AN EXAMINATION OF MUSCULOSKELETAL MARKERS TO ANALYZE OCCUPATIONAL TYPE OF A MODERN DOCUMENTED POPULATION USING THE COIMBRA METHOD

by

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DEDICATION

This thesis is dedicated to the donors of the Texas State University Donated Skeletal Collection as well as their families. Without their generosity, none of this research would be possible.

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TABLE OF CONTENTS

| | Page |
|---------------------------------------|------|
| ACKNOWLEDGEMENTS | v |
| LIST OF TABLES | viii |
| LIGHT OF FIGURES | |
| LIST OF FIGURES | 1X |
| LIST OF ABBREVIATIONS | X |
| ABSTRACT | xi |
| CHAPTER | |
| I. INTRODUCTION | 1 |
| Entheseal Sites | 3 |
| Definition of Terms | 3 |
| Anatomy of an Entheseal Site | 3 |
| Different Types of Entheseal Sites | |
| Important Features of Entheseal Sites | 5 |
| Empirical Bioarchaeological Studies | 6 |
| Labor and Activity | 7 |
| Documented Collections | 8 |
| The Coimbra Method | 9 |
| Forensic Anthropological Applications | |
| Research Questions | |
| Summary | 13 |
| II. MATERIALS AND METHODS | 14 |
| Labor Classification | 15 |
| Age Categorization | 16 |
| Socio-economic Status | |
| Chosen Entheseal Sites | |
| Collection of Ordinal Data | |
| Variety in Expression | 21 |
| Statistical Methods | 21 |

| III. RESULTS | 23 |
|---|----|
| Intra-Observer Error | 23 |
| Generalized Estimating Equations | |
| IV. DISCUSSION AND CONCLUSION | 35 |
| Research Question #1: Activity Levels and Occupational Type | 35 |
| Research Question #2: Other Explanatory Variables | 37 |
| Research Question #3: Error and Accuracy | 40 |
| Performance of Method | 41 |
| Conclusion | |
| APPENDIX SECTION | 45 |
| LITERATURE CITED | 54 |

LIST OF TABLES

| Table | Page |
|--|------|
| Demographics of Study Sample from TXSTDSC | 14 |
| 1. Number of TXSTDSC Individuals Per Category | 16 |
| 3. Selected Entheseal Sites | 18 |
| 4. Methodology of Scoring | 20 |
| 5. Explanatory Variables | 22 |
| 6. Descriptive Statistics for ECs on Humerus | 24 |
| 7. Descriptive Statistics for ECs on Radius and Ulna | 25 |
| 8. Intra-observer Error Percentage Agreements | 25 |
| 9. p-values (Pr(> W)) for <i>m. subscapularis</i> | 26 |
| 10. p-values (Pr(> W)) for Common Extensor Origin | 27 |
| 11. p-values (Pr(> W)) for Common Flexor Origin | 28 |
| 12. p-values (Pr(> W)) for m. supraspinatus | 29 |
| 13. p-values (Pr(> W)) for m. infraspinatus | 30 |
| 14. p-values (Pr(> W)) for m. biceps brachii | 31 |
| 15. p-values (Pr(> W)) for m. triceps brachii | 32 |
| 16. Summary of Correlations | 34 |

LIST OF FIGURES

| Figure | Page |
|---|------|
| 1. Age Category Distribution | 16 |
| 2. Age Separated by Sex | 16 |
| 3. Socio-economic Status Distribution | 17 |
| 4. Summary of scores pertaining to SES for <i>M. subscapularis</i> | 27 |
| 5. Summary of scores pertaining to AGE for <i>M. subscapularis</i> | 27 |
| 6. Summary of scores pertaining to AGE for CEO | 28 |
| 7. Summary of scores pertaining to SES for CFO | 29 |
| 8. Summary of scores pertaining to SES for <i>M. supraspinatus</i> | 30 |
| 9. Summary of scores pertaining to SEX and AGE for <i>M. infraspinatus</i> | 31 |
| 10. Summary of scores pertaining to OCCUPATION SEX for <i>M. biceps brachii</i> | 32 |
| 11. Summary of scores pertaining to AGE for <i>M. biceps brachii</i> | 32 |
| 12. Summary of scores pertaining to SES for <i>M. triceps brachii</i> | 33 |

LIST OF ABBREVIATIONS

Abbreviation Description

EC Entheseal Change

FC Fibrocartilaginous Enthesis

TXSTDSC Texas State Donated Skeletal Collection

SES Socio-economic Status

BF Bone Formation

E Erosion

FP Fine Porosity

MP Macro-Porosity

C Cavitation

TC Textural Change

GEE Generalized Estimating Equations

GLM Generalized Linear Model

ABSTRACT

Determining occupational type as part of the biological profile has significant implications for human identification in forensic anthropology. Overall increased activity levels have been shown to manifest in musculoskeletal markers, otherwise referred to as entheseal changes. Although several different methods have been used to assess entheseal changes in bioarchaeological collections, few approaches have been used to examine modern documented collections (Cardoso & Henderson, 2010; Villotte et al., 2010; Henderson, Mariotti, Pany-Kucera, Villotte, & Wilczak, 2013; Nolte & Wilczak, 2013; Mountrakis & Manolis, 2015; Henderson, Mariotti, Pany-Kucera, Villotte, & Wilczak, 2016; Henderson, Mariotti, Santos, Villotte, & Wilczak, 2017; Michopoulou, Nikita, & Henderson, 2017).

Henderson and colleagues (2013) created the Coimbra Method to score specific features of fibrocartilaginous entheseal changes in known bioarchaeological collections. This study applied the same method to a recently donated skeletal collection with self-reported information to examine whether the method could be used in a forensic anthropological context. Generalized Estimating Equations were used to see possible correlations between the different features of the entheseal sites and the activity levels based on the reported occupational type, specifically manual and non-manual labor. Correlations with variables such as socio-economic status, sex, and age were also compared to correlations seen with activity levels to assess whether the reported

occupational type aptly described the resulting entheseal changes compared to the outside variables.

The results showed that although the Coimbra Method was a useful method for examining the severity of entheseal changes in a modern individual, the entheseal changes did not often correlate to reported occupations and their assumed activity levels. Other variables such as age and sex showed mixed results dependent on the entheseal site and feature being examined. Out of all the variables, socio-economic status showed the most consistent correlation with many of the entheseal sites, indicating that the lower levels usually had more entheseal changes than the higher levels. However, all variables were inconsistent overall, suggesting that entheseal sites in general represent a mosaic of factors in an individual's life, and they should not be tied to only one variable when being analyzed. Results also suggested that recent documented populations with self-reported data do not showcase the full extent of the activity levels of each individual in their reported occupations, and research focused on such subjects should be careful and not limit their criteria to just occupation.

The Coimbra Method as a whole presented itself as a viable tool in examining the individual features seen in an entheseal site, but the method was rather complicated for researchers who are just learning the different rules and measurements included in the original and updated publications (Henderson et al., 2013; Henderson et al., 2016). A potential future focus for this method could be to include a full photography series indicating the different stages and scores of the entheseal sites to ensure better accuracy for future research.

I. INTRODUCTION

Knowledge of a deceased individual's previous activity patterns can provide crucial information for a biological profile. Forensic anthropologists often use ancestry, sex, and age to estimate the identity of an unknown individual, but background information of physical activity and occupation classification could potentially help narrow the search for the individual's identity even further. Studies focused on activity-related changes to bone have demonstrated reliable methods to determine general activity patterns, such as manual and non-manual labor, as well as possible occupational categories in known bioarchaeological skeletal collections (Cardoso & Henderson, 2010; Villotte et al., 2010; Henderson et al., 2013; Nolte & Wilczak, 2013; Mountrakis & Manolis, 2015; Henderson et al., 2016; Henderson et al., 2017; Michopoulou et al., 2017). Some of these studies have used methods that focus on specific muscle attachments visible on the bone, otherwise known as entheseal sites, to measure and analyze past activity patterns (Villotte et al., 2010; Henderson et al., 2013; Mountrakis & Manolis, 2015; Henderson et al., 2017; Michopoulou et al., 2017).

Studies on activity-related changes to bone have increased in popularity over the past decade in biological anthropological research (Milella, Cardoso, Assis, Lopreno, & Speith, 2015). Biological anthropologists are interested in entheseal sites for several reasons, the most important being: 1) entheses are under physical strain and pressure during rigorous activity done in an individual's lifetime; 2) there is varying severity according to the activity patterns of the individual seen in entheseal changes, making them possible to score; and 3) entheseal sites are usually clear and visible on dry bone and are thus easy to report. Because of these reasons, bioarcheologists have created

methods to use these entheseal sites to reconstruct past activities and possible occupations (Villotte & Knusel, 2013). These studies and methods vary significantly depending on the different types of entheseal sites selected, as well as which specific method to use and which population to examine. Many studies compare the results of the entheseal site examinations to each individual's occupation and associated activity level (Villotte et al., 2010; Cardoso & Henderson, 2010; Cardoso & Henderson, 2013). Others examine how other variables such as age might have a similar effect on entheseal changes (Weiss, 2007; Cardoso & Henderson, 2010). Several also examine the observer error that accompany each method (Henderson et al., 2013; Henderson et al., 2016; Henderson et al., 2017). Although the results show promising accuracy in identifying activity patterns in past populations, few activity-related studies have focused on recent or modern, well-documented skeletal samples and their associated occupations.

