussed in the previous chapter, both materials present in cellular fluid during the calcification process *in utero* (Neuman 1980), and those subsequently ingested or otherwise incorporated during life (Grupe 1987), may be prone to interaction in one or more of the four outlined processes of diffusion or exchange. The nature and degree of this interaction is dependent on the availability of elements to the organism and the

physiological reaction to them. Availability is chiefly a function of the characteristics of the local geochemical environment through the media of food, water, and air (Curzon 1983; Runia 1988). As trace element incorporation into an organism is primarily via a food chain originating in soils, the chemical composition of tissues varies from locality to locality. Reaction involves the processes of discrimination for or against the uptake into bone of certain elements, with the result that the composition of bone mineral does not necessarily reflect the activity and significance of all elements in the human organism as a whole.

The potential indicators of diet that are sought must not only be known to be subject to deposition in bone, but must also be present to a degree that conveys information, *i.e.*, that allows differentiation from the normally complex, but not necessarily informative, compositional background. In some cases, the absence of a certain element may also be revealing. A comparison of this nature is rendered difficult by the great variability in the type and quantity of elemental ingredients in bone stemming from variation in availability. Because of this, there can be no fixed reference of elemental content against which potential diet-indicating signs could be strictly measured.

Furthermore, the possible nutritional role and origin of many elements is either poorly understood or unknown (Bowen 1979), thereby often limiting their value as sources of information. Exceptions to this limitation include two of the most important elements for palaeodiet reconstruction: strontium and barium. Although there is some evidence for the interference of strontium with vitamin D metabolism (Wasserman 1974), both elements are presently designated nonessential and nontoxic, for their functions, if any, are unknown (Aufderheide 1989). Thus, although substances may be indicators of diet by virtue of the degree of their presence in ingested foodstuffs, they may not necessarily be indicators of nutritional or health status. The question of essentiality and nonessentiality is further discussed below.

Keeping in mind the relevance of these two major problems of variability and lack of much basic knowledge, one may proceed to examine the degree of diversity of elements present in bone. This is required as a steppingstone to identification both of substances that may disclose their dietary origins and, unavoidably, of further difficulties inherent in the process. Bowen (1979) has collected data from several studies concerned with the determination of element type and quantity in modern human bone. His compilation is summarized in

Table 3. While giving a good picture of the complexity of osseous tissue, such data are, unfortunately, often of limited worth for archaeological research. Specific complicating factors that are illuminated by these data and that stem from the problem of compositional variability are as follows:

1. Single values in the table indicate either single analyses or means for which the number of samples is not given. Thus, it may be impossible to evaluate statistical significance in comparisons. This is basically a problem of insufficient research or inadequate reporting of data.

2. Extreme ranges (*e.g.*, La, Sc, U) provide information that is too vague for comparison. Unless the metabolic pathway of such an element is understood, little of significance may be extracted from a range of such magnitude.

3. The elemental content of modern bone may differ from the antemortem composition of prehistoric bone, especially with respect to substances present in modern industrial pollutants and atomic fallout, *e.g.*, cadmium and strontium (Dehos and Schmier 1985; Jaworowski *et al.* 1985).

4. The distribution of elements between the organic and inorganic parts of bone is often unknown. This may

Table 3. Elemental composition of dry human bone (mg/kg). (from data in Bowen 1979)

Ag	0.01-0.44	Lu	<0.08
Al	4-27	Mg	700-1800
As	0.08-1.6	Mn	0.2-100
Au	0.016	Mo	<0.7
B	1.1-3.3	N	43k
Ba	3-70	Na	10k
Be	0.003	Nb	<0.07
Bi	<0.2	Nd	<0.28
Br	6.7	Ni	<0.7
C	360k	O	285k
Ca	170k	P	62k-71k
Cd	1.8	Pb	3.6-30
Ce	2.7	Ra	4×10^{-9}
Cl	900	Rb	0.1-5
Co	0.01-0.04	S	500-2400
Cr	0.1-33	Sb	0.01-0.6
Cs	0.013-0.052	Sc	0.001-6
Cu	1-26	Se	1-9
Dy	<0.36	Si	17
Er	<0.28	Sm	0.23
Eu	0.032	Sn	1.4
F	2k-12k	Sr	36-140
Fe	3-380	Ta	0.03
Gd	<0.4	Tb	0.05
H	52k	Th	0.002-0.012
Hf	0.12	Tl	0.002
Hg	0.45	Tm	<0.08
Ho	<0.08	U	0.00016-0.07
I	21	V	0.0035
K	2.1k	W	0.00025
La	<0.08-9	Y	0.07
Yb	<0.28		
Zn	75-170		
Zr	<0.1		

or may not have an effect on analysis for biogenic signals, depending on whether or not differential degrees of postmortem change in the two fractions of a bone sample can be expected.

5. No allowance is made in such tabulations for sex or age specificity. Although any difference in values between the sexes may ultimately relate to differences in diet, there may also be physiological factors involved (Sillen and Kavanagh 1982). Trace element content may also vary with age (Buikstra *et al.* 1989), and at least one component, calcium, follows a decline that is both age- and sex-related.

The factor of essentiality, mentioned above, may also complicate the issue. Essential trace elements are homeostatically controlled neither to fall below nor to rise above certain levels, even in storage (Grupe 1986b). Increased intake of these elements above the upper level presumably leads to increased excretion rather than increased storage in tissues. This process, when functioning normally, effectively limits the range of variability. Thus, the essential elements observed in bone may be less useful for dietary characterization than the nonessential ones, the bodily concentrations of which are not as subject to regulation in this manner. For evaluation of nutritional adequacy, however, essential

trace elements would be the indicators of choice. The presence of the third class of elemental substances, toxins, would be of interest in any quantity.

A further factor to consider, especially when evaluating multiple elements, is the variability in turnover rates. Table 4 provides estimated residence times in the human body for 22 elements. While analyses of more than one element can be advantageous (Buikstra *et al.* 1989), assessing dietary shifts may be problematic due to the large difference in the treatment of some elements by the body.

Finally, research may also be confounded by the variability in distribution of elements throughout the skeleton. Brätter *et al.* (1977) found that elemental content appears to be related to the functional and structural conditions of the sampling site. Larger amounts of many elements are present in epiphyseal areas, and the content of a bone sample depends largely on its cancellous/cortical mass ratio. Knowledge of this variability has led some researchers to stress the importance of consistent sampling strategies when analyzing for substances in bone (Grupe 1988a).

In summary, assigning significance to the amount of minor and trace elements in bone cannot be done meaningfully without realization of the current state of

Table 4. Residence times in the adult human body (days). (from data in Bowen 1979)

Element	Residence time
Hg, K, Mo	50-100
Co, Cs, Mn, Rb	100-200
As, Sb, Se	200-400
Cd	400-800
Ba, Ca, Cu, Ni, Ra, Sn, Sr	800-1600
Fe, Pb, Th, Zn	>1600

understanding of their roles and the factors affecting their deposition *in vivo*. These factors include chiefly variable availability and physiological discrimination, as well as behaviour and distribution of elements within bone, possible physiological consequences of essentiality and nonessentiality, and differences in elemental content of bone related to sex and age.

C. Extrinsic Complications

At death, the factors that may result in variability in the elemental content of living bone cease to be of direct influence. They are replaced, however, by developments that, for the researcher examining such content for palaeodiet reconstruction, are generally of even greater consequence. These developments fall under the rubric of dead bone decomposition and may involve both biogenic and nonbiogenic forms of chemical alteration. Gross physical changes in bone, in the form of deterioration or visible contamination, may or may not provide evidence of the nature and extent of alteration at the chemical level and thus in themselves are not of primary importance.

Biogenic decomposition resulting in chemical alteration normally begins soon after death and plays a

major role in the deterioration of bone (Baud and Lacotte 1984; Grupe and Piepenbrink 1988; Piepenbrink 1986a). This process may take the form of autolysis, bacterial putrefaction, or fungal or algal invasion. Autolysis, normally the first phenomenon to occur after life functions have ceased (Ascenzi 1986), is caused by the activation and release of intracellular enzymes and results in the partial or complete disappearance of marrow cells, osteoblasts, osteoclasts, and the cells of the vasculature. The effect of this development on the trace element content of the organic and inorganic fractions of bone is unclear, but may assumed to be minimal, as the hard tissues are initially not involved. Autolysis does, however, partly catalyze subsequent chemical changes that are discussed below.

Bacterial putrefaction, which results in the enzymatic decomposition of proteins, is usually the first form of deterioration at least partly involving invasion of foreign matter from the environment of the remains. Usually eliminating those cellular structures not removed by autolytic enzymes, putrefaction initiates a process of collagen breakdown that may ultimately have consequences for analyses involving the protein (Dennison 1980) and the associated mineral parts of bone. The resulting amino acids and other by-products of this reaction are

generally leached out of bone in a burial context (Lynch and Jefferies 1982). The effects of such bacterial action on trace elements associated with the hydroxyapatite crystal and the magnitude of their possible removal from bone are not known.

The most substantial biogenic deterioration of bone is caused by certain types of fungi, algae, and bacterial microorganisms invading hard tissue either by exploiting existing spaces in the structure of bone or by tunnelling (Hackett 1981; Piepenbrink 1986b). The boring of tunnels is effected by bone-dissolving metabolites that are secreted by fungi and possibly to a lesser extent by bacteria (Marchiafava *et al.* 1974). In addition to upsetting attempts to assess age histomorphometrically (Bruchhaus 1987; Lazenby 1984), tunnelling and void exploitation can introduce substances from the environment (Piepenbrink 1984). Especially barium seems to be prone to transport in this manner (Grupe and Piepenbrink 1988). Such materials may disrupt any trace element signal originating in diet both by their mere presence and their potential for exchanging with ions in the hydroxyapatite molecule. Furthermore, continued amorphous dissolution of the inorganic part of bone around the tunnels and voids may occur as a result of the generally acidic milieu produced by the secreted

metabolites. Under some circumstances, dissolved or otherwise altered mineral may recrystallize as a different compound (Fig. 6). On a gross level, fungal and algal invasion can cause fissuring and splitting of bone, facilitating increased incursion of foreign matter. Thus, what begins as encroachment by microorganisms can result in contamination, ion exchange, diagenesis, and destruction.

Microorganisms can also impregnate bone with chromophores and fluorophores (Keith and Armelagos 1988; Piepenbrink 1986a). Bone thus affected may not only be contaminated with certain elements but become discoloured or fluoresce, leading to misinterpretations regarding the origins of these phenomena. Piepenbrink (1986b) believes that especially postmortem fluorochroming may cause confusion, for similar effects may occur after ingestion of tetracycline or grain affected by certain moulds.

Nonbiogenic postmortem chemical alteration of bone potentially provides the greatest source of inaccuracy in trace element determinations. The chemical breakdown and restructuring of bone, possibly already in progress to a more minor extent as a result of the biogenic factors noted above, can be strongly promoted by contact with inorganic substances in the burial environment and can involve a number of processes: simple contamination

Fig. 6. Fungus hyphae with recrystallization in a human long bone fragment. (Schultz 1986: 99)

through occupation of pores and voids in bone (Fig. 7), dissolution of bone mineral, precipitation of some resulting compounds out of solution, obvious or latent recrystallization of others (Fig. 8), or ionic adsorption or substitution. One or a combination of these phenomena may considerably alter the elemental composition of bone between the time of interment and chemical analysis (Pate and Hutton 1988). Moreover, at the moment of retrieval, the chemical nature of the structure may vary within a single bone, depending on differing degrees of exposure to contamination and on how the different phases of calcium phosphate have reacted.

The most important source for contamination resulting in alteration, or the depository for unidirectional leaching out of bone, is the immediate environment of the bone. This is normally soil or water, or both. According to Pate and Hutton (1988), the most important variables to consider when the environment is soil are pH, chemical composition, and the amount, type, and pressure or flow of any groundwater. In water, these variables would also be fully applicable. It appears that combinations of these factors can result in extremely complex and varying effects that probably differ somewhat for practically every burial and immersion context (Nelson and Sauer 1984). The

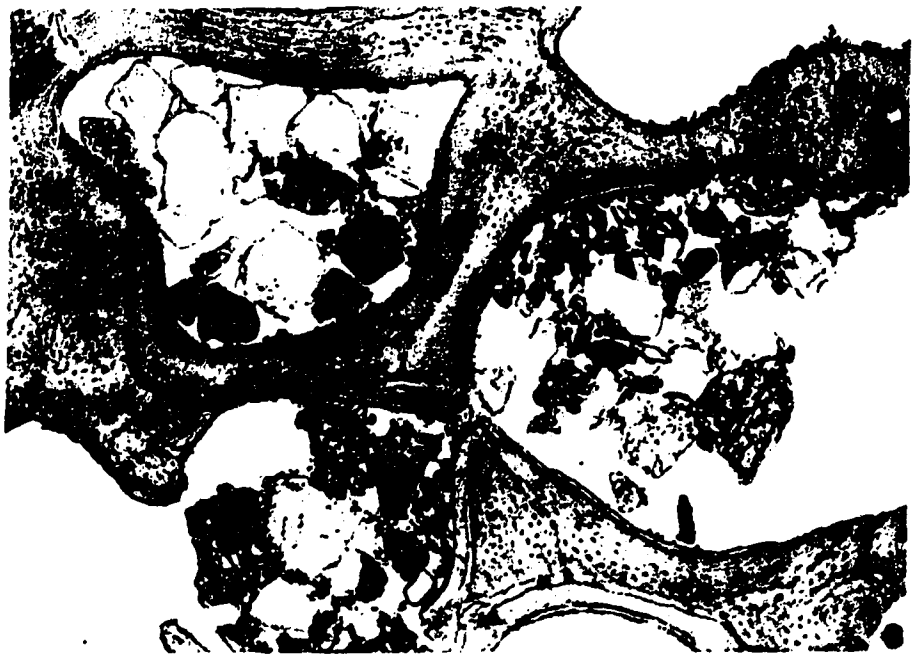


Fig. 7. Void occupation by soil particles in metaphyseal bone from a mammoth femur (x25). (Schultz 1986: 85)

Fig. 8. Recrystallization in a Haversian canal in human bone. (Schultz 1986: 86)

possibility of different conditions over time can further complicate the matter. However, a few generalizations may be made regarding pH and the transfer of several elements of interest.

As noted above, the excretions of microorganisms involved in the destruction of both the organic and inorganic parts of bone often produce a milieu of relatively low pH (Piepenbrink 1984). Consequently, the integrity of the protein-mineral bond may be greatly affected and stability decreased (Hanson and Buikstra 1987; Von Endt and Ortner 1984). This generally universal process of degradation within bone may be promoted, retarded, or halted by the pH of the surrounding soil or water and the degree to which the bone is prone to its influence.

The creation of an acidic microenvironment by a combination of a biological process and a low soil pH can result in considerable alteration of the mineral of bone itself (Herrmann and Newesely 1982). As microbial or fungal activity and an acidic burial context begin to dissolve the calcium phosphate phases in bone, calcium is released from the mineral (Grupe and Piepenbrink 1988; White and Hannus 1983). The less soluble phases of calcium phosphate (*cf.* Table 1) regress, if they precipitate and recrystallize, to more immature phases

due to the relative loss of the calcium ion to the typically calcium-poor nature of acidic soils. The degree of this change depends on the degree of acidity. According to Herrmann and Newesely (1982), the typical result of this process in a microenvironmental pH of 4.5 to 6.0 is the conversion of hydroxyapatite ($Ca_5[PO_4]_3OH$) to dicalcium phosphate dihydrate (brushite) ($Ca[HPO_4]2H_2O$) (Fig. 9). In a somewhat less acidic milieu (pH 6.0 to 7.0), hydroxyapatite may be transformed into octacalcium phosphate ($Ca_4H[PO_4]_3$). The more acidic the microenvironment, the greater the friability of the affected bone will be. Carried to its limit, this may result in bone losing all structural stability and crumbling, for the more immature the calcium phosphate phase, the more prone it is to structural collapse.

As in the case of fungal and algal invasion, the formation of brushite can result in mechanical destruction of bone (Herrmann and Newesely 1982). This effect, which may occur in conjunction with the crumbling caused by friability, is due to brushite expanding as it crystallizes and subsequently cracking the lamellar structure. Furthermore, in an earlier phase of this phenomenon in an absence of extracorporeal water, crystals of brushite may grow on the surface of bone, resulting in an appearance not dissimilar to that of

(copyright permission unavailable)

Fig. 9. Brushite formation in cranial bone. (Herrmann and Newesely 1982:23)

hoarfrost. Such surface growth may also be confused with postmortem adipocere formation caused by hydrolytic hardening of body fats in more recent remains (Herrmann 1981). This crystallization occurs commonly in crypts and vaults, where corporeal water has been sufficient to produce an acidic milieu and cause the dissolution of parts of the mature phases of calcium phosphate, but where the actual environment is relatively dry.

In a situation where environmental water is of such a quantity and endurance that recrystallization does not occur, and the milieu remains acidic, the inorganic part of bone may disappear entirely. In aerobic conditions, where bacterial and/or fungal activity is prevalent, the organic matrix would also vanish due to enzymatic catabolism and acidity. In unusual anaerobic surroundings, the result may be preservation of the organic portion not affected by autolytic processes, but dissolution of the mineral. The so-called "bog bodies" are an example of this phenomenon (Piepenbrink 1986b).

In some neutral to alkaline burial contexts, dissolution and ionic exchange initially caused by biogenic acidity is retarded and often halted (Gordon and Buikstra 1981; Hanson and Buikstra 1987; Pate and Hutton 1988). The lack of soil acidity can result in the calcium phosphate phases remaining more stable. However,

just as protein preservation may still be good even in the presence of acidic demineralization, gross structural integrity in some alkaline soils does not necessarily indicate biochemical integrity. Biogenic contamination may continue unabated in both acidic and alkaline contexts. Porous areas may be filled with soil. Calcite ($CaCO_3$) and sodium carbonate (Na_2CO_3), common in alkaline soils (Nelson 1982), may also fill voids in bone and result in overall enrichment of calcium and sodium. In dry contexts, this may promote gross structural stability (White and Hannus 1983). In the presence of even moderate amounts of groundwater, however, a degree of mineral dissolution and permeating contamination may occur regardless of soil pH. Carbonates may exchange with calcium phosphate (Grupe 1986b; Piepenbrink and Schutkowski 1987), and the bone may assume the consistency of chalk. Further degradation may result in disappearance.

In both acidic and alkaline contexts, then, the integrity of any trace element signature may be jeopardized. In the former, partial dissolution or a combination of dissolution and chemical reconstitution of bone mineral in the presence of contaminating soil or water can result in fundamental changes in the overall chemical composition of a piece of bone. In the latter

context, dryness may preserve bone intact, but the presence of contaminating and dissolvent water renders the advantage ineffectual.

The question of mineral transfer and ionic adsorption and substitution is obviously by no means unrelated to that of pH. As bone and environment come into contact with each other, a bidirectional flow of elements may exist. As noted, the relative degree of acidity and alkalinity can greatly affect the nature and direction of this flow. From the viewpoint of trace element analysis, the major consideration is net loss or gain of those substances that can reveal their dietary origins.

Strontium readily replaces calcium in hydroxyapatite at all four levels of crystal interaction (Table 2) and has been characterized as a bone-seeking element (Schroeder *et al.* 1972). Its tendency to substitute for calcium within the crystal interior gave initial impetus to the assumption that it remained relatively stable after death (Parker and Toots 1980). The fact that practically all of body strontium that is not excreted is deposited in bone mineral (Nielsen 1986) lent further credence to the belief that the strontium content of archaeological bone could infallibly inform the researcher regarding the diet of the deceased. However,

the bone-seeking behaviour of strontium does not cease with death. More recent research has shown that interred bone can act as a strontium sponge, exchanging postmortally with calcium if it is present in contaminating soil (Rado[illegible]89; Sealy and Sillen 1988). In such a case, i[illegible] washing of bone samples, as has been [illegible]y Sillen (1986), would do little or nothing to remove the strontium that has undergone heteroionic exchange (Tuross *et al.* 1989). If strontium is present in soil, the strontium content of bone buried in that soil cannot be assumed to be purely biogenic, especially if the biochemical stability of the bone is jeopardized by adverse pH conditions.

Barium, like calcium and strontium a member of the IIa group of elements and thus possessed of the same charge, is less well understood than strontium but is considered to behave similarly (Grupe 1986b; Sillen and Kavanagh 1982). It, too, is bone-seeking and is present chiefly in the mineral part of bone (Brätter *et al.* 1977). The presence of barium in soil from a burial context must therefore also be considered as a warning signal when determination of the barium content of bone is planned (Lambert *et al.* 1984b). Moreover, as noted above, this element appears to be especially prone to transport into bone by various types of fungus (Grupe and

Piepenbrink 1988).

Zinc, unlike strontium and barium, is essential to human metabolism and thus under homeostatic control (Hambidge *et al.* 1986). More than 90% of bone zinc is present in the mineral fraction (Brätter *et al.* 1977). An expected relatively narrow range of variation of this essential element's content in bone is borne out by the data in Table 3 (39% for zinc, compared to 59% for strontium and 92% for barium), although this cannot be taken as firm support due to the inadequate reporting method in this table. Postmortem enrichment or depletion may be easier to identify for such an element with a more controlled presence in the living body.

The hydroxyapatite binding position for zinc is disputed. As a cation, it would theoretically interact with calcium. Vincent (1963) believes it does not undergo substitution, while Lappalainen *et al.* (1982) claim it forms an integral part of the molecule. Lambert *et al.* (1985) hold that zinc does replace calcium on the basis of artificial inducement experiments, a process that may not, however, parallel physiological conditions. Thus, the relative proneness of this element to postmortem alteration is difficult to estimate. Zinc does not appear to be a bone-seeking substance (Hambidge *et al.* 1986); therefore, it may not be subject to

diffusion toward bone in a burial context to the same extent as strontium and barium. This hypothesis is supported by the findings of Whittaker and Stack (1984) and Byrne and Parris (1987), who report little obvious change in the zinc content of teeth and bones, respectively, from archaeological contexts.

Buikstra *et al.* (1989) indicate that the sodium content of bone may also be a valuable dietary indicator. Strictly speaking not a trace element, the amount of this essential substance varies little in fresh bone (Klepinger 1984), although the value of 10,000 ppm dry weight in Table 3 cannot be taken as fixed. It is not believed to be a bone-seeker like strontium or barium. The postmortem stability of sodium may be less than that of these two elements, for it is believed only to adsorb onto the hydroxyapatite crystal surface (Table 2). Previous studies on the behaviour of sodium in a burial context have produced varying results. Byrne and Parris (1987), Lambert *et al.* (1982), and Parker *et al.* (1974) conclude that the element is leached out of bone, while Edward *et al.* (1984), Henderson *et al.* (1983), and Klepinger *et al.* (1986) observed a net gain of sodium. The ambiguity may result from the general failure in most studies to pay primary attention to the composition, pH, and water content of the burial contexts.

While an observed quantity of one of the above elements in archaeological bone that lies far outside the known range of variation for living bone would in all likelihood be indicative of postmortem enrichment or depletion, it would not be possible to rule out depositional alteration even in the other case, namely the discovery of agreement between the determinations for archaeological and fresh bone. It is possible to envision mass misinterpretation arising from the combination of such an occurrence and ignorance of one or both of soil conditions and the nature of the interaction between individual elements and bone mineral. Indeed, all studies that have proceeded on the assumption that an element present in an "acceptable" amount is of physiological origin are suspect. In an instance where it is not possible to determine from the element itself or to estimate from soil conditions (if known) whether or not postdepositional alteration may have occurred, the presence of certain other elements in the bone sample that are obviously contaminants may point to latent alteration of the diet-indicating substances.

Known contaminating elements that are very common in soils of many types are aluminum, iron, and manganese (Buikstra *et al.* 1989; Lambert *et al.* 1982, 1983, 1984a, 1985). Aluminum can be present in a form that is freely

exchangeable with other cations such as calcium in both acidic and alkaline soils (Barnhisel and Bertsch 1982). Thus, its presence in archaeological bone in an amount well above the range in fresh bone is indicative of not only surface contamination but also substitution of calcium. Iron, too, exists in many soils as an exchangeable cation with a +2 charge (Olson and Ellis 1982), but its possibly wider range of variation in human bone (Table 3) may make it less valuable as an indicator of contamination than aluminum. Exchangeable manganese is present chiefly in neutral to acidic soils and tends to be bound in less soluble oxides in alkaline contexts (Gambrell and Patrick 1982). This, combined with its possibly fairly wide range of occurrence as indicated in Table 3, may also make it less suitable than aluminum for suggesting postdepositional alteration of the diet-indicating elements.

In summary, both biogenic and nonbiogenic factors can cause changes to occur in the chemical composition of interred bone. Autolysis, bacterial putrefaction, and fungal, algal, and bacterial invasion often initiate a process of alteration in elemental content that is promoted or retarded by environmental conditions, depending on contextual pH, chemical composition, and water availability. The chief diet-indicating elements,

strontium, barium, zinc, and sodium, are by no means immune to this process. A knowledge of soil conditions is a key factor in the analysis of archaeological bone, unless the presence of known contaminants can be used as indicators. However, a lack of these contaminants in the absence of soil data cannot be used as proof of the biochemical integrity of a bone sample.

4. MATERIALS AND METHODS

The material examined in this study was excavated in Harappa cemetery R37 in 1987. Excavation staff cut and retained femoral midshaft cross-sections from both primary and secondary context human remains and collected soil from within burial pits, primarily for use in elemental and isotopic analyses. The provenience of faunal remains was also noted as to burial or secondary depositional context.