This present study explored one of these methods, the Coimbra Method, to assess whether the method can be applied in a forensic anthropological setting by examining the relationship between occupational activity and entheseal sites seen in recently deceased individuals. This study examined the possible impacts that various confounding variables may have on the results. By focusing on a recently deceased sample, this study was able to rely on self-reported data regarding the individuals' occupations, ages, socio-economic statuses, and other such variables. By comparing the known information from a recently documented sample with the entheseal site activity seen through the lens of the Coimbra Method (Henderson et al. 2013; Henderson et al. 2016; Henderson et al. 2017), this study explored the potential use of occupational stress markers to predict activity level and occupational type.

Entheseal Sites

Definition of Terms

The term "entheseal site" or "enthesis" is used to define the musculoskeletal markers that show tendon attachments. "Entheseal change" loosely defines any osseous change to an enthesis based on the visible features of the site. An enthesopathy or an "enthesophyte" is the possible pathology that can occur in these sites and is thus not the focus of this study (Villotte & Knusel, 2013).

Various osteological terms are assigned to the features seen in entheses, such as musculoskeletal markers and muscular crests and markings. Depending on the study or the researcher, different terms are often used to describe the features seen in the entheseal sites.

Anatomy of an Entheseal Site

An entheseal site is a concentrated site of stress where a tendon attaches its associated muscle to bone. These sites serve as the location for fibrous connective tissue to attach the tendon to the periosteum, and they are complex loading sites that transfer force to and from the skeleton, allowing for multi-axis bending and compounding the stress exerted on the site (Benjamin et al., 2006). Since most tendons attach obliquely to bone, the actual contact is fanned out from the initial point of attachment. This phenomenon dissipates the stress at the entheseal site, and often results in formations of fibrocartilages (Benjamin et al., 2006).

The fibrocartilage is separated into two distinct parts of calcification. The uncalcified portion of the fibrocartilage sits atop the calcified fibrocartilage, which has direct contact with the bone, creating a boundary between the cartilage and the

periosteum. This is referred to as a "tidemark" (Villotte, 2012). The outermost area of the tidemark creates the margin of a normal fibrocartilaginous entheseal site, and the acute angle created by the tidemark emphasizes the directional "pull" of the tendon and marks the center of the entheseal site. This study will focus and expand upon the mechanical factors that influence each of these different zones of the entheseal sites.

Different Types of Entheseal Sites

The two main categories of entheseal sites that researchers have generally agreed upon are fibrocartilaginous entheses and fibrous entheses. Fibrocartilaginous entheses (FC) are characterized by smooth, well defined bone with no foramina, and they are located at cartilaginous tendon attachment sites on bone. Any deviation from this could be categorized as an entheseal change (Villotte et al., 2016). Fibrous entheses, which are located on cortical bone, are found on long bones and vertebrae. Although both types of entheses have been used in entheseal studies, FC entheseal sites are more commonly chosen because the baseline appearance of an FC entheseal site (what an observer would normally score as a zero) is clearer and easier to identify than the normal appearance of a fibrous entheseal site (Henderson et al., 2013). Fibrous entheseal site topography is often dominated by porosity, roughness, and other alterations, making scoring more difficult (Michopoulou et al., 2017). Also, and more importantly for this particular study, FC entheseal sites have been shown to have a higher correlation with physical activity than fibrous entheseal sites (Villotte et al., 2010; Weiss, 2015; Michopoulou et al., 2017).

There are several FC entheseal sites in the human body, although some have proven to be more informative in physical activity studies than others. General

conclusions from previous studies have shown that most FC entheseal sites are mosaics showing a broad range of activities of an individual (Kennedy, 1998). They display the basic patterns of stress resulting from habitual activities of that individual. However, certain markers of stress can show more specific strenuous labor based on where the entheseal site is located. According to Kennedy (1998), the upper and lower extremities have certain entheseal site locations that can show isolated markers of repeated physical stress. These markers are more visible on the bone because of the elevated activity resulting in increased bone growth, which creates more prominent crests, ridges, and pitting (Michopoulou et al., 2017).

Important Features of Entheseal Sites

The visible features of entheseal sites are numerous, and highlighted features vary from study to study. According to Villotte et al. (2016), possible categories for entheseal changes (ECs) are surface discontinuity, mineralized tissue formation, and a complete lack of original morphology. Hawkey also used robusticity, stress-induced lesion, and ossification as prominent features for her study (1998). Henderson and her colleagues used similar characteristics to Hawkey's (1998) study in their own analysis, but they expanded on ossification, including extoses, which are spurs on the bone, as well as added erosion and subcortical cavitations, which are deep cavities in the bone (Henderson et al., 2013; Henderson et al., 2016). The basic features and characteristics are relatively similar between studies, but the differences seen depend mainly on the focus of the respective study. Different methods have attempted to utilize these differences to predict specific physical activity patterns, such as the Villotte (2006) method and the Coimbra Method (Henderson et al., 2013; Henderson & Nikita, 2016).

Empirical Bioarchaeological Studies

Entheseal changes (ECs) have long been used as a tool to reconstruct past activity of archaeological collections (Lopreno, Cardoso, Assis, Milella, & Speith, 2013).

However, the overall focus of such studies has been varied and inconsistent. Many studies have been conducted by several different anthropologists using internationally based collections from different time periods in human history. Many researchers have also created their own method to follow when measuring entheseal changes, which has resulted in an overall lack of standardization.

Hawkey and Merbs (1995) analyzed an ancient Alaskan population to assess whether daily habitual activity patterns could be seen in musculoskeletal markers as well as stress lesions. They found that certain activities that used specific movements caused noticeable wear on the different bones they examined. Robb (1998) analyzed eighteen entheseal sites in individuals from the Iron Age and found that variations in entheseal sites could be linked to activity. In the same year, Stirland (1998) used a sample consisting of all male individuals from two different archaeological collections. She studied muscle insertion sites of the humerus and took two different types of data by visibly scoring the entheseal sites and taking measurements on radiographs. She determined that the scoring must be done using ordinal data since the assigned scores were not actually numerical values with equal distances between each rank (1998). Villotte, following the same logic as Stirland (1998), created a new scoring method in 2006, which reduced inter-observer error levels up to 15% lower than previous studies (Davis, Shuler, Danforth, & Herndon, 2013).

All these studies added important information to what was known about ECs, but their methods remained varied and inconsistent in their scoring and their criteria.

Labor and Activity

Several entheseal site studies focused on correlations with activity patterns typically seen in various occupational types. In general, manual and non-manual laborers were among the most commonly used categories (Kennedy, 1998; Cardoso & Henderson, 2010; Cardoso & Henderson, 2013, Lopreno et al. 2013). These studies focused on the distinction between the two groups by analyzing the biomechanical aspects of the entheseal sites. As a result, many studies found that the lower limbs were useful in assessing occupational use and weight-bearing; however, entheseal sites of the upper limb better reflected activity level, due to the arm, forearm, and hand being utilized more during occupation-related tasks than the lower limbs (Eshed, Gopher, Galili, & Hershkovitz, 2004; Villotte et al, 2010; Cardoso & Henderson, 2013; Mountrakis & Manolis, 2015).

Some studies have also broken each labor category down further into groups regarding activity levels (Villotte et al. 2010, Cardoso & Henderson 2013, Milella et al. 2015). Although the types of occupational groups remain similar across the different studies, the criteria used to differentiate manual from nonmanual labor differed depending on the researchers and their intended study population. Villotte et al. (2010) decided on two main occupational risk factors to help split the laborers into further subgroups: manual activities and forceful tasks, such as lifting or moving heavy loads. However, their results showed few differences between the sub-labor groups. Cardoso and Henderson (2010) also used their own criteria to separate the groups. They looked at

historical records to differentiate the more "physically demanding" occupations with the "less demanding jobs." Lopreno et al. (2013) looked at several of these studies and focused on their grouping criteria specifically. They found that the most important factors to consider based on the typical criteria seen in previous studies were movement of the limbs, carrying of heavy loads, and overall physical tasks required to perform the job successfully.

Documented Collections

An increasing number of researchers also turned their focus from the entheseal sites themselves to the collections from whence they came. Lopreno et al (2013) determined that although many documented collections have useful background data such as occupation, sex, and age, unclear or incomplete information could affect the interpretation of the biological and socio-cultural data. Cardoso and Henderson (2010) also cautioned that due to the individual's socio-economic status at birth, the individual could have undertaken other forms of physical activity more common for someone his or her status, and these activities could have been performed outside the main occupation and thus unlisted in the occupational section for the skeletal collection. Both authors pointed out in 2010 and later on in 2013 that it is also paramount to standardize occupational categories and to always include age as a confounding variable.

Age has been investigated several times as a possible confounding agent to occupational markers examined in entheseal site studies (Milella, Belcastro, Zollikofer, & Mariotti, 2012; Cardoso & Henderson, 2013; Henderson et al., 2013; Henderson et al., 2016; Henderson et al., 2017; Michopoulou et al., 2017). Some results have shown age to correspond strongly with ECs, while others have shown weaker correlations. However,

most researchers agree that age is a variable that also must be considered when analyzing activity patterns and their impact on entheseal sites, regardless of which population is being examined.