The samples were identified by codes expressing project, year of excavation, provenience (lot and feature numbers), and, in the case of commingled remains, bone numbers. These codes are used for identification purposes throughout this study. Table 5 lists the material made available. All human bone samples are adult femoral midshaft cross-sections except H87/197-194b, which is a neonatal tibia from an individual in contact with skeleton H87/197-194a. All samples underwent analysis except the human bone from feature H87/40-34a, which was found to be impregnated with an unidentified glue or preservative.

The technique selected for the examination of both bone and soil was instrumental neutron activation analysis (INAA). This method was chosen for several reasons. First, INAA is a multielement technique

Table 5. Identification of available material.

Number	Human	Faunal	Soil
H87/40-34a	(*)[1]	*Bos*	
H87/60-46a-26	*	*Bos, Canis*	
H87/60-46a-77	*	*Bos, Canis*	
H87/85-49d.2	*	*Bos*	*
H87/136-147a	*		*
H87/137-148a	*		*
H87/197-194a	*		
H87/197-194b	*		

1. Unsuitable for analysis.

suitable for determining the presence, both qualitatively and quantitatively, of a large number of elements (Willard *et al.* 1988). Second, the physical state of the sample has little effect on the results. Third, the technique is extremely sensitive for most elements. Fourth, the technique is nondestructive, thus allowing sample reuse if required. Fifth, a SLOWPOKE research nuclear reactor and associated laboratories were accessible and affordable. Sixth, INAA has been shown to be an excellent technique for trace element analysis of bone and soil (*e.g.*, Grynpas *et al.* 1987; Hancock *et al.* 1989; Salmon and Cawse 1983).

While previous studies of gradients of postmortem change across a bone sample have used the method of electron probe microanalysis (EPMA), this investigation used INAA throughout. Three factors supported the use of INAA over EPMA for the gradient study. First, samples of bone may experience burning with the latter technique (M. Wayman, pers. comm.). Moreover, the chemical composition of a substance may be altered through the application of intense heat (Grupe 1988b). Second, the sensitivity of EPMA is in many cases insufficient for the quantification of trace elements. Third, methodological consistency contributes to the reliability of results.

Initial attempts were made with a coring drill bit

to extract from the surface of a femoral cross-section a series of subsamples, the analysis of which would presumably reveal any gradient of change in elemental content from the periosteal to the endosteal surfaces. This failed due to the inability of the bit to produce a discrete, intact core. It was then decided to cut subsamples manually with a jeweller's saw. After ultrasonic cleaning of blades in ethanol for approximately one minute, obvious gross surface contamination was abraded away from the femoral cross-sections according to the recommendation of Lambert *et al.* (1989). The blades were recleaned, and three consecutive subsamples were cut in a row from the endosteal to the periosteal surfaces of the cross-sections. A larger number of subsamples was not possible due to the physical limitations of the cutting method. These three subsamples were identified as E, C, or P, depending on whether they were cut from the endosteal, centre, or periosteal part of a femoral cross-section. As human bone sample H87/85-49d.2 was heavily contaminated with an unidentified glue or preservative, the E and P subsamples could not be cut. The small size of human bone sample H87/197-194b precluded the cutting of consecutive subsamples from it. Thus, an entire cross-section was cut and used for analysis.

As the faunal samples were not homogeneous as to skeleton sampling locus and consisted chiefly of trabecular bone, only single subsamples were cut from each after removal of obvious gross surface contamination. After completion of cutting, a blade sample was recleaned and included for analysis in order to control for the possibility of contamination of the bone by the metal of the blade.

In order to drive off any moisture, the relative mass of which may have varied from sample to sample and thus interfered with elemental quantification, the bone, soil, and blade samples intended for analysis were heated at approximately 104 degrees C. for approximately 18 hours. The samples were then weighed to the nearest 0.0001 g (Table 6, column 1) and encapsulated in polyethylene vials that had previously been cleaned for approximately 24 hours in 5% nitric acid in order to remove any residue from the manufacturing process. In the case of bone and blade samples, small empty vials were placed in the larger main vials as filling material so that the samples would remain in the same location throughout the analytical procedure. In the case of the soil, smaller vials containing the samples were placed within the main vials. All main vials were then labelled with indelible ink (Faber-Castell OH-LUX 513 blue). One

Table 6. Sample masses (g).

Sample	1. Initial	2. Subsequent	3. Adjusted
H87/40-34a *Bos*	.4204	.4995[1]	.4183
H87/60-46a *Bos*	.2411	.2526	.2116
Canis	.3587	.3905	.3271
H87/60-46a-26 E[2]	.1299	.1461	.1224
C[3]	.1394	.1590	.1332
P[4]	.2009	.2248	.1883
H87/60-46a-77 E	.1936	.2300[1]	.1926
C	.1115	.1223	.1024
P	.2286	.2440	.2044
H87/85-49d.2 C	.1260	.1110	.0930
Bos	.4337	.5185[1]	.4343
soil	.3201		
H87/136-147a E	.1135	.1239	.1038
C	.1669	.2008[1]	.1682
P	.2721	.2971	.2488
soil	.3475		
H87/137-148a E	.2336	.2658	.2226
C	.1501	.1654	.1385
P	.2973	.3381	.2832
soil	.1520		
H87/197-194a E	.1267	.1344	.1126
C	.0990	.0914	.0765
P	.1385	.1497	.1254
H87/197-194b	.2442	.2400	.2010
Saw Blade	.0440	.0514[5]	.0440

1. No apparent loss of mass during repackaging.
2. Endosteal portion of femoral cross-section.
3. Centre portion of femoral cross-section.
4. Periosteal portion of femoral cross-section.
5. No apparent loss of mass during repackaging; adjusted to initial mass.

cleaned, empty, labelled vial was included in order to be able to address the possibility of the composition of the ink interfering with analysis.

An irradiation strategy was chosen by the reactor staff on the basis of the half-lives of certain elements of primary interest. For reasons outlined in chapter 3, part C, this strategy was developed with the elements aluminum (Al), barium (Ba), iron (Fe), manganese (Mn), sodium (Na), strontium (Sr), and zinc (Zn) in mind. As a major constituent of bone heavily involved in exchange processes and thus also potentially useful for evaluating postdepositional alteration, calcium (Ca) was considered as well. It was not possible to determine the relationship of calcium to phosphorus, the other main ingredient of bone, for this element is not detectable by the techniques used here. The nuclear properties of the elements of interest are given in Table 7 in order of increasing half-life.

The irradiation and counting procedure was as follows:

- a first irradiation of 30 s at a flux of 1 x 10^{11} neutrons/cm^2 s with an initial counting period of 60 s after a 30 s decay, and a second counting period of approximately 240 s after a decay of approximately 4 h;

- a second irradiation of 1 h at a flux of 1 x 10^{12}

Table 7. Nuclear properties of elements of interest (in order of increasing half-life).

Element	Employed Radioisotope	Half-life (s)[1]	Primary Gamma Ray Energy (keV)
Al	^{28}Al	134.4	1781
Mn	^{56}Mn	9282.6	847
Na	^{24}Na	52,772.4	1368
Ca	^{47}Ca	391,910.4	1297
Ba	^{131}Ba	1,019,520.0	496
Fe	^{59}Fe	3,844,454.4	1099
Sr	^{85}Sr	5,602,176.0	514
Zn	^{65}Zn	21,090,240.0	1115

1. Browne and Firestone (1986).

neutrons/cm^2 s with an initial counting period of approximately 1200 s after a decay of approximately 6 days, and a second counting period of approximately 10,000 s after approximately 21 days.

The gamma-ray spectrometer used in the counting procedure consisted of a lithium-drifted germanium detector in conjunction with a 4096 channel peak analyzer.

Upon request of the reactor staff, the bone samples were repackaged for the second irradiation in order to save reactor time. This procedure consisted of opening the vials, wrapping each sample in a polyethylene bag, and placing as many wrapped samples as possible in a vial. This did not involve the soil samples, for they were sealed in their own small vials that were not reopened. Before the six day count, the bone samples were unwrapped and replaced in their original vials. This process created three problems. First, it was impossible to know whether any contamination was introduced during the repackaging. Second, as many of the samples were quite friable, mass was lost in the transfer. Third, upon reweighing the samples, it was discovered that those samples that had remained completely intact throughout the repackaging process had increased in mass. This was undoubtedly due to the

absorption of atmospheric water. Rather than attempt to redry all the samples, which would again have involved some loss of material from the friable samples, it was decided to correct the masses of the bone samples using the completely intact samples as guidelines. Unfortunately, only four out of 21 bone samples could be used for this purpose, but their mass increase was quite consistent (mean = +19.4%, sd = 0.66%). Table 6, columns 2 and 3, present the masses after repackaging and after adjustment, respectively. These adjusted values were then used for quantification of elements observed during the six and 21 day counts of the second irradiation.

With the first irradiation, ten reference standards containing 1 mg each of Al and Mn were included. With the first and second irradiations, respectively, two and four bovine bone reference standards (International Atomic Energy Agency animal bone H-5 [Table 8]) were included. For bone analyses, this reference material is preferred over the usual mineralogical standards because compositional agreement between standard and sample leads to more precise results. Furthermore, to promote consistency in analysis and reporting of results, animal bone H-5 has been recommended by the most recent symposium on the chemical analysis of archaeological human bone (Price *et al.* 1989). As H-5 does not contain

Table 8. H-5 bovid bone reference material published elemental content. (International Atomic Energy Agency, Vienna)

Element	Unit[1]	Concentration	% error[2]	95% CI
Ba	mg/kg	79	16	67-92
Br	mg/kg	3.5	14	3.0-4.0
Ca	g/kg	212	3.8	204-220
Cl	mg/kg	550	18	450-650
Fe	mg/kg	79	7.5	73-85
K	mg/kg	680	17	560-800
Mg	g/kg	3.55	2.5	3.46-3.64
Na	g/kg	5.0	5.6	4.7-5.3
P	g/kg	102	8.4	93-110
Pb	mg/kg	3.1	18	2.6-3.7
Sr	mg/kg	96	8.6	88-105
Zn	mg/kg	89	5.9	84-95

1. Dry weight.
2. 95% confidence interval.

published amounts of Al and Mn, mineralogical standards had to be used to quantify these elements.

The obtained gamma-ray spectra were interpreted, in the case of the first count of the first irradiation, by a PC/MCA neutron activation analysis computer program, and in all other cases, by a SPAN analysis program. These programs correct for background and present the net area of energy peaks and the associated error. The programs were instructed to ignore all peaks with an error greater than 30% at one standard deviation. The identification of peak energies was also checked manually, as the programs can misinterpret energies that result from complex or abnormal peak configurations. In those cases where PC/MCA or SPAN were unable to calculate a peak area at all, the spectrum was examined manually, the peak area and background determined digitally, and the associated error calculated by the formula

$$\text{error} = \frac{100\%(A + 2B)^{0.5}}{A} ,$$

where A = peak area,
and B = background.

When an element of interest appeared as a peak on more than one activity measurement, the value with the lowest related error was used in subsequent calculations. This strategy resulted in the following association of

elements with irradiations and counting periods:

- first irradiation, first count: Al
- first irradiation, second count: Na, Mn
- second irradiation, first count: -
- second irradiation, second count: Ba, Ca, Fe, Sr, Zn

In the case of the second count of the first irradiation and both counts of the second irradiation, the differences in time elapsed between the end of bombardment and the start of the activity measurement for each sample in the series required that the peak areas be corrected for the differential decay. Such correction involves determining peak areas of the elements of interest at time zero, *i.e.*, at the end of bombardment (eob), according to the following procedure:

$$counts_{eob} = counts_{observed}(\exp[(\ln 2)(tc)/t_{0.5}]),$$

where ln2 = the natural logarithm of 2,
tc = centre time of the activity measurement,
and $t_{0.5}$ = half-life of element under consideration.

For the sake of convenience, the amount

$$\ln 2/t_{0.5}$$

is expressed as a constant, λ. Therefore,

$$counts_{eob} = counts_{observed}(\exp[\lambda tc]),$$

where $(\exp[\lambda tc])$ is termed the decay correction factor.

As radioactive isotopes decay according to an exponential curve, the centre time of the activity measurement cannot be determined by simply taking the point equidistant from the beginning and the end of the counting period. The centre time is calculated as follows:

$$\lambda tc = \frac{([\lambda tb + 1]\exp^{-\lambda tb}) - ([\lambda te + 1]\exp^{-\lambda te})}{(\exp^{-\lambda tb} - \exp^{-\lambda te})},$$

where tb = time elapsed between end of bombardment and beginning of activity measurement,
and te = time elapsed between end of bombardment and end of activity measurement.

Peak areas, peak area errors, decay correction factors, and corrected peak areas are presented in the Appendix. Iron is not included, for reasons explained below.