Although studies done on entheseal sites have been numerous, their methods regarding which features or variables to examine and consider during analysis have been extremely inconsistent and dependent on the researcher. This has resulted in a field of study that has access to a wealth of information on the general topic but has very little standardization in practice. In an attempt to create a standardized method of examining ECs, Henderson et al. (2013) proposed a new methodology that specifically examined FC entheses of documented skeletal populations, formally known as the Coimbra Method.

The Coimbra Method

The Coimbra Method was introduced after a working group met in 2013 and held a workshop on musculoskeletal stress markers to help standardize entheseal research. The group combined various aspects of past studies with new concepts to create a method that measured fibrocartilaginous entheses by scoring different features separately (Henderson et al., 2013; Henderson et al., 2016). The group split the entheseal sites into two separate zones, each with scoreable features. They chose the historical Coimbra collection, comprised of identified skeletons with physically strenuous occupations, for the original study, using only five generic entheseal sites from the upper and lower appendages. They analyzed the effects of activity and age on the scores and examined the resulting observer error. They found that although the method was not yet ready for widespread use, it provided basic tools to analyze the effect of different factors on entheseal sites such as activity and age.

The same authors wrote a follow-up report on the method in 2016 (Henderson et al., 2016). They changed the scoring slightly by adding a new feature as well as narrowing the scoring range, resulting in the elimination of 5-10% of their previously reported inter-observer error (Henderson et al., 2016). The simplification of the scoring process made it easier to reproduce. Although not all scores were improved and only two entheseal sites were used to compare percentage agreements, the authors recommended the method for widespread use based on an overall 80% agreement rate for observations of both the *m. subscapularis* and the common extensor origin entheseal sites (Henderson et al., 2016).

The most recent study done using the Coimbra Method was conducted by Michopoulou et al. (2017). These authors used a different skeletal collection and focused on the statistical approaches. They used generalized linear models to test if there were recordable activity-related patterns in the ECs. Only FC entheseal sites in the upper limbs were used in the study. The results showed that the revisions to the method done in 2016 did not necessarily improve accuracy of identifying activity-related changes. However, the revisions did show that the impact of body mass was less significant than previously thought, and the correlation between age and entheseal sites was still strong but not as systematic as expected (Michopoulou et al., 2017).

In all, the Coimbra Method is currently the only tested method that scores FC entheses features separately in an attempt to enable the study of the different causations of each independent feature (Henderson et al., 2016). However, it is important to note that this method has been primarily tested on European archaeological and historical samples and collections, with very few studies conducted on any recent reference

samples (Henderson et al., 2013; Henderson et al., 2016; Henderson et al., 2017; Michopoulou et al., 2017).

Forensic Anthropological Application

Occupation is a valuable piece of information for identification of an unknown deceased individual. Musculoskeletal markers are not often used in the past to examine activity patterns in forensic cases, but other body markers have been used to ascertain possible occupations in forensic anthropological studies. Kulshreshtha and Mondal (2017) used scars caused by injuries occurring in various workplaces to determine occupation. They examined scars located on the torso, skull, and appendages of various workers from different Indian villages to examine soft tissue markers related to accidental injuries such as finger pricks, burns, and welding marks. They successfully used distributional patterns of the scars to group the individuals into specific occupational groups. Although they did not use entheseal sites to determine occupation, their study showcased how occupational groupings can be estimated from activity markers relating to their specific occupation.

This present research used the Coimbra Method on a recent collection with birthyears spanning the middle to the late 20th century to analyze the impact of modern physical labor on the skeleton and how different occupational groups could potentially be estimated from the entheseal sites. This study followed the methodology of the Coimbra Method and divided each entheseal site into specific areas to better understand the associated activity levels as well as other variables that can affect the attachment sites. This type of exploratory research was more applicable to modern-day forensic cases due

to the sample demographics as well the associated occupations being more modern than the previously used archaeological populations or historical collections.

Original publications introducing the Coimbra Method have discussed variables such as age in great detail as possible confounding variables when analyzing activity levels (Henderson et al., 2013; Henderson et al., 2016; Henderson et al, 2017; Michopoulou et al., 2017). However, various characteristics of human populations such as sexual dimorphism, economies, and subsistence patterns have changed over time between and among populations. Thus, to properly test these secular changes in a modern population, this study also included other variables such as sex and socio-economic status (SES) to further explore variations in expression.

Based on empirical research, entheseal sites appear to represent a mosaic of different experiences humans have throughout their lifetime, and further research using the Coimbra Method could deconstruct the components of entheseal changes to better understand the types of changes occurring.

Research Questions

This study assessed whether the Coimbra Method, which has been applied primarily to European archeological and historical collections, could be applied to a recently documented North American-based skeletal collection to accurately estimate the individuals' generalized occupational category of either manual or nonmanual labor. It also examined whether variables other than physical activity could significantly affect the entheseal sites. By doing this, the study addressed the following research questions:

- 1) Using the Coimbra Method, can scored fibrocartilaginous entheseal changes seen in the upper limb be used to estimate Texas State University Donated Skeletal Collection's generalized reported occupations?
- 2) Do other variables such as age, sexual dimorphism, and SES significantly alter the entheseal changes and the resulting scores?
- 3) Is there significant intra-observer error (>80% agreement) when visibly scoring musculoskeletal markers on a recent collection using the Coimbra method?

Summary

This study could potentially be applied to several different aspects of anthropology. Although the initial goal was to predict possible occupational groups by observing the entheseal sites on an unidentified set of remains, which forensic anthropologists can then use for identification, this type of study is potentially applicable to bioarcheology and paleoanthropology as well (Agarwal, 2016). Much of the previous studies done on entheseal sites have been on past archaeological populations to assess possible occupational and social roles (Henderson et al., 2013; Lopreno et al., 2013; Milella et al., 2015; Wilcsak, Mariotti, Pany-Kucera, Villotte, & Henderson, 2017). Testing this method on a modern skeletal collection not only tests its repeatability with a different population, but it shows future steps that need to be taken in order to improve its accuracy for future applications in the different realms of anthropology.

II. MATERIALS AND METHODS

Fifty individuals from the Texas State Donated Skeletal Collection (TXSTDSC) were selected and examined following the Coimbra method. Individuals from this collection were willed body donations placed at the Forensic Anthropology Research Facility at Freeman Ranch in San Marcos, Texas. Once the remains reached late decompositional stages, they were brought to the Osteological Research and Processing Laboratory on the same ranch to be processed and cleaned. The remains were finally curated in the TXSTDSC at the Grady Early Forensic Anthropology Research Laboratory.

All individuals included in the sample were born in the mid to late 20th century and died between the years of 2008 and 2015. Each individual's background and history, including age, sex, ancestry, SES, and occupation, were recorded either by the individual before death or by next of kin shortly after death, and all information was compiled by the Forensic Anthropology Center at Texas State University (Table 1).

Table 1. Demographics of Study Sample from TXSTDSC

| Sex and Age Distribution | Ancestry Distribution | Birthplace | | |
|----------------------------|-----------------------|-----------------|--|--|
| Male=34 (avg age: 56.09) | White/Caucasian=47 | Texan=13 | | |
| Female=16 (avg age: 54.94) | Hispanic=2 | Non-Texan=34 | | |
| | African American=1 | International=3 | | |

In order to avoid examining entheseal sites that were too damaged or altered from degenerative changes, an age range of 16-70 years was implemented when selecting each individual for examination (Henderson et al., 2017; Michopoulou et al, 2017). As a result, the average age of the entire sample was 55.7 years. Any individuals exhibiting pathology such as diffuse idiopathic skeletal hyperostosis and seronegative

spondyloarthropathies were excluded to avoid the presence of erosion in the fibrocartilaginous entheses, which have been documented to mimic ECs (Benjamin & McGonagle, 2001; Villotte & Knusel, 2013; Henderson, Mariotti, Santos, Villotte, & Wilczak, 2017; Weiss, 2017). Other general pathologies such as healed fractures and abnormal fusing, as well as any other pathology otherwise obscuring the entheseal sites included in the study, were also excluded.

Labor Classification

All selected individuals had well-defined occupations, each chosen in order to facilitate easier classification as either manual or non-manual labor (Appendix A). Individuals with occupations that did not inherently contain enough information to determine the general amount of workload were excluded (e.g. truck drivers, housewife). All jobs were assumed to be habitual and not as a result of exceptional incidents resulting in lesions (Jurmain, Cardoso, Henderson, & Villotte, 2011). Each job was also assumed to have fit the standard definition of the job title with its typical requirements and procedures in order to classify as manual or nonmanual labor. Jobs requiring moderate to intense levels of physical work and weight bearing/lifting were considered manual labor, while less demanding jobs that primarily involved desk work or computer analytics as well as simple movements such as walking or writing, were considered non-manual. Examples of manual labor frequently seen included occupations such as carpenter (n=3) and mechanic (n=5); common examples of non-manual labor included occupations such as teacher (n=3), real-estate agent (n=3), and architect (n=3).

Out of the fifty individuals selected, twenty-one (42%) were classified as manual laborers, and twenty-nine (58%) were classified as non-manual laborers, each with

varying age ranges. No females with manual labor jobs were included due to a lack of information available or the individual not meeting the basic requirements (Table 2).

Table 2. Number of TXSTDSC Individuals Per Category

| Sex | Occupation | Number Present |
|--------|------------|----------------|
| Male | Manual | 21 |
| Male | Non-manual | 13 |
| Female | Manual | 0 |
| Female | Non-manual | 16 |

Age Categorization

Following a similar model created by Nikita (2014), all ages were compiled into three general categories (Figure 1). The youngest category, ages 16-35, was the smallest of the three (n=3) due to a lack of representation in the collection as a whole, a bias seen in most documented collection samples (Lopreno et al., 2013). The middle-aged category, ages 36-55, contains the majority of a typical American's working life (n=18). The oldest category, ages 56-70, was the most numerous (n=29). The population was overall varied in age between both sexes, but no individuals between the ages of thirty-six and forty fit the criteria necessary to be included in this study (Figure 2).

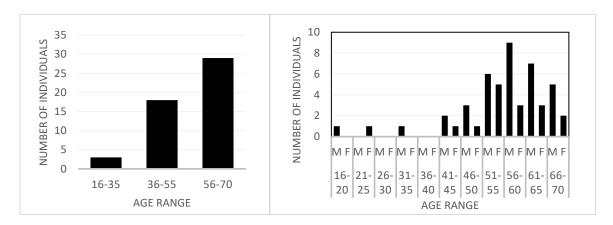


Figure 1. Age Category Distribution.

Figure 2. Age Separated By Sex.

Socio-economic Status

All statuses documented were childhood SES reported by either the individual or next of kin (Figure 3). The typical five categories were collapsed to become a simpler three-tiered system to help bolster the first and last categories: lower to lower middle (n=16), middle (n=23), and middle high to high (n=9). Two individuals had unknown SES and thus were excluded from the statistical tests regarding SES.

It is important to note that when the sample was compared to an overall report on SES in America collected by the National Health and Nutrition Examination Survey between the years of 1999-2004, the distribution of this sample was not reflective of the national average at that time. Family income was calculated as a percentage of the federal poverty line, and 38.5% of those surveyed had an income that was equal to or more than 400% of the poverty line compared to the 12.5% that were 100% or lower (Braveman, Cubbin, Egerter, Williams, & Pamuk, 2010). Due to the lower and middle SES being highest in number for this particular sample, it was unfortunately not entirely representative of the typical SES seen in the United States. Thus, any correlation seen with EC was not assumed to be immediately applicable to American workers today.

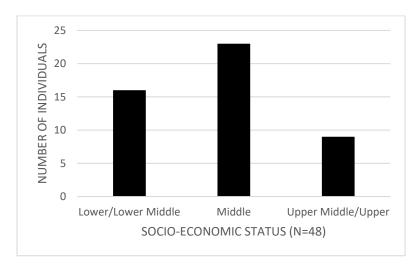


Figure 3. Socio-economic Status Distribution.

Chosen Entheseal Sites

Seven different entheseal sites located on both humeri, radii, and ulnae of each individual were analyzed (Table 3). These entheseal sites were specifically chosen for their high involvement in common movements seen in physical activity and strenuous labor and for being previously highlighted in prominent entheseal site studies (Villotte et al., 2010; Henderson et al., 2013; Milella et al., 2015; Mountrakis & Manolis, 2015; Henderson et al., 2016; Henderson et al. 2017). Although many other fibrocartilaginous (FC) entheseal sites are located throughout the body, these specific sites were selected because all seven involved the arm, forearm, and hand, which muscles have been shown to correspond more with occupation rather than mobility, as opposed to the lower limbs (Cardoso & Henderson 2010, 2013). Each individual had all six skeletal elements present, ensuring that all fourteen entheseal sites were examined for all fifty individuals.

Table 3. Selected Entheseal Sites

| Entheseal site | Location | Related Movement |
|-------------------------|---------------------------|-------------------------------|
| M. subscapularis | Lesser tubercle of the | Medial rotation of shoulder |
| | humerus | and arm |
| Common extensor origin* | Lateral epicondyle of the | Extension of wrist/fingers; |
| (CEO) | humerus | abduction of hand |
| | | |
| Common flexor origin* | Medial epicondyle of the | Pronation of the forearm; |
| (CFO) | humerus | flexion of the wrist/fingers; |
| | | abduction of the hand |
| M. supraspinatus | Greater tubercle of the | Abduction of shoulder and |
| | humerus | arm |
| M. infraspinatus | Greater tubercle of the | Lateral rotation of shoulder |
| | humerus | and arm |
| M. biceps brachii | Radial tuberosity of the | Flexion of shoulder and |
| | radius | arm; supination of forearm |
| M. triceps brachii | Olecranon process of the | Extension of elbow, |
| | ulna | shoulder, and arm |

^{*}The common extensor and flexor origins consist of multiple muscle attachments.

Collection of Ordinal Data

All seven entheseal sites were bilaterally located on each individual, with each site containing eight separate scoreable features following the corrections made to the Coimbra Method after the original publication by Henderson et al. (2013). The updated method by the same authors in 2016 included an additional transitional feature of "textural change" to help collapse bone formation from four possible scores down to three. With eight features per site, and fourteen total sites observed per individual, a total of 112 observations were recorded per individual.

The method split each entheseal site into two different zones, the first adhering strictly to the margin of the entheseal site, with the second zone encompassing the remainder of the entheseal site. The only exception was the *m. triceps brachii*, which the authors suggested scoring on overall appearance only and not dividing into two zones (Speith, 2017). Six categories were used to rank the different zones: bone formation (BF), erosion (E), fine porosity (FP), macro-porosity (MP), textural change (TC), and cavitation (C) (Henderson et al., 2016). Bone formation and erosion were the only two categories recorded in both zones, while the other four were only recorded from zone 2 (Table 4). Each feature was assigned a ranked number from zero to one or two based on a specialized ordinal system, with zero representing little to no expression and the maximum number dependent on the feature being measured (Appendix B).

Table 4. Methodology of Scoring

| Entheseal Site | Zone (if applicable) | Scoreable Feature |
|-----------------------|----------------------|------------------------|
| M. subscapularis | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| CEO | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| CFO | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| M. supraspinatus | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| M. infraspinatus | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| M. biceps brachii | Zone 1 | BF1, E1 |
| | Zone 2 | BF2, E2, FP, MP, TC, C |
| M. triceps brachii | | BF, E, FP, MP, TC, C |

All fifty individuals were scored over the span of several weeks after careful examination of publications pertaining to the Coimbra Method as well as personal guidance from Dr. Nivien Speith, who is a co-speaker for the international working group of 'Occupational Markers' from the original workshop on musculoskeletal stress markers held at the University of Coimbra in Portugal in 2009. All scoring that required measuring features in millimeters were measured using sliding calipers. All scores were also taken without the use of magnification tools in a laboratory setting with no closeup artificial lighting, as per suggestion from Henderson et al. (2016).

To calculate intra-observer error, full sets of ordinal data for the first five individuals and the final five individuals were scored four weeks after initial observations. This was done to calculate initial errors while observing the ECs, as well as to assess whether any experience with the method altered the resulting scores. Specific percentage agreements for both the first five and the last five individuals were calculated for each entheseal site overall, as well as for each independent feature scored.

Variety in Expression

Since each entheseal site was differently shaped and unique in size, most features had a significant amount of variation in their expression. Certain features were never prominent enough to be scored at various levels, especially those in the *m. triceps* entheseal site. However, general trends were photographed using a Canon® EOS 80D camera with an EF-S 18-135mm lens. Resulting photographs in Appendix C depict a selection of typical stages seen in various entheseal sites used in this study. Each photograph shows only one or two features at a time in a single entheseal site to avoid confusion.

Statistical Methods

Following the methods recommended used by Nikita (2014), this study used Generalized Estimating Equations (GEE) to explore the interactions and impacts each variable had on the entheseal changes. Although Generalized Linear Models (GLM) can and have been used to analyze ECs with multiple variables (Michopoulou et al., 2017), Nikita (2014) found that GEE statistically performed better than GLM when both sides of each entheseal site were included in the interactions. Other previous studies such as Villotte et al. (2010) also used GEE when examining entheseal sites in relation to activity levels. Since both the left and right sides of each entheseal site were scored for this study, GEE was used instead of the GLM. All tests were run in RStudio version 1.1.442 using the geeglm() function in the package titled "geepack" version 1.2-1.

The explanatory variables included in this study (Table 5) consisted of elements included in previous studies as well elements unique to this study. All information was self-reported by either the individual prior to death or their next of kin after death. Each

variable was treated as an ordinal response, following Nikita's (2014) methodology. All were assigned simple codes given to either indicate one or the other or a different stage or level. All variables were tested independently with side included in each formula.

Occupation and sex were also tested together to assess the different groups from Table 2.