Quantitative analysis in INAA is based upon the following equation:

$$A_{sample}/A_{standard} = C_{sample}/C_{standard},$$

where A = activity (measured in corrected peak areas),
and C = concentration of parent nuclide.

In order to take into account the varying masses of the samples and the reference standards, this formula was amended as follows:

$$C_{sample} = (A_{sample}/y)/Mass_{sample},$$

where constant $y = A_{standard}/([Mass_{standard}][C_{standard}])$.

Constant y is a mean obtained from analysis of the ten mineralogical standards for Al and Mn, the two H-5 standards for Na, and the four H-5 standards for Ba, Ca, Sr, and Zn. The means obtained, with their standard deviations and coefficients of variation, are given in Table 9. As the coefficient of variation for iron was 22.7%, and some peak area errors were over 100%, it was decided to abandon analysis for this element. Better results can be obtained for iron when an irradiation period longer than 1 hour is used (G. Haverland, pers. comm.).

Resulting elemental concentrations in parts per million or per cent dry weight are given for Ba, Sr, and Zn in Table 10, for Ca and Na in Table 11, and for Al and Mn in Table 12. Associated imprecision data defining a 95% confidence interval were determined, in the case of Ba, Ca, Na, Sr, and Zn, by combining the errors presented for the animal bone standard H-5 (Table 8) with those calculated by SPAN (or manually, if SPAN was unable to interpret properly) according to the formula

$$error_{final} = ([error_1]^2 + [error_2]^2)^{0.5}.$$

SPAN errors, which are calculated at one standard

Table 9. Quantification constants (y) derived from reference standards.

Mineralogical	Al	Mn
n	10	10
mean (y)	2163.8	82.7
sd	64.8	3.0
CV	3.0%	3.6%

Animal bone (H-5)	Na
n	2
mean (y)	0.86
sd	0.02
CV	2.3%

Animal bone (H-5)	Ba	Ca	Sr	Zn
n	4	4	4	4
mean (y)	17.5	984.0	5.4	15.1
sd	2.3	120.8	0.4	1.4
CV	13.1%	12.3%	7.4%	9.3%

deviation, were doubled in order to make them compatible with the H-5 errors presented in Table 8. In the case of Al and Mn, the doubled coefficients of variation (Table 9) were combined in the same manner with the PC/MCA errors (calculated at two standard deviations) and the doubled SPAN errors, respectively.

The empty labelled control vial is not included in Tables 10 to 12, for the first and second counting periods of the first irradiation resulted in spectra that contained only normal background radioisotopes. Upon recommendation of the reactor staff, this vial was not included in the second irradiation.

Testing for pH was performed on the three soil samples. Soils from the Harappan excavations of 1988 have generally produced pH values of between 6.5 and 8.9, indicating that calcium carbonate is a dominant feature (Pendall 1989). Some values higher than this range are believed to be the result of the increased presence of sodium carbonate. The pH of the soil samples analyzed in this study was determined after mixing samples with distilled water with a known pH of 6.36. The values obtained were adjusted to compensate for the value of the water and are listed in Table 13.

Table 10. Concentrations of Ba, Sr, & Zn with errors[1] (ppm dry weight).

Sample		Ba	Sr	Zn
H87/40-34a	*Bos*	232 +/- 43	1708 +/- 167	173 +/- 16
H87/60-46a	*Bos*	232 +/- 81	2033 +/- 210	189 +/- 21
	Canis	191 +/- 58	1806 +/- 178	134 +/- 14
H87/60-46a-26	E[2]	118 +/- 72	1154 +/- 156	107 +/- 19
	C[3]	95 +/- 71	1378 +/- 170	124 +/- 20
	P[4]	56 +/- 67	1241 +/- 146	139 +/- 18
H87/60-46a-77	E	65 +/- 55	996 +/- 123	97 +/- 15
	C	133 +/- 100	1561 +/- 200	146 +/- 24
	P	103 +/- 58	1122 +/- 131	128 +/- 17
H87/85-49d.2	C	709 +/- 159	1981 +/- 245	238 +/- 33
	Bos	250 +/- 59	831 +/- 93	103 +/- 11
	soil	574 +/- 132	209 +/- 97	114 +/- 17
H87/136-147a	E	1475 +/- 273	1720 +/- 217	171 +/- 27
	C	1694 +/- 295	1921 +/- 209	198 +/- 24
	P	1878 +/- 316	1984 +/- 202	207 +/- 21
	soil	630 +/- 131	320 +/- 92	113 +/- 17
H87/137-148a	E	621 +/- 122	2319 +/- 232	107 +/- 15
	C	444 +/- 107	2089 +/- 229	98 +/- 17
	P	389 +/- 82	2062 +/- 203	119 +/- 14
	soil	436 +/- 138	185 +/- 144	125 +/- 24
H87/197-194a	E	590 +/- 136	1950 +/- 230	120 +/- 21
	C	521 +/- 144	1995 +/- 258	115 +/- 25
	P	694 +/- 149	1940 +/- 222	129 +/- 21
H87/197-194b		655 +/- 131	2926 +/- 287	190 +/- 22
Saw Blade		0	0	0

1. 95% confidence interval.
2. Endosteal portion of femoral cross-section.
3. Centre portion of femoral cross-section.
4. Periosteal portion of femoral cross-section.

Table 11. Concentrations of Ca (% dry weight) & Na (ppm dry weight) with errors[1].

Sample		Ca	Na
H87/40-34a	*Bos*	46.2 +/- 4.7	31660 +/- 1907
H87/60-46a	*Bos*	49.4 +/- 6.6	21360 +/- 1420
	Canis	40.2 +/- 4.9	25065 +/- 1560
H87/60-46a-26	E[2]	38.9 +/- 6.4	23471 +/- 1695
	C[3]	36.9 +/- 6.0	35200 +/- 2485
	P[4]	45.9 +/- 5.8	25820 +/- 1708
H8	77 E	29.0 +/- 4.6	20954 +/- 1440
	C	42.6 +/- 7.6	20148 +/- 1562
	P	35.3 +/- 5.0	21900 +/- 1456
H87/85-49d.2	C	41.9 +/- 8.3	30361 +/- 2111
	Bos	40.6 +/- 4.3	34078 +/- 2041
	soil	9.8 +/- 3.5	15481 +/- 1039
H87/136-147a	E	43.5 +/- 8.6	11950 +/- 1062
	C	50.0 +/- 7.3	14584 +/- 1111
	P	47.3 +/- 6.0	13515 +/- 946
	soil	12.9 +/- 3.7	13085 +/- 891
H87/137-148a	E	44.5 +/- 5.8	11761 +/- 872
	C	45.3 +/- 7.4	12630 +/- 1026
	P	37.4 +/- 4.7	10481 +/- 758
	soil	11.2 +/- 4.9	13637 +/- 1096
H87/197-194a	E	43.8 +/- 8.3	38776 +/- 2594
	C	45.1 +/- 10.2	35697 +/- 2529
	P	48.9 +/- 8.4	37428 +/- 2484
H87/197-194b		40.9 +/- 6.1	49577 +/- 3009
Saw Blade		0	0

1. 95% confidence interval.
2. Endosteal portion of femoral cross-section.
3. Centre portion of femoral cross-section.
4. Periosteal portion of femoral cross-section.

Table 12. Concentrations of Al & Mn with errors[1] (ppm dry weight).

Sample		Al		Mn	
H87/40-34a	*Bos*	239 +/-	37	6 +/-	3
H87/60-46a	*Bos*	456 +/-	65	24 +/-	5
	Canis	435 +/-	56	13 +/-	4
H87/60-46a-26	E[2]	423 +/-	81	46 +/-	8
	C[3]	272 +/-	62	21 +/-	6
	P[4]	388 +/-	92	47 +/-	7
H87/60-46a-77	E	461 +/-	72	65 +/-	9
	C	261 +/-	68	18 +/-	6
	P	455 +/-	68	106 +/-	12
H87/85-49d.2	C	436 +/-	84	41 +/-	9
	Bos	233 +/-	38	13 +/-	4
	soil	49788 +/-	3046	766 +/-	60
H87/136-147a	E	189 +/-	69	14 +/-	5
	C	269 +/-	57	9 +/-	4
	P	189 +/-	48	39 +/-	6
	soil	50337 +/-	3080	662 +/-	52
H87/137-148a	E	419 +/-	63	18 +/-	4
	C	240 +/-	56	18 +/-	5
	P	264 +/-	43	33 +/-	5
	soil	61363 +/-	3795	802 +/-	67
H87/197-194a	E	467 +/-	87	34 +/-	10
	C	294 +/-	76	0	
	P	444 +/-	81	69 +/-	11
H87/197-194b		3985 +/-	299	114 +/-	13
Saw Blade		441 +/-	139	8498 +/-	648

1. 95% confidence interval.
2. Endosteal portion of femoral cross-section.
3. Centre portion of femoral cross-section.
4. Periosteal portion of femoral cross-section.

Table 13. Soil sample pH.

Sample	H87/85-49d.2	H87/136-147a	H87/137-148a
pH	8.5	7.8	9.7

5. DISCUSSION

This discussion begins with a consideration of the soil data and proceeds with an examination of the elemental content of the human and animal bone samples.

Tables 10 to 12 reveal that all elements of interest are present in greater than trace quantities in all three soil samples analyzed. Al, Ca, and Na, with concentrations of over 1%, form substantial constituents. Taking into account the associated imprecision data, homogeneity is shown between samples in the case of Ca, while the data for Al and Na indicate a somewhat wider range of variation. This may reflect greater intrasite diversity for these two elements than for Ca.

Given the high pH of these soils, the large absolute amounts of Ca and Na are not surprising. These data are basically in agreement with the observations of Pendall (1989), who found that Ca and Na, bound as carbonates, were dominant in alkaline and very alkaline Harappan soils, respectively. Soil sample H87/137-148a, with a pH of 9.7, may have been expected to contain more Na than H87/85-49d.2 and H87/136-147a, if indeed the element is present as a carbonate. This, however, is an assumption not tested here. Moreover, the samples analyzed by INAA do not contain exactly the same soil as the material submitted for pH testing. Intrasample homogeneity of

elemental content and pH for soil retrieved from the excavation, for the portion taken from it for this study, and for the separate subsamples used in INAA and pH testing has also been assumed, but is not necessarily the case.

The diet-indicating elements, Ba, Sr, and Zn, are present in concentrations of between 0.01 and 0.1% in all three soil samples. The quantity of Mn also falls into this range. Imprecision ranges show homogeneity between samples for Ba, Sr, and Zn and nearly so for Mn. Again, sampling limitations have their effect on the final results. Of major importance, in particular for the known bone-seeking indicators of diet, is the fact that presence in greater than trace quantities, *i.e.*, in substantial amounts, has been demonstrated.

Only three human femoral samples (H87/85-49d.2, H87/136-147a, and H87/137-148a) are associated with soil in this study, and of these, only two allowed a gradient analysis. Values for Ba and Sr undoubtedly point to postmortem enrichment in all three, for these values are larger than presumed physiological levels by more than a factor of ten. Even when the considerable limitations of Table 3 are taken into consideration, no other alternative is remotely possible. Probably both simple contamination as well as ionic adsorption and

substitution have resulted in these high values, for Sr and very likely Ba, both IIa elements, participate in exchange processes on the surface and in the interior of the hydroxyapatite crystal. More difficult to explain is the intersample variability for these elements. In the case of Ba, concentrations in H87/136-147a are more than twice as large as in the other two bone samples, yet the soil values vary little. Gradients of concentration present in H87/136-147a and H87/137-148a run in opposite directions, with a decrease from the periosteal to the endosteal surfaces in the former and an increase in the latter. This difference may be due to the presence of soil in the medullary cavity of one and not the other. However, the large intersample variation in concentrations is not explained by reference to the gradient question. As the differences are far too large to be attributable to variation in antemortem levels, it is possible that H87/136-147a was exposed to higher levels of exchangeable soil Ba, perhaps due to greater flow of groundwater. Areas of cemetery R37 have probably experienced variable exposure to water flow as a result of irrigation practices (N. Lovell, pers. comm.) The bone-seeking nature of Ba is definitely shown by all three bone samples.

Variation between the three bone samples associated

with soil is less in the case of Sr. When imprecision data are take into consideration, intersample homogeneity is approached. Gradient directions for Sr are in agreement with those for Ba, lending support to the possibility that medullary cavity soil played a greater role in H87/137-148a than H87/136-147a. However, the similar if not larger overall amount of Sr in the former compared to the latter at first glance does not agree with the theory that H87/136-147a was exposed to more exchangeable ions in solution. An answer may lie in the possibility that differences in groundwater were indeed present, but that Sr is a more active bone seeker than Ba and does not rely as heavily on water as a transport medium. Although the oxidation state of both is +2, the atomic radius of Sr (2.45 A) is closer to that of Ca (1.99 A) than is Ba (2.78 A) (Sargent-Welch 1980). Thus, Sr may be more prone to deposition in the crystal structure than Ba.

The data for Zn provide less obvious indicators of postmortem enrichment than Ba and Sr. Some values for the bone samples associated with the Zn-bearing soil are within the possible physiological range. It is precisely in such a case that soil data can reveal their worth. While in the case of Ba and Sr postmortem contamination is obvious even in the lack of such data, one might

assume that Zn has not experienced similar alteration. Knowledge of the presence of soil Zn, however, forces one to take into consideration the likely possibility of contamination. An examination of the gradients is less informative, for only the absolute values themselves reveal patterns that are more likely in keeping with postmortem change than with physiological factors, namely either a steady decrease from the periosteal to the endosteal surface in H87/136-147a (in agreement with the pattern of Ba and Sr) or a lowest value in the more inaccessible centre portion of H87/137-148a. The imprecision data associated with these absolute values, however, could contradict any such assumed patterns. The overall relatively low values for Zn as compared with Ba and Sr lend support to the theory that this element is not a bone seeker. The fact that bone Zn levels are higher than those for soil in the case of H87/85-49d.2 and H87/136-147a may point to the possibility of identifiable antemortem variation. However, as the actual mechanisms of change in each bone sample are unknown, such differences in level would be an unstable basis upon which to build a theory of variation in physiological Zn and consequently diet.

The bone samples for which associated soil was not available must, for the purposes of this study, be

assumed to have been interred in a context similar to the soil analyzed here. However, the levels of Ba present in bone samples H87/60-46a-26 and H87/60-46a-77 may point to less available Ba in the vicinity of feature 46a, a possibility that would be in agreement with the variable intrasite Ba availability hypothesized above. Upon first glance, the agreement between some of these low values and the physiological range could indicate lack of postmortem alteration, but the knowledge that Ba exists elsewhere in the site should be sufficient to discount this idea if one wishes to remain conservative. No such ambiguity exists for H87/197-194a and 194b, the Ba content of which is in line with H87/137-148a. In the case of all four human bone samples unassociated with soil, the large error values render the usefulness of any gradient study doubtful.

The amount of Sr in H87/60-46a-26 and 77 and in H87/197-194a and 194b is, like in the other human bone samples discussed above, undoubtedly due to postmortem deposition. No knowledge of soil composition is required in order to come to this conclusion. The much higher level in the neonatal tibia (H87/197-194b) as compared to the adult from the same burial supports the findings elsewhere that the porosity of infant bone renders it even more susceptible to chemical alteration (Gordon and

Buikstra 1981). While the gradients of H87/60-46a-26 and H87/197-194a indicate homogeneity (taking imprecision data into account), the centre portion of H87/60-46a-77, from the same context as 26, contains more Sr than the periosteal and endosteal portions. There is no obvious explanation for this apparent anomaly.

The data for Zn, again, are less easy to interpret in the case of these human bone samples unassociated with soil. As in the case of Ba, knowledge that Zn exists in the site should be sufficient to eliminate the Zn content of these bone samples as a potential source for dietary reconstruction, if one wishes to proceed with care. The higher value for the neonatal tibia may support the theory of postmortem absorption for all these samples, but as total-body concentrations of Zn have been found to be high in fetuses near or at term (Hambidge *et al.* 1986), this is not conclusive. Gradients are again nonexistent except in H87/60-46a-77, the high centre value of which probably stems from the same unknown source as its Sr value.

Intersample variation of Na, believed not to interact with the hydroxyapatite crystal interior but only adsorb onto its surface (Table 2), appears to be more difficult to account for than that of Ba, Sr, or Zn. Examination of the data for H87/136-147a and H87/137-148a

may lead one initially to suggest that the bone samples have become impregnated with soil Na and that, thus, their values for this element have been raised from the physiological level of approximately 10,000 ppm (Table 3) to a level between this amount and the soil level of roughly 12,000 to 15,000 ppm (imprecision data included). However, an apparently anomalous situation is then present in the case of H87/85-49d.2, which reveals a bone Na level of approximately twice that of the soil. Bringing the human bone samples not associated with soil into view, one finds a range of values from intrasample homogeneity at a high level (H87/197-194a), to intrasample homogeneity at an intermediate level (H87/60-46a-77), to lack of intrasample homogeneity covering a span from intermediate to high (H87/60-46a-26). The extremely high Na level of the neonatal tibia as compared to the associated adult femur may point to preferential absorption of Na into more porous bone. This theory, however, helps little when attempting to account for the overall variation in the adult bones from 10,000 to 15,000 ppm on the one extreme and 35,000 to 40,000 ppm on the other, unless variable bone porosity has indeed been present and has played a role. Reference to the distribution patterns of the other elements discussed does not elucidate the matter. This variability in Na

content can tentatively only be explained as differential proneness of this bone material to contamination by this element, perhaps related to bone porosity and readily available crystal surface and space. The identical atomic radii of Na and Ca may indicate that the two are quite interchangeable.

Increase in overall Ca content with little intra- or intersample variation is apparent in all bone samples. The high but homogeneous distribution of Ca between soil samples, which may be extrapolated to the soil elsewhere in the site, is probably reflected in this lack of variability in enrichment. The high Ca level in the bone samples resulting from impregnation with and substitution by calcium carbonate presumed to be present in the alkaline soil may also have contributed to the magnitude of enrichment involving Ba, Sr, and possibly Na in some cases.

The usefulness of Al as an indicator of postmortem compositional change is supported by this study. It is present in all human bone samples in amounts far higher than physiological levels or than could have arisen from Al in the saw blade, demonstrating extensive postmortem alteration of chemical composition in the form of contamination and possibly Ca substitution. An examination of gradients of change is not revealing, for

no clear pattern is apparent. A comparison of these gradients with those of other elements also does not disclose any agreement. The neonatal tibia, with approximately ten times the Al content of the adult bone associated with it, again portrays the susceptibility of porous, immature bone to postmortem compositional alteration, in this case largely attributable to simple filling of spaces.

Like Zn, Mn is present in all the samples at a level within a possible antemortem range. Any postmortem contamination has remained moderate, in keeping with the restricted activity of this element as a relatively insoluble oxide in alkaline soils. The concentration of Mn in the neonatal tibia is not sufficiently high to indicate firmly a postmortem external source, but an examination of the gradients is more informative. Mn is the only element in this study that almost always occurs at the highest levels in the periosteal portion, at lower levels in the endosteal portion, and at the lowest levels in the centre portion. This is a reasonable pattern when one assumes that most contamination should occur at the periosteal margin, less from any soil in the medullary cavity, and the least in the relatively inaccessible centre section. In this particular case, the high Mn level of the saw blade may have resulted in raised Mn

values for all the bone samples, but presumably by a relatively equal amount. Thus, the gradient study has shown that Mn, too, has been subject to postmortem enrichment.

The analysis of faunal remains in this study proved to be of limited value due to the availability of only one sample in association with soil. In the case of Ba, Sr, Zn, Al, and Mn, this single *Bos* sample appeared to be less subject to contamination than the human bone sample H87/85-49d.2 from the same context. Without a knowledge of bovine physiology, however, a frame of reference is lacking. If the composition of the bovine reference standard H-5 (Table 8) is any indication, the levels of Ba, Sr, Ca, and Na have, like the associated human sample, been considerably augmented. The *Bos* sample H87/40-34a, not in association with soil or human material, has been similarly altered, with only its Sr content considerably higher.

A comparison of the *Bos* and *Canis* samples from feature H87/60-46a with the associated human bone again does not add any extra information to this analysis. Both may be even more contaminated than the human samples with respect to Sr and possibly Ba. In the case of Ba, this may be related to the hypothesized variable intrasite availability of this element. In the case of

Sr, the more porous nature of the chiefly trabecular faunal bone samples, including H87/40-34a, may have rendered them more prone to contamination and ionic exchange with this element. This hypothesis is in agreement with the Sr data for the neonatal tibia H87/197-194b. Zn levels could at first glance be of physiological origin, but unless bovine and canine metabolisms result in differential deposition of this element in bone, these levels would indicate that the bovid ate more meat than the canid. Thus, patternless postmortem alteration again appears to be the case.

In summary, all seven of the elements of interest quantified here have experienced transport from soil to bone. The values for Ba and Sr provide sound evidence for the postmortem bone-seeking behaviour of these two elements. Zn does not appear to possess this characteristic, but the knowledge that Zn is present in the soil, and, as a cation, could interact with Ca, is sufficient to classify it as postmortally altered. A gradient study shows that Mn, too, has entered the bone from the soil. The large amounts of Al, Ca, and Na present in the soil have also undoubtedly influenced the chemical composition of the bone, but in the case of Na this has not been clear in every subsample. Because the contamination of the human bone samples is quite obvious,

analysis of faunal remains has not added information to this study. It appears very likely, however, that the animal bone has been influenced by the same factors as the human bone and has undergone postmortem enrichment of all the elements of interest.

6. CONCLUSION

The goal of this study was to evaluate the suitability of the elemental content of bone mineral for reconstruction of Harappan palaeodiet using information obtained from instrumental analysis of bone samples and associated soil and faunal remains. It was revealed that, in the case of all elements of interest (Ba, Sr, Zn, Na, Ca, Al, and Mn), enrichment had occurred after interment. This substantial extrinsic, postmortem alteration effectively obliterated any dietary signature that may have been present in the bone mineral and obviated any consideration of the effect of intrinsic, antemortem factors on the results obtained. Several points were raised in the analysis and discussion that may warrant attention in future analyses of this nature. These points are considered in the following paragraphs.

In order to make meaningful interpretations of trace element data obtained from human and faunal archaeological bone samples, a frame of reference in the form of a possible antemortem range of values must be present. In this study, information in Table 3 served this purpose for the human material. However, such a compilation may be of limited worth. Elemental content can vary considerably in living bone and, moreover, there is no guarantee that the values in such a table can be

applied to prehistoric populations. The amount of strontium, one of the major diet-indicating elements, has risen appreciably in the global trophic system over the last few decades. Physiological continuity is generally assumed in palaeobiological studies, but the possibility of aspects that may vary diachronically, for example the level of bone strontium, should be kept in mind.

Another factor may render a compilation of concentrations of elements for modern bone less applicable to archaeological remains. Interred bone may experience differential decomposition of its protein and mineral fractions. Modern concentration values in ppm or mg/kg are based on intact bone. If, for example, very severe protein degradation has occurred, elements such as carbon, nitrogen, and oxygen, which are major ingredients of fresh bone (Table 3), may be eliminated to a great extent. This would raise the expected antemortem values for the elements chiefly or wholly associated with the mineral fraction. In such a case, modern values for ashed bone would, if available, be preferred. Unfortunately, the actual degree of variable degradation between the protein and mineral parts of archaeological bone may be impossible to ascertain.

In situations where postmortem contamination is severe, as in this study, the problems associated with

assumed diachronic physiological continuity and differential protein/mineral alteration would not be of concern. However, when values obtained from archaeological bone are found to be within or close to a possible antemortem range, difficulties in interpretation may arise. In such instances, soil data are indispensable in order to determine whether or not a potential contaminant is present.

When animal bones of known species with known diet are being treated as possible indices of postmortem alteration, a suitable frame of reference of antemortem values for trace elements must also be known. It is generally assumed that the behaviour of elements of interest in, for example, bovid or canid physiology is similar to that of humans. In the case of bovids, the elemental content of the H-5 reference material (Table 8) suggests that differences indeed exist. When comparing a herbivore to a human, one would expect higher strontium and lower zinc values in the former than in the latter, unless the human in question were completely vegetarian. However, the ranges for these two elements in Table 8 fall completely within those of humans in Table 3, indicating an incorrect assumption at some point. The use of faunal comparisons in palaeodiet reconstruction definitely requires further study.

Looking specifically at the techniques used in this analysis, it may be stated that, when postmortem alteration is severe, gradient analysis and the use of contamination indicators like aluminum and manganese are normally superfluous. However, in more ambiguous cases where alteration has not been great and the obtained values are close to assumed physiological levels, these two methods may provide assistance in evaluating the usefulness of a bone sample, whether or not soil data are also available. Regarding the gradient analysis, it would also be advisable to determine the presence, by simple gross examination, of any medullary cavity soil before the sample is subjected to further handling. This would facilitate the interpretation of any gradient.

The technique of INAA proved to be sufficient for the needs of this study. It can only be suggested that irradiation and counting strategies be selected precisely for the elements in question in order to reduce the associated error as much as possible. Furthermore, samples should be handled as little as possible after initial encapsulation. In instances where postmortem alteration has been severe, the slight errors introduced by repackaging, as in this study, would have a negligible effect on the final conclusions. When values are closer to physiological levels and a dietary signature may be

present, however, accuracy becomes more important and the method must be as rigourous as possible.