Table 5. Explanatory Variables

| Variables | Available Scores (n) | | | | |
|-------------------|----------------------------|--|--|--|--|
| Occumation | 0=Manual (n=21) | | | | |
| Occupation | 1=Nonmanual (n=29) | | | | |
| | 0=16-35 (n=3) | | | | |
| Age | 1= 36-55 (n=18) | | | | |
| | 2=56-70 (n=29) | | | | |
| Sex | 0=Male (n=34) | | | | |
| Sex | 1=Female (n=16) | | | | |
| | 0=Lower/Middle (n=16) | | | | |
| SES | 1=Middle (n=23) | | | | |
| | 2=Middle/High (n=9) | | | | |
| | 00=Nonmanual/Male (n=13) | | | | |
| Occupation Sour | 01=Nonmanual/Female (n=16) | | | | |
| Occupation Sex | 10=Manual/Male (n=21) | | | | |
| | 11=Manual/Female (n=0) | | | | |

III. RESULTS

Table 6 shows resulting frequencies for all scores examined on the humerus, as well as the minimum and maximum scores observed. Table 7 shows the same information for the two remaining entheseal sites examined on the lower forearm. Certain features such as cavitation (C) and textural change (TC) were seen less frequently than other features such as bone formation (BF) or fine porosity (FP). Similarly, certain entheseal sites showed less variation in their observed ECs than others. The common flexor origin site, for example, showed little deviation from zero except for BF scores in both zones. The *m. triceps* origin site in particular showed very few changes in almost all features apart from consistent scores for BF. Both the left and right sides of all entheseal sites showed similar minimum and maximum scores, with most of the higher maximum scores showing on the right side.

Intra-observer Error

Intra-observer error was calculated through percentage agreement between two different sessions of data collection for the first five and the final five individuals (Table 8). The lowest feature score seen in any entheseal site was 60% agreement with bone formation 2 (BF2) in the *m. subscapularis* with the first five individuals scored. Most scores showed improvement with time when comparing the first five individuals' scores versus the final five, with the main exceptions of fine-porosity and macro-porosity having higher scores in the first five individuals scored. Macro-porosity (MP), cavitation (C) and textural change (TC) had very high, consistent percentage agreements. As a whole, the final five individuals had higher overall percentage agreements than the first five with the sole exception of the *m. infraspinatus* entheseal site showing an overall decrease of 1%.

Table 6. Descriptive Statistics for ECs on Humerus (n=50)

| Enthesis | Feature | M | in | M | ax | | | % Free | quency | | |
|-----------------|-----------|---|----|---|----|------|------|--------|--------|------|------|
| | | L | R | L | R | | L | | | R | |
| | | | | | | 0 | 1 | 2 | 0 | 1 | 2 |
| Subscap | BF1 | 0 | 0 | 2 | 2 | 44.0 | 50.0 | 6.0 | 30.0 | 64.0 | 6.0 |
| | E1 | 0 | 0 | 2 | 2 | 60.0 | 38.0 | 2.0 | 60.0 | 36.0 | 4.0 |
| | BF2 | 0 | 0 | 2 | 2 | 34.0 | 62.0 | 4.0 | 26.0 | 68.0 | 6.0 |
| | E2 | 0 | 0 | 2 | 2 | 48.0 | 46.0 | 6.0 | 40.0 | 58.0 | 2.0 |
| | FP | 0 | 0 | 1 | 2 | 78.0 | 22.0 | | 74.0 | 24.0 | 2.0 |
| | MP | 0 | 0 | 2 | 1 | 70.0 | 28.0 | 2.0 | 78.0 | 22.0 | |
| | C | 0 | 0 | 2 | 2 | 94.0 | 4.0 | 2.0 | 92.0 | 6.0 | 2.0 |
| | TC | 0 | 0 | 1 | 0 | 98.0 | 2.0 | | 100 | | |
| CEO | BF1 | 0 | 0 | 2 | 2 | 10.0 | 76.0 | 14.0 | 12.0 | 62.0 | 26.0 |
| | E1 | 0 | 0 | 1 | 2 | 86.0 | 14.0 | | 72.0 | 26.0 | 2.0 |
| | BF2 | 0 | 0 | 2 | 2 | 34.0 | 64.0 | 2.0 | 40.0 | 58.0 | 2.0 |
| | E2 | 0 | 0 | 1 | 2 | 56.0 | 44.0 | | 46.0 | 48.0 | 6.0 |
| | FP | 0 | 0 | 2 | 2 | 70.0 | 28.0 | 2.0 | 68.0 | 30.0 | 2.0 |
| | MP | 0 | 0 | 1 | 2 | 94.0 | 6.0 | | 88.0 | 10.0 | 2.0 |
| | C | 0 | 0 | 0 | 1 | 100 | | | 98.0 | 2.0 | |
| | TC | 0 | 0 | 0 | 0 | 100 | | | 100 | | |
| CFO | BF1 | 0 | 0 | 2 | 2 | 58.0 | 36.0 | 6.0 | 48.0 | 50.0 | 2.0 |
| | E1 | 0 | 0 | 1 | 2 | 94.0 | 6.0 | | 92.0 | 6.0 | 2.0 |
| | BF2 | 0 | 0 | 2 | 2 | 66.0 | 32.0 | 2.0 | 62.0 | 36.0 | 2.0 |
| | E2 | 0 | 0 | 2 | 2 | 84.0 | 14.0 | 2.0 | 84.0 | 14.0 | 2.0 |
| | FP | 0 | 0 | 1 | 1 | 90.0 | 10.0 | | 92.0 | 8.0 | |
| | MP | 0 | 0 | 1 | 0 | 98.0 | 2.0 | | 100 | | |
| | C | 0 | 0 | 0 | 0 | 100 | | | 100 | | |
| | TC | 0 | 0 | 0 | 0 | 100 | | | 100 | | |
| Supra | BF1 | 0 | 0 | 2 | 2 | 70.0 | 24.0 | 6.0 | 64.0 | 30.0 | 6.0 |
| | E1 | 0 | 0 | 2 | 2 | 78.0 | 18.0 | 4.0 | 74.0 | 22.0 | 4.0 |
| | BF2 | 0 | 0 | 1 | 2 | 66.0 | 34.0 | | 62.0 | 24.0 | 14.0 |
| | E2 | 0 | 0 | 2 | 2 | 58.0 | 24.0 | 18.0 | 74.0 | 18.0 | 8.0 |
| | FP | 0 | 0 | 2 | 2 | 82.0 | 16.0 | 2.0 | 68.0 | 30.0 | 2.0 |
| | MP | 0 | 0 | 2 | 2 | 80.0 | 18.0 | 2.0 | 86.0 | 12.0 | 2.0 |
| | C | 0 | 0 | 2 | 2 | 88.0 | 8.0 | 4.0 | 90.0 | 8.0 | 2.0 |
| | TC | 0 | 0 | 0 | 1 | 100 | | | 98.0 | 2.0 | |
| Infra | BF1 | 0 | 0 | 2 | 2 | 60.0 | 36.0 | 4.0 | 48.0 | 50.0 | 2.0 |
| | E1 | 0 | 0 | 1 | 2 | 90.0 | 10.0 | | 88.0 | 10.0 | 2.0 |
| | BF2 | 0 | 0 | 1 | 1 | 70.0 | 30.0 | | 66.0 | 34.0 | |
| | E2 | 0 | 0 | 2 | 1 | 68.0 | 24.0 | 8.0 | 76.0 | 24.0 | 2.0 |
| | FP | 0 | 0 | 1 | 2 | 84.0 | 16.0 | | 80.0 | 18.0 | 2.0 |
| | MP | 0 | 0 | 1 | 2 | 90.0 | 10.0 | 4.0 | 82.0 | 16.0 | 2.0 |
| | C | 0 | 0 | 2 | 2 | 92.0 | 4.0 | 4.0 | 88.0 | 10.0 | 2.0 |
| | TC | 0 | 0 | 1 | 0 | 98.0 | 2.0 | | 100 | | |

Table 7. Descriptive Statistics for ECs on Radius and Ulna (n=50)

| Enthesis | Feature | M | in | M | ax | | | % Free | quency | | |
|----------|--------------|---|----|---|----|------|------|--------|--------|------|------|
| | | L | R | L | R | | L | | | R | |
| | | | | | | 0 | 1 | 2 | 0 | 1 | 2 |
| Biceps | BF1 | 0 | 0 | 2 | 2 | 32.0 | 52.0 | 16.0 | 22.0 | 66.0 | 12.0 |
| | E1 | 0 | 0 | 2 | 2 | 88.0 | 10.0 | 2.0 | 90.0 | 6.0 | 4.0 |
| | BF2 | 0 | 0 | 2 | 2 | 66.0 | 32.0 | 2.0 | 50.0 | 48.0 | 2.0 |
| | E2 | 0 | 0 | 2 | 2 | 88.0 | 10.0 | 2.0 | 64.0 | 34.0 | 2.0 |
| | FP | 0 | 0 | 1 | 1 | 72.0 | 28.0 | | 80.0 | 20.0 | |
| | MP | 0 | 0 | 1 | 1 | 96.0 | 4.0 | | 94.0 | 6.0 | |
| | C | 0 | 0 | 0 | 1 | 100 | | | 98.0 | 2.0 | |
| | TC | 0 | 0 | 1 | 1 | 98.0 | 2.0 | | 98.0 | 2.0 | |
| Triceps | BF | 0 | 0 | 2 | 2 | 44.0 | 52.0 | 4.0 | 36.0 | 58.0 | 6.0 |
| | \mathbf{E} | 0 | 0 | 1 | 2 | 90.0 | 10.0 | | 92.0 | 6.0 | 2.0 |
| | FP | 0 | 0 | 0 | 1 | 100 | | | 96.0 | 4.0 | |
| | MP | 0 | 0 | 0 | 1 | 100 | | | 98.0 | 2.0 | |
| | C | 0 | 0 | 0 | 0 | 100 | | | 100 | | |
| | TC | 0 | 0 | 0 | 0 | 100 | | | 100 | | |

Table 8. Intra-observer Error Percentage Agreements

| | Err | BF1 | E1% | BF2 | E2% | FP% | MP% | С% | TC | Overall |
|---------------|-----------------|-----|-----|-----|-----|-----|-----|-----|-----|---------|
| | | % | | % | | | | | % | |
| Subscapularis | 1^{st} | 70 | 80 | 60 | 80 | 90 | 100 | 100 | 100 | 85 |
| | 2^{nd} | 100 | 80 | 80 | 90 | 90 | 90 | 100 | 100 | 91 |
| CEO | 1 st | 80 | 80 | 90 | 90 | 70 | 90 | 100 | 100 | 88 |
| | 2^{nd} | 80 | 100 | 80 | 90 | 100 | 100 | 100 | 100 | 94 |
| CFO | 1 st | 70 | 100 | 90 | 90 | 90 | 100 | 100 | 100 | 92.5 |
| | 2^{nd} | 90 | 90 | 100 | 90 | 90 | 100 | 100 | 100 | 95 |
| Supraspinatus | 1 st | 70 | 90 | 90 | 90 | 80 | 100 | 100 | 100 | 90 |
| | 2^{nd} | 80 | 90 | 100 | 90 | 80 | 100 | 90 | 100 | 91 |
| Infraspinatus | 1 st | 70 | 90 | 80 | 80 | 100 | 100 | 100 | 90 | 89 |
| | 2^{nd} | 100 | 90 | 60 | 80 | 90 | 90 | 90 | 100 | 88 |
| Biceps | 1 st | 100 | 90 | 90 | 70 | 100 | 100 | 100 | 100 | 94 |
| | 2^{nd} | 80 | 100 | 100 | 90 | 90 | 100 | 100 | 100 | 95 |
| Triceps | 1 st | 80 | 100 | N/A | N/A | 100 | 100 | 100 | 100 | 97 |
| | 2^{nd} | 90 | 100 | N/A | N/A | 100 | 100 | 100 | 100 | 98 |

Generalized Estimating Equations (GEE)

Tables 9-15 show the resulting p-values from the GEE tests run using the geeglm() function within RStudio. Side was also included as a variable for all five tests to ensure the full spectrum of the ECs seen in each individual was included in the calculations. Almost all EC showed a lack of correlation with occupational type and activity level in comparison to the other explanatory variables included in the statistical model.

Each entheseal site highlighted different explanatory variables, although the results were inconsistent. The *m. subscapularis* entheseal site showed more correlation to socio-economic status (SES) than any other variable (Table 9). However, when all scores were summarized and broken down into the different SES levels, the middle status proved to have the highest average (Figure 4). Age, the next highest variable, showed a general increase of scores over time when summarized scores were compared (Figure 5). However, the p-values indicated that this was mainly seen in only BF1 and C.

Table 9. p-values (Pr(>|W|)) for m. subscapularis

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .508 | .002 | .012 | .909 | .075 |
| E1 | .746 | .906 | .080 | .273 | .911 |
| BF2 | .216 | .288 | .581 | .474 | .024 |
| E2 | .255 | .279 | .592 | .701 | .890 |
| FP | .935 | .371 | .004 | .059 | .042 |
| MP | .205 | .246 | .062 | .356 | .105 |
| C | .531 | .005 | .462 | .901 | .673 |
| TC | .003 | .792 | .666 | .474 | .000 |

^{*}Bolded/Italicized values are significant at α =0.05.

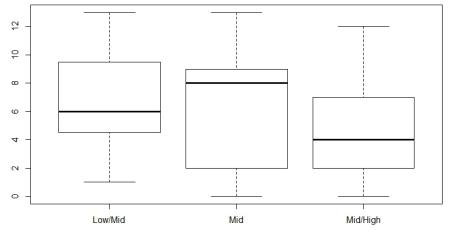


Figure 4. Summary of scores pertaining to SES for M. subscapularis

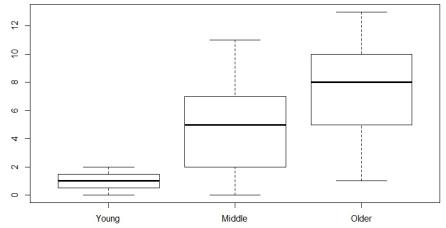


Figure 5. Summary of scores pertaining to AGE for M. subscapularis

The CEO site showed directional correlation to age more than occupational type (Table 10). Figure 6 shows a summary of scores of the CEO site indicating similar results.

Table 10. p-values (Pr(>|W|)) for Common Extensor Origin

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .795 | .004 | .006 | .292 | .150 |
| E1 | .007 | .001 | .143 | .000 | .550 |
| BF2 | .237 | .366 | .049 | .201 | .540 |
| E2 | .086 | .027 | .493 | .856 | .690 |
| FP | .379 | .231 | .155 | .820 | .210 |
| MP | .077 | .398 | .526 | .050 | .800 |
| C | .051 | .102 | .866 | .058 | .460 |
| TC | - | | | | |

^{*}Bolded/Italicized values are significant at α =0.05.

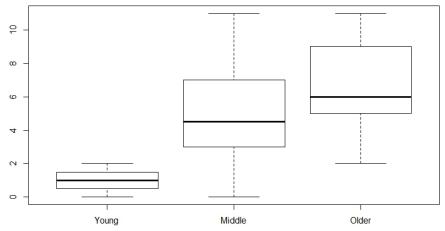


Figure 6. Summary of scores pertaining to AGE for CEO

The CFO site showed some correlation with SES, but both E2 and MP had very low frequencies (Table 6), which potentially skewed the results (Table 11). However, the same results were also reflected in the summarized scores, showing a clear directional distribution with the low/middle SES having the highest average.

Table 11. p-values (Pr(>|W|)) for Common Flexor Origin

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .476 | .150 | .220 | .330 | .962 |
| E1 | .468 | .240 | .600 | .840 | .429 |
| BF2 | .592 | .180 | .490 | 160 | .744 |
| E2 | .836 | .200 | .370 | .660 | .046 |
| FP | .923 | .560 | .760 | .740 | .234 |
| MP | .001 | .110 | .330 | .600 | .005 |
| C | | | | | |
| TC | | | | | |

^{*}Bolded/Italicized values are significant at α =0.05.

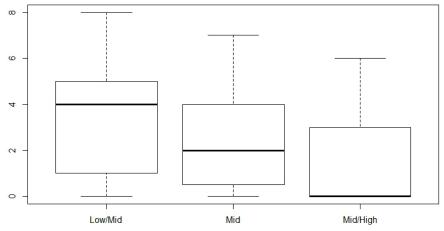


Figure 7. Summary of scores pertaining to SES for CFO

M. supraspinatus appeared to have a strong connection to SES, although the summarized scores showed a nearly opposite directional distribution compared to the results seen in the previous entheseal sites. Although *m. supraspinatus* had the strongest set of p-values indicating significance with SES in general (Table 12), the summarized scores showed that the middle/high level of SES had the highest average, completely opposite of the results found for *m. subscapularis*.

Table 12. p-values (Pr(>|W|)) for m. supraspinatus

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .400 | .345 | .770 | .969 | .022 |
| E1 | .250 | .661 | .164 | .334 | .439 |
| BF2 | .090 | .094 | .143 | .247 | .067 |
| E2 | .150 | .596 | .000 | .114 | .006 |
| FP | .840 | .112 | .353 | .460 | .492 |
| MP | .500 | .277 | .364 | .532 | .018 |
| C | .280 | .898 | .063 | .188 | .316 |
| TC | .000 | .099 | .050 | .000 | .000 |

^{*}Bolded/Italicized values are significant at α =0.05.

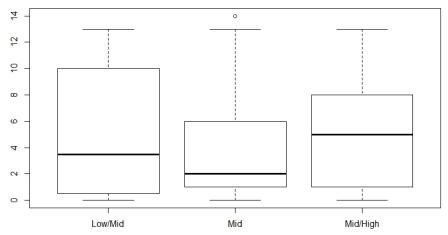


Figure 8. Summary of scores pertaining to SES for M. supraspinatus

M. infraspinatus showed strong correlations with sex in four features (Table 13). The highlighted features also had the lowest scoring EC, with frequencies usually <10% (Table 6). The summarized scores did show females to have the higher scores, however, which is significant given that the males outnumbered the females almost 2:1 (Figure 9).