The analysis of a neonatal bone from a skeleton that was in contact with an adult provided useful information regarding bone porosity and susceptibility to postmortem alteration. In the case of strontium, sodium, aluminum, and manganese, concentrations were found to be far higher in the porous neonatal tibia than in the associated adult femoral compact bone. Aluminum and manganese, which are present in much greater amounts in the soil than in the bone, may merely be present in the soil that has presumably filled the numerous pores of this immature sample. Strontium and sodium, however, appear to have been more actively attracted to the porous than to the compact bone. The crystalline surface available for ionic adsorption and substitution in the Haversian systems of compact bone is indeed enormous, but the more readily accessible trabeculae of the porous bone of the neonatal tibia may result in greater exchange capability. Moreover, mature bone is more highly crystallized than immature bone, and ionic exchange may not occur as easily in the former as in the less organized structure of hydroxyapatite in the latter. Further research needs to be conducted into the mechanisms behind variation in susceptibility to postmortem chemical alteration of

compact versus trabecular and mature versus immature bone.

In conclusion, a combination of data obtained from elemental analysis of human bone and associated soil has provided evidence that any dietary signature formerly present in these bone samples has been destroyed. Examination of gradients of contamination and analysis of associated animal bones contribute secondary support in some cases. Thus, reconstruction of palaeodiet of individuals from this Harappan cemetery must proceed by other means. In future trace element studies, it is recommended that bone and associated soil be analyzed whenever possible so that the possibility of postmortem alteration can be rigourously addressed. Where soil samples are not available, the initial priority of the researcher must still be to seek any change resulting from interment before attempting to identify any dietary signature.

BIBLIOGRAPHY

Agrawal, D.P.
1982 The Indian Bronze Age Cultures and Their Metal Technology. *Advances in World Archaeology* 1: 213-264.

Allchin, Bridget and Raymond Allchin
1982 *The Rise of Civilization in India and Pakistan.* Cambridge University Press, Cambridge.

anonymous
1974 Preface. In *Geochemistry and the Environment* (vol. 1), pp. v-vi. National Academy of Sciences, Washington, D.C.

Armelagos, George J., Barrett Brenton, Michael Alcorn, Debra Martin, and Dennis P. Vangerven
1989 Factors affecting elemental and isotopic variation in prehistoric human skeletons. In *The Chemistry of Prehistoric Human Bone*, edited by T. Douglas Price, pp. 230-244. Cambridge University Press, Cambridge.

Ascenzi, Antonio
1986 Microscopy and ultramicroscopy in palaeopathology. In *Innovative Trends in der prähistorischen Anthropologie*, edited by Bernd Herrmann, pp. 149-156. (*Mitteilungen der Berliner Gesellschaft für Anthropologie, Ethnologie und Urgeschichte* 7).

Aufderheide, Arthur C.
1989 Chemical Analysis of Skeletal Remains. In *Reconstruction of Life From the Skeleton*, edited by Mehmet Yasar Iscan and Kenneth A.R. Kennedy, pp. 237-260. Alan R. Liss, Inc., New York.

Aufderheide, Arthur C., J. Lawrence Angel, Jennifer O. Kelley, Alain C. Outlaw, Merry A. Outlaw, George Rapp Jr., and Lorentz E. Wittmers Jr.
1985 Lead in Bone III. Prediction of Social Correlates From Skeletal Lead Content in Four Colonial American Populations (Catoctin Furnace, College Landing, Governor's Land, and Irene Mound). *American Journal of Physical Anthropology* 66: 353-361.

Aufderheide, Arthur C., Lorentz E. Wittmers, George Rapp, and J. Wallgren
1988 Anthropological applications of skeletal lead analysis. *American Anthropologist* 90: 932-936.

Barnhisel, Richard and Paul M. Bertsch
1982 Aluminum. In *Methods of Soil Analysis* (part 2), edited by A.L. Page, R.H. Miller, and D.R. Keeney, pp. 275-300. American Society of Agronomy, Inc., and Soil Science Society of America, Inc., Madison, Wisconson.

Baud, Charles-Albert and Danielle Lacotte
1984 Etude au microscope électronique à transmission de la colonisation bactérienne de l'os mort. *Comptes Rendus de l'Academie des Sciences Ser. 2* 298: 507-510.

Bell, Lynne S.
1990 Palaeopathology and Diagenesis: An SEM Evaluation of Structural Changes Using Backscattered Electron Imaging. *Journal of Archaeological Science* 17: 85-102.

Bethell, P.H. and J.U. Smith
1989 Trace-element Analysis of an Inhumation from Sutton Hoo, Using Inductively Coupled Plasma Emission Spectrometry: An Evaluation of the Technique Applied to Analysis of Organic Residues. *Journal of Archaeological Science* 16: 47-55.

Blakely, Robert L.
1989 Bone Strontium in Pregnant and Lactating Females From Archaeological Samples. *American Journal of Physical Anthropology* 80: 173-185.

Bonar, L.C., M.D. Grynpas, J.E. Roberts, R.G. Griffin, and M.J. Glimcher
1985 Physical and Chemical Characterization of the Development and Maturation of Bone Mineral. In *The Chemistry and Biology of Mineralized Tissues*, edited by William T. Butler, pp. 226-233. William T. Butler, Birmingham, Alabama.

Bowen, H.J.M.
1979 *Environmental Chemistry of the Elements.* Academic Press, London.

Brätter, P., D. Gawlik, J. Lausch, and U. Rösick
1977 On the Distribution of Trace Elements in Human Skeletons. *Journal of Radioanalytical Chemistry* 37: 393-403.

Brätter, P., H. Gessner, B. Herrmann, J. Lausch, and U. Rösick
1980 Zur Identifizierung menschlicher Knochen durch neutronenaktivierungsanalytisch bestimmte Spurenelementverteilungsmuster. *Berliner Beiträge zur Archäometrie* 5: 187-195.

Browne, Edgardo and Richard B. Firestone
1986 *Table of Radioactive Elements.* John Wiley and Sons, New York.

Bruchhaus, Horst
1987 Zur Problematik der Verwendung histologischer Methoden an menschlichen Skelettresten für die Sterbealterbestimmung. *Alt-Thüringen* 22/23: 35-54.

Buikstra, Jane E., Susan Frankenberg, Joseph B. Lambert, and Liang Xue
1989 Multiple elements: multiple expectations. In *The Chemistry of Prehistoric Human Bone*, edited by T. Douglas Price, pp. 155-210. Cambridge University Press, Cambridge.

Byrne, Kevin B. and David C. Parris
1987 Reconstruction of the Diet of the Middle Woodland Amerindian Population at Abbott Farm by Bone Trace-Element Analysis. *American Journal of Physical Anthropology* 74: 373-384.

Curzon, M.E.J.
1983 Epidemiology of Trace Elements and Dental Tissues. In *Trace Elements and Dental Disease*, edited by M.E.J. Curzon and T.W. Cutress, pp. 11-30. John Wright and Sons Ltd., Bristol.

Dales, George F.
1989 Harappa: A New Look at the Type Site of the Indus Civilization. In *Old Problems and New Perspectives in the Archaeology of South Asia*, edited by Jonathan Mark Kenoyer, pp. 127-131. (*Wisconsin Archaeological Reports* 2).

Dales, George F. and J. Mark Kenoyer
1988 *Preliminary Report on the Third Season (January - March 1988) of Research at Harappa, Pakistan.* Unpublished manuscript.

Dallemagne, Marcel J. and Leon J. Richelle
1973 Inorganic Chemistry of Bone. In *Biological Mineralization*, edited by Isadore Zipkin, pp. 23-42. John Wiley & Sons, New York.

Davies, David R., James B. Bassingthwaighte, and Patrick J. Kelly
1979 Blood Flow and Ion Exchange in Bone. In *Skeletal Research: An Experimental Approach*, edited by David J. Simmons and Arthur S. Kunin, pp. 397-419. Academic Press, New York.

Dehos, R. and H. Schmier
1985 Strontium-90 Content in Human Bone in West German Residents. In *Metals in Bone*, edited by Nicholas D. Priest, pp. 87-95. MTP Press Ltd., Lancaster, England.

Dennison, K.J.
1980 Amino Acids in Archaeological Bone. *Journal of Archaeological Science* 7: 81-86.

di Fiore, Mariano S.H.
1989 *Atlas of Normal Histology.* Lea and Fibeger, Philadelphia.

Eanes, E.D. and A.S. Posner
1970 Structure and Chemistry of Bone Mineral. In *Biological Calcification: Cellular and Molecular Aspects*, edited by Harald Schraer, pp. 1-26. Meredith Corporation, New York.

Edward, Jeremy, John M. Fossey, and Leo Yaffe
1984 Analysis by Neutron Activation of Human Bone from the Hellenistic Cemetery at Asine, Greece. *Journal of Field Archaeology* 11: 37-46.

El-Kammar, A., R.G.V. Hancock, and Ralph O. Allen
1989 Human Bones as Archaeological Samples. In *Archaeological Chemistry IV*, edited by Ralph O. Allen, pp. 337-352. American Chemical Society, Washington, D.C.

Engstrom, Arne
1972 Aspects of the Molecular Structure of Bone. In *The Biochemistry and Physiology of Bone* (vol. 1), edited by Geoffrey H. Bourne, pp. 237-257. Academic Press, New York.

Francalacci, Paolo
1989 Dietary Reconstruction at Arene Candide Cave (Liguria, Italy) by Means of Trace Element Analysis. *Journal of Archaeological Science* 16: 109-124.

Gambrell, Robert P. and William H. Patrick Jr.
1982 Manganese. In *Methods of Soil Analysis* (part 2), edited by A.L. Page, R.H. Miller, and D.R. Keeney, pp. 313-322. American Society of Agronomy, Inc., and Soil Science Society of America, Inc., Madison, Wisconsin.

Glimcher, Melvin J.
1981 On the Form and Function of Bone: From Molecules to Organs. In *The Chemistry and Biology of Mineralized Connective Tissue*, edited by Arthur Veis, pp. 617-673. Elsevier North Holland, New York.

Gordon, Claire G. and Jane E. Buikstra
1981 Soil pH, Bone Preservation, and Sampling Bias at Mortuary Sites. *American Antiquity* 46: 566-571.

Grupe, Gisela
1986a Rekonstruktion bevölkerungsbiologischer Parameter aus dem Elementgehalt bodengelagerter Knochen. In *Innovative Trends in der prähistorischen Anthropologie*, edited by Bernd Herrmann, pp. 39-44. (*Mitteilungen der Berliner Gesellschaft für Anthropologie, Ethnologie und Urgeschichte* 7)

1986b *Multielementanalyse: Ein neuer Weg für die Paläodemographie*. Bundesinstitut für Bevölkerungsforschung, Wiesbaden. (*Materialien zur Bevölkerungswissenschaft Sonderheft* 7)

1987 Spurenelemente in bodengelagerten menschlichen Knochen und ihre Aussagen. Ein Überblick. *Anthropologischer Anzeiger* 45: 19-28.

1988a Impact of the Choice of Bone Samples on Trace Element Data in Excavated Human Skeletons. *Journal of Archaeological Science* 15: 123-129.

1988b Metastasizing Carcinoma in a Medieval Skeleton: Differential Diagnosis and Etiology. *American Journal of Physical Anthropology* 75: 369-374.

Grupe, Gisela and Hermann Piepenbrink
1988 Trace element contaminations in excavated bones by microorganisms. In *Trace Elements in Environmental History*, edited by Gisela Grupe and Bernd Herrmann, pp. 103-112. Springer Verlag, Berlin.

Grynpas, Marc D., Laurence C. Bonar, and Melvin J. Glimcher
1981 On the Question of Amorphous Calcium Phosphate in Bone Mineral. In *The Chemistry and Biology of Mineralized Connective Tissue*, edited by Arthur Veis, pp. 279-283. Elsevier North Holland, New York.

Grynpas, M.D., K.P.H. Pritzker, and R.G.V. Hancock
1987 Neutron Activation Analysis of Bulk and Selected Trace Elements in Bones Using a Low Flux SLOWPOKE Reactor. *Biological Trace Element Research* 13: 333-344.

Hackett, C.J.
1981 Microscopical focal destruction (tunnels) in exhumed human bones. *Medicine, Science, and the Law* 21: 243-265.

Hambidge, K. Michael, Clare E. Casey, and Nancy F. Krebs
1986 Zinc. In *Trace Elements in Human and Animal Nutrition - Fifth Edition* (vol. 2), edited by Walter Mertz, pp. 1-137. Academic Press, Inc., Orlando.

Hancock, R.G.V., M.D. Grynpas, and K.P.H. Pritzker
1989 The Abuse of Bone Analyses for Archaeological Dietary Studies. *Archaeometry* 31: 169-179.

Hancox, N.M.
1972 *Biology of Bone*. Cambridge University Press, Cambridge.

Hanson, Douglas B. and Jane E. Buikstra
1987 Histomorphological Alteration in Buried Human Bone from the Lower Illinois Valley: Implications for Palaeodietary Research. *Journal of Archaeological Science* 14: 549-563.