Table 13. p-values (Pr(>|W|)) for m. infraspinatus

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .820 | .009 | .078 | .387 | .730 |
| E1 | .850 | .842 | .892 | .709 | .486 |
| BF2 | .970 | .792 | .477 | .500 | .510 |
| E2 | .990 | .826 | .005 | .905 | .004 |
| FP | .560 | .954 | .411 | .586 | .249 |
| MP | .400 | .245 | .016 | .018 | .802 |
| C | .730 | .484 | .004 | .274 | .936 |
| TC | .310 | .000 | .000 | .000 | .002 |

^{*}Bolded/Italicized values are significant at α =0.05.

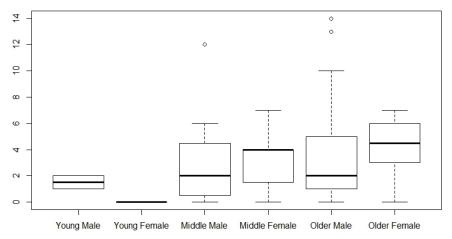


Figure 9. Summary of scores pertaining to SEX and AGE for M. infraspinatus

The *m. biceps brachii* site was the only entheseal site of all seven included in this study to show consistent correlation with both occupation and sex combined (Table 14). However, the summarized scores indicated that the trend for higher entheseal site scores was more often found in nonmanual workers rather than manual workers, a finding that directly disagrees with the hypothesis of activity related entheseal changes (Figure 10). Age was found to have the second highest correlation with the EC, and the summarized scores showed a normal directional distribution with the older individuals displaying the higher average (Figure 11).

Table 14. p-values (Pr(>|W|)) for m. biceps brachii

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF1 | .092 | .042 | .916 | .054 | .800 |
| E1 | .354 | .841 | .119 | .006 | .980 |
| BF2 | .149 | .155 | .041 | .017 | .450 |
| E2 | .035 | .149 | .212 | .019 | .690 |
| FP | .697 | .997 | .061 | .955 | .330 |
| MP | .089 | .090 | .115 | .218 | .910 |
| C | .000 | .000 | .007 | .098 | .580 |
| TC | .679 | .321 | .103 | .074 | .690 |

^{*}Bolded/Italicized values are significant at α =0.05.

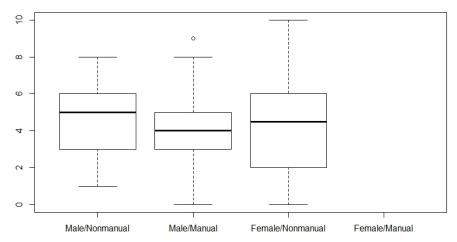


Figure 10. Summary of scores pertaining to OCCUPATION|SEX for M. biceps brachii

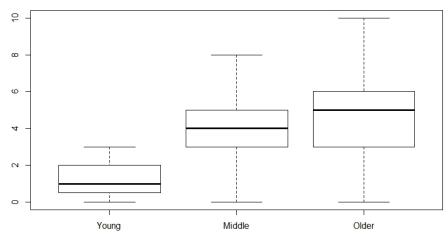


Figure 11. Summary of scores pertaining to AGE for M. biceps brachii

The p-values for *m. triceps brachii* displayed a relatively strong correlation with SES (Table 15). However, the frequencies of scored EC for this site were generally either very low or nonexistent. The summarized scores show a slight directional distribution with the low/middle and middle SES both having the higher average (Figure 12).

Table 15. p-values (Pr(>|W|)) for m. triceps brachii

| Variable | Occupation | Age | Sex | Occ Sex | SES |
|----------|------------|------|------|---------|------|
| BF | .069 | .120 | .017 | .898 | .131 |
| E | .723 | .100 | .746 | .005 | .025 |
| FP | .331 | .270 | .099 | .701 | .003 |
| MP | .952 | .000 | .000 | .000 | .012 |
| C | | | | | |
| TC | | | | | |

^{*}Bolded/Italicized values are significant at α =0.05.

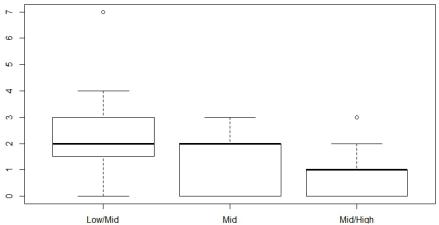


Figure 12. Summary of scores pertaining to SES for M. triceps brachii

Summary of Results

Almost all the entheseal sites displayed a lack of correlation with occupation. The one entheseal site that did display significance was the opposite of what was expected, with nonmanual workers having the higher scored entheseal changes.

Other explanatory variables had mixed results. SES had the highest amount of correlation amongst all the entheseal sites as a whole, but its significance often relied on scores that had low frequencies. Sex did not show much correlation compared to the other variables, but it did have one strong correlation with promising results. Age often showed typical directional distributions that suggested a correlation between older individuals and higher scores, but due to the low number of younger individuals available for this study, much of the summarized score distributions simply mirrored the overall distribution seen throughout the sample population. A general synopsis of all the variables and their respective correlations can be found in Table 16.

These results overall showed unorganized patterns amongst the entheseal sites. Some EC appeared more successful in showcasing specific variables while others tended to show more of an equal correlation between more than one variable. Some features also appeared more frequently than others (Tables 6&7). Overall, the results were inconsistent

with previous findings that emphasized occupation and suggest that other explanatory variables may be more impactful than previously considered.

Table 16. Summary of Correlations

| Enthesis | Strong | Moderate | Weak | No Correlation |
|--------------------|------------------|-----------------------------|----------------------------|----------------|
| | Correlation | Correlation | Correlation | |
| M. subscapularis | SES | Age, Sex | Occupation | Occupation Sex |
| CEO | Age | Sex, Occupation Sex | Occupation | SES |
| CFO | | SES | Occupation | Age, Sex, SES |
| M. supraspinatus | (SES) | Sex | Occupation, Occupation Sex | Age |
| M. infraspinatus | (Sex) | Age, SES, Occupation Sex | | Occupation |
| M. biceps brachii | (Occupation Sex) | Age, Occupation | | Sex, SES |
| M. triceps brachii | SES | Sex, Occupation Sex | Age | Occupation |

^{*}Parentheses indicate unexpected results.

IV. DISCUSSION AND CONCLUSION

Research Question #1: Activity Levels and Occupational Type

This study attempted to apply a primarily bioarchaeological method to a temporally different population, assuming that by doing so, the differences in occupation and other variables would not affect the performance of the method as a whole and potentially in a forensic anthropological context. The results were inconclusive regarding activity levels in association with entheseal changes as a whole, but the *m. biceps brachii* attachment site showed surprising results. Although the initial report came back showing correlation to occupation and sex combined, the actual groups showing the higher numbers belonged to the nonmanual laborers. Even females, who have statistically been shown in the past to be smaller than men in both enthesis size and change (Weiss, 2003), working nonmanual labor jobs had more entheseal changes than male manual laborers. Many studies using the Coimbra Method have commented on the muted correlation that activity seems to have with entheseal sites compared to other variables. This study's findings also indicated that certain entheseal sites might show unexpected results when compared to occupational categorization.

Although several studies have used entheseal sites to analyze activity patterns in general, most have examined the ECs using bioarchaeological samples, whose occupations are rather different from a modern collection (Hawkey & Merbs, 1995; Villotte et al., 2010; Henderson et al., 2013; Weiss, 2015). Occupations seen in the archaeological record are more diversified in their movements because of the many different responsibilities an individual may have during a regular work day, as opposed to modern day occupations being more specialized in their work responsibilities due to

larger numbers in the workforce and overall technological innovation. Archaeological populations worked to survive each day by gathering, hunting, harvesting, or trading in various levels of social organization, and although certain distinctions have been found when examining the concept of sexual division of labor (Ogilvie & Hilton, 2011), modern workers are in general more able to conserve energy for extracurricular activities. Thus, the activities inside and outside the workplace are vastly different than they were hundreds of years ago.

This study was able to demonstrate that occupational type did not overall correlate with entheseal changes in the TXSTDSC population as strongly as the other variables included in the equations did, which is not completely uncommon in entheseal site studies (Cardoso & Henderson, 2010; Michopoulou et al., 2017). However, there are some limitations that are difficult to avoid when conducting studies done on activity-related changes and must be addressed. Although careful steps were taken while analyzing the different occupations in this study, labor is generally difficult to categorize, and some studies have merely focused on the potential bias that could be introduced simply by using manual and nonmanual labor as variables (Cardoso & Henderson, 2013). Other factors such as duration, intensity, and postural demands of the workload are important in understanding expected activity levels in a job, and these details are often not given in self-reported occupations in documented collections (Jurmain et al., 2011).

Self-reported occupations are arguably more accurate than bioarchaeological records, but occupation at death only provides a snapshot of the entire life history of an individual and is based entirely on the individual's (or next of kin's) opinion of what he or she does. Cardosa and Henderson (2013) also pointed out that occupation at death does

not account for any job changes the individual might have had during his or her lifetime, which would have the potential to significantly alter results. Thus, despite the precautions taken in selecting the various individuals with their listed occupations, the natural limitations inherent in this study could have led to the understated role of occupation and its related activity in entheseal changes.