Hassan, Afifa A., John D. Termine,
and C. Vance Haynes, Jr.
1977 Mineralogical Studies on Bone Apatite and Their Implications for Radiocarbon Dating. *Radiocarbon* 19: 364-374.

Henderson, Julian, R.C. Janaway, and J.R. Richards
1987 Cremation slag: a substance found in funerary urns. In *Death, Decay and Reconstruction*, edited by A. Boddington, A.N. Garland, and R.C. Janaway, pp. 81-100. Manchester University Press, Manchester.

Henderson, P., C.A. Marlow, T.I. Molleson,
and C.T. Williams
1983 Patterns of Chemical Change during Bone Fossilization. *Nature* 306: 358-360.

Herrmann, B.
1981 Eine Möglichkeit der makroskopischen Fehlbeurteilung von Dekompositionserscheinungen des Knochens. *Zeitschrift für Rechtsmedizin* 87: 275-278.

Herrmann, B. and H. Newesely
1982 Dekompositionsvorgänge des Knochens unter langer Liegezeit. *Anthropologischer Anzeiger* 40: 19-31.

Hisanaga, Akira, Yukuo Eguchi, Miyuki Hirata,
and Noburu Ishinishi
1988 Lead Levels in Ancient and Contemporary Japanese Bones. *Biological Trace Element Research* 16: 77-85.

Howell, David S.
1971 Current Concepts of Calcification. *Journal of Bone and Joint Surgery* 53A: 250-258.

Jansen, M.
1989 Water supply and sewage disposal at Mohenjo-Daro. *World Archaeology* 21: 177-192.

Jaworowski, Z., F. Barbalat, C. Blain, and E. Peyre
1985 Historical Changes of Trace Metals in Human Bones from France. In *Metals in Bone*, edited by Nicholas D. Priest, pp. 383-393. MTP Press Ltd., Lancaster, England.

Katzenberg, Mary Anne
1984 *Chemical Analysis of Prehistoric Human Bone from Five Temporally Distinct Populations in Southern Ontario.* National Museum of Man Mercury Series, Archaeological Survey of Canada Paper 129.

Keith, Margaret S. and George J. Armelagos
1988 An Example of *In Vivo* Tetracycline Labelling: Reply to Piepenbrink. *Journal of Archaeological Science* 15: 595-601.

Klepinger, Linda L.
1984 Nutritional assessment from bone. *Annual Review of Anthropology* 13: 75-96.

Klepinger, Linda L., John K. Kuhn, and Wendell S. Williams
1986 An Elemental Analysis of Archaeological Bone From Sicily as a Test of Predictability of Diagenetic Change. *American Journal of Physical Anthropology* 70: 325-331.

Lambert, Joseph B., Sharon M. Vlasak, Anne C. Thometz, and Jane E. Buikstra
1982 A comparative study of the chemical analysis of ribs and femurs in Woodland populations. *American Journal of Physical Anthropology* 59: 289-294.

Lambert, Joseph B., Sharon Vlasak Simpson, Jane E. Buikstra, and Douglas Hanson
1983 Electron microprobe analysis of elemental distribution in excavated human femurs. *American Journal of Physical Anthropology* 62: 409-423.

Lambert, Joseph B., Sharon Vlasak Simpson, Jane E. Buikstra, and Douglas K. Charles
1984a Analysis of Soil Associated with Woodland Burials. In *Archaeological Chemistry III*, edited by Joseph B. Lambert, pp. 97-113. American Chemical Society, Washington, D.C.

Lambert, J.B., S.V. Simpson, C.B. Szpunar, and Jane E. Buikstra
1984b Copper and Barium as Dietary Discriminants: The Effects of Diagenesis. *Archaeometry* 26: 131-138.

Lambert, Joseph B., Sharon Vlasak Simpson, Susan Gorell Weiner, and Jane E. Buikstra
1985 Induced Metal-Ion Exchange in Excavated Human Bone. *Journal of Archaeological Science* 12: 85-92.

Lambert, Joseph B., Liang Xue, and Jane E. Buikstra
1989 Physical Removal of Contaminative Inorganic Material from Buried Human Bone. *Journal of Archaeological Science* 16:427-436.

Lappalainen, R., M. Knuuttila, S. Lammi, E.M. Alhava, and H. Olkkonen
1982 Zn and Cu content in human cancellous bone. *Acta Orthopaedica Scandinavica* 53: 51-55.

Lazenby, Richard A.
1984 Inherent Deficiencies in Cortical Bone Microstructural Age Estimation Techniques. *OSSA* 9-11: 95-103.

Leeson, C. Roland, Thomas C. Leeson, and Anthony A. Paparo
1985 *Atlas of Histology*. W.B. Saunders, Philadelphia.

Lynch, B. Mark and Richard W. Jefferies
1982 A Comparative Analysis of the Nitrogen Content of Bone as a Means of Establishing a Relative Temporal Ordination of Prehistoric Burials. *Journal of Archaeological Science* 9: 381-390.

Marchiafava, Valeria, E. Bonucci, and A. Ascenzi
1974 Fungal Osteoclasia: a Model of Dead Bone Resorption. *Calcified Tissue Research* 14: 195-210.

McConnell, D.
1973 *Apatite*. Springer Verlag, Vienna.

Nelson, B., M.J. DeNiro, M.J. Schoeninger, and D.J. DePaolo
1983 Strontium isotope evidence for diagenetic alteration of bone: consequences for diet reconstruction. *Geological Society of America Bulletin* 15: 652.

Nelson, Dorothy A. and Norman J. Sauer
1984 An Evaluation of Postdepositional Changes in the Trace Element Content of Human Bone. *American Antiquity* 49: 141-147.

Nelson, R.E.
1982 Carbonate and Gypsum. In *Methods of Soil Analysis* (part 2), edited by A.L. Page, R.H. Miller, and D.R. Keeney, pp.181-197. American Society of Agronomy, Inc., and Soil Science Society of America, Inc., Madison, Wisconsin.

Neuman, William F.
1980 Bone Material and Calcification Mechanisms. In *Fundamental and Clinical Bone Physiology*, edited by Marshall R. Urist, pp. 83-107. J.B. Lippincott, Philadelphia.

Neuman, William F. and Margaret W. Neuman
1958 *The Chemical Dynamics of Bone Mineral*. University of Chicago Press, Chicago.

Newesely, Heinrich
1980 Ab- und Umbauvorgänge der biologischen Hartgewebe (Knochen, Zähne) unter langer Liegezeit. *Berliner Beiträge zur Archäometrie* 5: 175-186.

1988 Chemical Stability of Hydroxyapatite under Different Conditions. In *Trace Elements in Environmental History*, edited by Gisela Grupe and Bernd Herrmann, pp. 1-16. Springer Verlag, Berlin (West).

Nielsen, Forrest H.
1986 Other Elements: Sb, Ba, B, Br, Cs, Ge, Rb, Ag, Sr, Sn, Ti, Zr, Be, Bi, Ga, Au, In, Nb, Sc, Te, Tl, W. In *Trace Elements in Human and Animal Nutrition - Fifth Edition* (vol. 2), edited by Walter Mertz, pp. 415-463. Academic Press, Inc., Orlando.

Olson, R.V. and Roscoe Ellis Jr.
1982 Iron. In *Methods of Soil Analysis* (part 2), edited by A.L. Page, R.H. Miller, and D.R. Keeney, pp. 301-312. American Society of Agronomy, Inc., and Soil Science Society of America, Inc., Madison, Wisconsin.

Ortner, Donald J. and Walter G.J. Putschar
1985 *Identification of Pathological Conditions in Human Skeletal Remains*. Smithsonian Institution Press, Washington, D.C. (reprint of 1981 edition)

Parker, Ronald B. and Heinrich Toots
1980 Trace Elements in Bones as Paleobiological Indicators. In *Fossils in the Making*, edited by Anna K. Behrensmeyer and Andrew P. Hill, pp. 197-207. University of Chicago Press, Chicago.

Parker, Ronald B., Heinrich Toots, and J.W. Murphy
1974 Leaching of Sodium from Skeletal Parts during Fossilization. *Geochimica et Cosmochimica Acta* 38: 1317-1321.

Pate, F. Donald and John T. Hutton
1988 The Use of Soil Chemistry Data to Address Post-mortem Diagenesis in Bone Mineral. *Journal of Archaeological Science* 15: 729-739.

Pendall, Elise Gislaine
1989 *Natural and Anthropogenic Influences on Soil Development at the Harappa Archaeological Site, Pakistan.* Master of Arts thesis, University of of Califiornia at Berkeley.

Piepenbrink, Hermann
1984 Beispiele biogene Dekompositionserscheinungen an Knochen unter längerer Liegezeit. *Anthropologischer Anzeiger* 42: 241-251.

1986a Two Examples of Biogenous Dead Bone Decomposition and their Consequences for Taphonomic Interpretation. *Journal of Archaeological Science* 13: 417-430.

1986b Prinzipien der Knochendekomposition und ihre Konsequenzen für die diagnostische Bearbeitung von Skeletfunden. In *Innovative Trends in der prähistorischen Anthropologie*, edited by Bernd Herrmann, pp. 15-21. (*Mitteilungen der Berliner Gesellschaft für Anthropologie, Ethnologie und Urgeschichte* 7).

Piepenbrink, H. and H. Schutkowski
1987 Decomposition of skeletal remains in desert dry soil. A roentgenological study. *Human Evolution* 2: 481-491.

Posner, Aaron S.
1969 Crystal Chemistry of Bone. *Physiological Reviews* 49: 760-792.

Posner, Aaron S. and Ralph A. Beebe
1975 The Surface Chemistry of Bone Mineral and Related Calcium Phosphates. *Seminars in Arthritis and Rheumatism* 4: 267-291.

Posner, Aaron S., F. Betts, and N.C. Blumenthal
1979 Bone Mineral Composition and Structure. In *Skeletal Research: An Experimental Approach*, edited by David J. Simmons and Arthur S. Kunin, pp. 167-192. Academic Press, New York.

Price, T. Douglas
1989 Multi-element studies of diagenesis in prehistoric bone. In *The Chemistry of Prehistoric Human Bone*, edited by T. Douglas Price, pp. 126-154. Cambridge University Press, Cambridge.

Price, T. Douglas, Margaret J. Schoeninger, and George J. Armelagos
1985 Bone chemistry and past behavior: an overview. *Journal of Human Evolution* 14: 419-447.

Price, T. Douglas, George J. Armelagos, Jane E. Buikstra, M. Pamela Bumsted, Brian S. Chisholm, Jonathon E. Ericson, Joseph B. Lambert, Nikolaas J. van der Merwe, Margaret J. Schoeninger, and Andrew Sillen
1989 The chemistry of prehistoric human bone: recommendations and directions for future study. In *The Chemistry of Prehistoric Human Bone*, edited by T. Douglas Price, pp. 245-252. Cambridge University Press, Cambridge.

Radosevich, Stefan
1989 Geochemical Techniques Applied to Bone from South Asia and Alaska: Neither God's Truth or Hocus-Pocus. In *Old Problems and New Perspectives in the Archaeology of South Asia*, edited by Jonathan Mark Kenoyer, pp. 93-102. (*Wisconsin Archaeological Reports* 2).

Renfrew, Colin
1989 *Archaeology and Language*. Penguin Books, London. (reprint of 1987 Jonathan Cape edition)

Rogers, Juliet and Tony Waldron
1985 Lead Concentrations in Bones from a Neolithic Long Barrow. *Journal of Archaeological Science* 12: 93-96.

Runia, Lex
1988 Discrimination factors on different trophic levels in relation to the trace element content in human bones. In *Trace Elements in Environmental History*, edited by Gisela Grupe and Bernd Herrmann, pp. 53-66. Springer Verlag, Berlin (West).

Sagne, Sören
1978 The use of scanning electron microscope with x-ray equipment for the analysis of elements in medieval bones. *OSSA* 5: 85-92.

Salmon, Leonard and Peter A. Cawse
1983 Instrumental Neutron Activation Analysis. In *Soil Analysis*, edited by Keith A. Smith, pp. 299-353. Marcel Dekker, Inc., New York.

Sargent-Welch
1980 *Table of Periodic Properties of the Elements*. Sargent-Welch Scientific Company, Skokie, Illinois.

Schoeninger, Margaret J.
1979 Diet and Status at Chalcatzingo: Some Empirical and Technical Aspects of Strontium Analysis. *American Journal of Physical Anthropology* 51: 295-310.

Schroeder, Henry A., Isabel H. Tipton, and Alexis B. Nason
1972 Trace Metals in Man: Strontium and Barium. *Journal of Chronic Diseases* 25: 491-517.

Schultz, Michael
1986 *Die mikroskopische Untersuchung prähistorischer Skeletfunde*. Amt für Museen und Archäologie, Liestal, Switzerland. (*Archäologie und Museum* 6)

Sealy, Judith C. and Andrew Sillen
1988 Sr and Sr/Ca in Marine and Terrestrial Foodwebs in the Southwestern Cape, South Africa. *Journal of Archaeological Science* 15: 425-438.

Sillen, Andrew
1981 Strontium and Diet at Hayonim Cave. *American Journal of Physical Anthropology* 56: 131-137.

1986 Biogenic and diagenetic Sr/Ca in Plio-Pleistocene fossils of the Omo Shungura Formation. *Paleobiology* 12: 311-323.

1989 Diagenesis of the inorganic phase of cortical bone. In *The Chemistry of Prehistoric Human Bone*, edited by T. Douglas Price, pp. 211-229. Cambridge University Press, Cambridge.

Sillen, Andrew and Maureen Kavanagh
1982 Strontium and Paleodietary Research: A Review. *Yearbook of Physical Anthropology* 25: 67-90.

Sillen, Andrew and Patricia Smith
1984 Weaning Patterns are Reflected in Strontium-Calcium Ratios of Juvenile Skeletons. *Journal of Archaeological Science* 11: 237-245.

Triffit, J.T.
1985 Receptor Molecules, Coprecipitation and Ion Exchange Processes in the Deposition of Metal Ions in Bone. In *Metals in Bone*, edited by Nicholas D. Priest, pp. 3-20. MTP Press, Lancaster.

Tuross, Noreen, Anna K. Behrensmeyer, and E.D. Eanes
1989 Strontium Increases and Crystallinity Changes in Taphonomic and Archaeological Bone. *Journal of Archaeological Science* 16: 661-672.

Urist, Marshall R.
1976 Biochemistry of Calcification. In *The Biochemistry and Physiology of Bone* (vol. 4), edited by Geoffrey H. Bourne, pp. 1-59. Academic Press, New York.

Vaughan, Janet
1981 *The Physiology of Bone.* Clarendon Press, Oxford.

Vincent, Jacques
1963 Microscopic Aspects of Mineral Metabolism In Bone Tissue With Special Reference to Calcium, Lead and Zinc. *Clinical Orthopaedics and Related Research* 26: 161-175.

Von Endt, David W. and Donald J. Ortner
1984 Experimental Effects of Bone Size and Temperature on Bone Diagenesis. *Journal of Archaeological Science* 11: 247-253.

Wasserman, Robert H.
1974 Strontium. In *Geochemistry and the Environment* (vol. 2), pp. 73-87. National Academy of Sciences, Washington, D.C.

White, E.M. and L.A. Hannus
1983 Chemical Weathering of Bone in Archaeological Soils. *American Antiquity* 48: 316-322.

Whittaker, D.K. and M.V. Stack
1984 The Lead, Cadmium and Zinc Content of Some Romano-British Teeth. *Archaeometry* 26: 37-42.

Willard, Hobart H., Lynne L. Merritt Jr., John A. Dean, and Frank A. Settle Jr.
1988 *Instrumental Methods of Analysis* (seventh edition). Wadsworth Publishing, Belmont, California.

Zipkin, I.
1970 The Inorganic Composition of Bones and Teeth. In *Biological Calcification: Cellular and Molecular Aspects*, edited by Harald Schraer, pp. 69-103. Meredith Corporation, New York.

APPENDIX

Peak Areas (PA), Peak Area Errors, Decay Correction Factors (DCF), and Corrected Peak Areas (CPA) for Bone and Soil Samples

1. Barium 131

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	1130.2	4.6	3.85	4351.3
H87/60-46a	*Bos*	220.6	15.5	3.89	858.1
	Canis	274.4	12.8	3.97	1089.4
H87/60-46a-26	E[2]	73.3	29.4	3.44	252.2
	C[3]	64.0	36.6	3.46	221.4
	P[4]	53.0	59.5	3.49	185.0
H87/60-46a-77	E	62.0	41.2	3.51	217.6
	C	67.0	36.7	3.54	237.2
	P	103.0	25.9	3.56	366.7
H87/85-49d.2	C	320.8	7.9	3.59	1151.7
	Bos	484.6	8.8	3.91	1894.8
	soil	767.7	8.3	4.18	3209.0
H87/136-147a	E	740.5	4.7	3.61	2673.2
	C	1367.1	3.4	3.64	4976.2
	P	2228.9	2.6	3.66	8157.8
	soil	910.1	6.6	4.20	3822.4
H87/137-148a	E	654.6	5.7	3.69	2415.5
	C	289.7	9.0	3.71	1074.8
	P	515.9	7.0	3.73	1924.3
	soil	273.3	13.6	4.23	1156.1
H87/197-194a	E	309.1	8.2	3.75	1159.1
	C	185.0	11.3	3.76	695.6
	P	398.7	7.2	3.81	1519.0
H87/197-194b		600.5	6.0	3.83	2299.9
Saw Blade		0			

1. % at 1 sd.
2. Endosteal portion of femoral cross-section.
3. Centre portion of femoral cross-section.
4. Periosteal portion of femoral cross-section.

2. Strontium 85

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	3002.7	2.3	1.28	3843.5
H87/60-46a	*Bos*	1804.3	2.9	1.28	2309.5
	Canis	2468.9	2.4	1.28	3160.2
H87/60-46a-26	E	605.8	5.2	1.25	757.3
	C	786.0	4.4	1.25	982.5
	P	999.7	4.0	1.26	1259.6
H87/60-46a-77	E	819.2	4.4	1.26	1032.2
	C	682.1	4.7	1.26	859.4
	P	977.5	4.0	1.26	1231.7
H87/85-49d.2	C	784.3	4.4	1.26	988.2
	Bos	1511.8	3.4	1.28	1935.1
	soil	276.4	22.8	1.30	359.3
H87/136-147a	E	758.9	4.6	1.26	956.2
	C	1405.2	3.4	1.26	1770.6
	P	2092.7	2.7	1.27	2657.7
	soil	460.1	13.7	1.30	598.1
H87/137-148a	E	2185.7	2.5	1.27	2775.8
	C	1223.5	3.4	1.27	1553.8
	P	2468.0	2.4	1.27	3134.4
	soil	116.0	38.6	1.30	150.8
H87/197-194a	E	926.7	4.0	1.27	1176.9
	C	644.1	4.8	1.27	818.0
	P	1024.4	3.8	1.28	1311.2
H87/197-194b		2473.3	2.4	1.28	3165.8
Saw Blade		0			

1. % at 1 sd.

3. Zinc 65

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	1022.9	3.5	1.07	1094.5
H87/60-46a	*Bos*	564.1	4.7	1.07	603.6
	Canis	618.7	4.5	1.07	662.0
H87/60-46a-26	E	187.1	8.4	1.06	198.3
	C	234.8	7.3	1.06	248.9
	P	371.8	5.8	1.06	394.1
H87/60-46a-77	E	265.7	6.9	1.06	281.6
	C	212.8	7.6	1.06	225.6
	P	370.8	5.8	1.06	393.0
H87/85-49d.2	C	314.4	6.4	1.06	333.3
	Bos	630.3	4.4	1.07	674.4
	soil	515.7	7.0	1.07	551.8
H87/136-147a	E	251.6	7.2	1.06	266.7
	C	473.0	5.2	1.06	501.4
	P	731.6	4.2	1.06	775.5
	soil	553.8	6.8	1.07	592.6
H87/137-148a	E	338.4	6.2	1.07	362.1
	C	191.5	8.3	1.07	204.9
	P	477.3	5.1	1.07	510.7
	soil	266.7	8.9	1.07	285.4
H87/197-194a	E	190.6	8.3	1.07	203.9
	C	124.3	10.3	1.07	133.0
	P	228.3	7.6	1.07	244.3
H87/197-194b		541.7	4.8	1.07	579.6
Saw Blade		0			

1. % at 1 sd.

4. Calcium 47

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	568.3	4.7	33.44	19004.0
H87/60-46a	*Bos*	301.7	6.4	34.09	10285.0
	Canis	280.0	5.8	35.94	12938.4
H87/60-46a-26	E	188.5	8.0	24.86	4686.1
	C	190.8	8.0	25.34	4834.9
	P	330.1	6.1	25.78	8510.0
H87/60-46a-77	E	209.1	7.6	26.30	5499.3
	C	160.2	8.7	26.76	4287.0
	P	259.8	6.8	27.29	7089.9
H87/85-49d.2	C	138.5	10.0	27.66	3830.9
	Bos	498.5	5.0	34.76	17327.9
	soil	74.7	17.5	41.34	3088.1
H87/136-147a	E	157.1	9.7	28.25	4438.1
	C	287.4	7.0	28.78	8271.4
	P	394.6	6.1	29.34	11577.6
	soil	105.8	14.3	41.82	4424.6
H87/137-148a	E	326.3	6.2	29.86	9743.3
	C	202.9	7.9	30.40	6168.2
	P	339.6	6.0	30.67	10415.5
	soil	39.4	22.0	42.57	1677.3
H87/197-194a	E	155.2	9.2	31.28	4854.7
	C	107.9	11.1	31.43	3391.3
	P	186.0	8.3	32.41	6028.3
H87/197-194b		245.8	7.2	32.87	8079.4
Saw Blade		0			

1. % at 1 sd.

5. Sodium 24

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	9453.3	1.1	1.21	11438.5
H87/60-46a	*Bos*	3650.3	1.8	1.21	4416.9
	Canis	6333.3	1.4	1.22	7726.6
H87/60-46a-26	E	2241.2	2.3	1.16	2599.8
	C	2519.5	2.2	1.17	2947.8
	P	3797.9	1.8	1.17	4443.5
H87/60-46a-77	E	2964.4	2.0	1.17	3468.3
	C	1638.2	2.7	1.17	1916.7
	P	3643.5	1.8	1.18	4299.3
H87/85-49d.2	C	2778.3	2.1	1.18	3278.4
	Bos	10454.5	1.1	1.21	12649.9
	soil	3434.6	1.9	1.24	4258.9
H87/136-147a	E	983.1	3.5	1.18	1160.1
	C	1760.8	2.6	1.18	2077.7
	P	2654.7	2.1	1.19	3159.1
	soil	3145.3	1.9	1.24	3900.2
H87/137-148a	E	1979.2	2.4	1.19	2355.2
	C	1363.0	3.0	1.19	1622.0
	P	2235.8	2.3	1.19	2660.6
	soil	1430.8	2.9	1.24	1774.2
H87/197-194a	E	3518.0	1.8	1.20	4221.6
	C	2525.5	2.1	1.20	3030.6
	P	3696.8	1.8	1.20	4436.2
H87/197-194b		8616.2	1.2	1.20	10339.4
Saw Blade		0			

1. % at 1 sd.

6. Aluminum 28

Sample		PA	Error[1]
H87/40-34a	*Bos*	217	14.2
H87/60-46a	*Bos*	238	13.0
	Canis	338	11.3
H87/60-46a-26	E	119	18.3
	C	82	22.1
	P	90	22.8
H87/60-46a-77	E	193	14.4
	C	63	25.2
	P	225	13.3
H87/85-49d.2	C	119	18.3
	Bos	219	14.2
	soil	34488	1.2
H87/136-147a	E	37	36.0
	C	97	20.3
	P	111	24.8
	soil	37853	1.2
H87/137-148a	E	212	13.7
	C	78	22.6
	P	170	15.3
	soil	20184	1.5
H87/197-194a	E	128	17.7
	C	63	25.2
	P	133	17.3
H87/197-194b		2106	4.5
Saw Blade		42	30.9

1. % at 2 sd.

7. Manganese 56

Sample		PA	Error[1]	DCF	CPA
H87/40-34a	*Bos*	76.5	23.4	2.89	221.1
H87/60-46a	*Bos*	166.9	10.0	2.92	487.3
	Canis	127.8	14.4	3.03	387.2
H87/60-46a-26	E	212.5	8.1	2.38	505.8
	C	100.9	13.6	2.40	242.2
	P	323.5	6.7	2.43	786.1
H87/60-46a-77	E	422.8	5.8	2.46	1040.1
	C	66.3	16.9	2.49	165.1
	P	796.1	4.1	2.52	2006.2
H87/85-49d.2	C	167.7	10.2	2.55	427.6
	Bos	153.7	15.4	2.96	455.0
	soil	6105.0	1.4	3.32	20268.6
H87/136-147a	E	49.9	19.1	2.57	128.2
	C	49.4	21.1	2.60	128.4
	P	332.1	6.4	2.63	873.4
	soil	5660.5	1.4	3.36	19019.3
H87/137-148a	E	124.1	11.2	2.67	331.3
	C	88.1	13.1	2.70	237.9
	P	297.3	6.9	2.73	811.6
	soil	2965.1	2.0	3.40	10081.3
H87/197-194a	E	128.6	14.2	2.76	354.9
	C	0			
	P	281.0	7.4	2.82	792.4
H87/197-194b		806.7	4.4	2.86	2307.2
Saw Blade		9418.6	1.1	3.28	30893.0

1. % at 1 sd.