The results (or lack thereof) regarding the impact of occupational category and overall correlation with other variables could also have modern implications as well. Since workers in modern societies generally have more time on their hands than their bioarchaeological ancestors due to a mandated maximum eight-hour workday, habitual activities (non-work related) could have affected the entheseal sites analyzed in this particular study, such as outdoor activities or exercise. Due to a lack of information available, active hobbies or habits were not considered in this study, and both of those factors could have easily affected the activity levels of an individual and potentially impacted the resulting ECs as well.

Research Question #2: Other Explanatory Variables

The effects of the explanatory variables in this study were varied depending on the entheseal site. Out of all the variables included, SES was the most significant in general. Since information regarding SES is not always available for bioarchaeological collections, this suggests that different socio-economic situations may cause differential entheseal use and wear. However, the reported SES for this particular sample were all based on childhood socio-economic conditions, which further complicates the matter. Entheseal changes have been shown to mostly manifest between the ages of 35-55, especially regarding the changes associated with labor (Villotte et al., 2010). The

reported SES data does not involve those later years. However, socio-economic mobility is not impossible in the United States when compared to other countries: data taken from the 1980-1982 birth cohort showed that a child born into the lowest income percentile was still half as likely as a child born in the upper percentile to become middle class or higher (Laurin, Engstrom, & Alec, 2019). However, the discrepancy in the United States is still rather large. A child born in the poorest percentile is three times more likely to remain in that percentile as an adult than children born in the richest percentile (Laurin et al, 2019). Given that this sample from the TXSTDSC does not necessarily reflect the general SES seen in America between 1999-2004 (Braveman et al., 2010), as was discussed previously, the issue regarding childhood SES status and any correlation with entheseal changes remains unclear. If these individuals had little mobility in social class and remained in their reported socio-economic statuses through adulthood, this potentially suggests that outside activity seen more commonly in lower to middle socioeconomic statuses plays a bigger role in entheseal changes than was previously thought. However, even though SES was significant for most of the EC, it also showed no significance for three out of the seven entheseal sites, suggesting that there was no single trend that could be observed in this sample.

Age did appear to affect various entheseal sites and their associated changes, but due to an unfortunate lack of younger individuals available for this study, this could be a result of low numbers. However, almost all of the age distributions by entheseal site showed the older population having higher average summarized scores. Since the rate of enthesopathies has been shown to slow after around 50 years of age, these results suggest

that age does play a role in entheseal changes, which supports previous findings (Dutour, 1992; Villotte et al., 2010; Henderson et al., 2013).

Sex did not correlate to many of the entheseal sites individually, apart from *m*. *infraspinatus*; however, females did show more overall changes than males. Given that males heavily outnumbered the females in this study, and there were no manual female workers included, these results suggest that the modern females in the TXSTDSC are potentially more active than the men when outside of their day jobs. Although all females that were listed as housewives were excluded from this study due to ambiguous descriptions of workload, some of the females included might have been working mothers, resulting in an unknown amount of work in their home as well as in their occupation. These circumstances would possibly have increased the rate of their entheseal changes.

Ultimately, all variables were shown to be significant at certain points, but overall consistency was lacking. The main takeaway seems to be a word of caution: entheseal changes are complex and are caused by a multitude of factors, all of which should be addressed in the research as interacting variables. In terms of utilizing this method for potential forensic anthropological analysis of occupational type, examining entheseal changes involves multiple variables, and based on the results from this particular study, it seems unwise at this point in time to assume that occupation can be successfully predicted using the Coimbra Method. However, this is not necessarily because the method itself is lacking in accuracy; due to changes seen in variables such as SES, this study suggests that modern populations and their activity levels are not solely contained and aptly described in their occupational classifications. Other factors such as outside

hobbies or activities that are not job related could play significant roles and thus alter the entheseal changes and their resulting scores.

It should also be noted that although these entheseal changes occurred before death most likely due to a myriad of reasons, individuals are also not equal in their responses to stress and environment (Wood, Milner, Harpending, & Weiss, 1992), and these limitations should always be considered when examining skeletal changes that occurred during life.

Research Question #3: Error and Accuracy

Due to this study having one sole observer collect the data, only intra-observer error was calculated to assess consistency in scoring (Table 7). The results were mainly in the higher percentages, with overall percentages all above the previously reported 80% (Henderson et al., 2016). Certain entheseal sites improved more than others, with only the *m. infraspinatus* showing a negative change when comparing the repeated scores against the first and the final five individuals scored. This potentially could have been caused by difficulties in seeing the outline or margin of the entheseal site next to the *m. supraspinatus*, with which it shares a border.

Certain scores also showed consistent results, though some proved to be more difficult than others. Both cavitation and textural change were rarely observed, but when they were scored, the results were relatively accurate. Bone formation in both zones did indicate that there were some initial difficulties in over-scoring during the first round of data collection, which was to be expected and has been documented before (Henderson et al., 2013). In general, the scores with the highest frequencies (BF, E, and FP) all showed improvement through experience with the method.

Performance of Method

Based on the results seen in this particular study with this sample, the Coimbra Method appears to be appropriate in assessing the severity of the different features of the entheseal sites. However, although repeatability was demonstrated through the low error rates seen in this study, the method itself is rather complicated with instructions and scoring systems specific to each feature. Steps have been taken by the original authors to simplify these instructions to make the method more user-friendly and repeatable, but the method is still fairly complicated, which is understandably difficult to avoid given the complex nature of ECs (Henderson et al., 2016). The authors themselves suggest receiving in-person training from one of the authors themselves to best learn how to observe ECs using the Coimbra Method, such as in a workshop (Henderson et al., 2016). Although they are commendable in their efforts to ensure the method is being applied correctly, that is often not an easy task for new observers. If the method is to become a universal method for analyzing entheseal sites, the authors should use full disclosure in describing the method in both text and illustration in their publications to help other researchers replicate their results and use the method themselves.

More illustrations of the entheseal changes in the Coimbra Method publications would be especially helpful. Although there are photos included in each, the authors indicate multiple elements in each photo, resulting in confusing arrows and unclear distinctions of features involving percentage of coverage on the entheseal site. Textural change does not even have a true representative photograph. Although the authors originally wrote that photographing an entire series on each enthesis is paramount to

improving its reproducibility (Henderson et al., 2013), the updated publications do not have complete series of photographs (Henderson et al., 2016; Henderson et al., 2017).

The authors also articulate that photographs are not equivalent to seeing the ECs in person on actual bone. However, the instructions for the method are difficult to learn, and published photographs for individual scores on the most popular entheseal sites would be a tremendous help in assisting new observers who are unfamiliar with the method. For this particular study, only an abridged collection of photographs was included to showcase the typical examples seen in the TXSTDSC during this study (Appendix C). However, each photograph only represents one score of a feature found on a designated entheseal site in order to avoid confusion.

Conclusion

This study assessed whether a method that examined entheseal changes in bioarchaeological and historical collections could still be used on a modern collection for forensic anthropological purposes (Henderson et al., 2013). Although the Coimbra Method originally showed lower observer scores in its initial introduction, an updated version of the method with condensed scoring methods was introduced in 2016 (Henderson et al., 2016), and the observer agreement rates reached 80% for common entheseal sites. Other studies (Weiss, 2007; Cardoso & Henderson, 2010; Michopoulou et al., 2017) began to analyze the different variables that could potentially affect entheseal changes and found that variables other than activity level had an unsystematic but significant effect on the entheseal changes.

Following the examples of these previous studies, this study examined the relationship between the entheseal changes and variables such as occupation, socio-

economic status, and age in a modern collection at Texas State University to identify possible trends. The results showed that there was once again an inconsistent pattern seen throughout the entheseal sites, with the various sites correlating with different variables, some expected and others unexpected. The results overall showed that all alternate explanatory variables showed higher correlations to the various entheseal sites than the assumed occupation or activity levels.

These results lead to an ultimate and unavoidable conclusion: although systematic in its application and complex in its approach to analyzing entheseal sites, the Coimbra Method may not be the best method to assess occupation classification in a modern collection due to secular changes. Circumstances more commonly seen today may have the potential of affecting the resulting scores. Trends such as hobbies or activities done outside the main occupation could potentially cause an individual to have more severe entheseal changes, despite what their occupational type may be. Other variables such as socio-economic status, which are not always documented for bioarchaeological populations, could potentially play a role in entheseal changes according to the multiple correlations seen in this study. Degenerative changes that come with age (the average of which has been steadily increasing with time) most likely mask or at least affect the entheseal changes based on past results from other studies as well as those seen in this study. From all appearances, it appears that entheseal sites are indeed mosaics affected by a multitude of factors and should be analyzed as such (Kennedy, 1998).

Finally, the Coimbra Method as a whole has proven to be an adept method in scoring the individual features of an entheseal site, but its complexity makes repeatability difficult, especially for new observers who have not had specific training from the

authors of the method themselves. The photographs included in the original and subsequent publications could be much improved by showcasing the different stages of the entheseal changes per entheseal site for each individual feature. Although there are photos showing generic entheseal changes, the publications specifically detailing the Coimbra Method do not include a series of photographs showing a progression of intensity or severity of the entheseal changes. Detailed photographs with only one or two features with a single score per photograph could greatly reduce observer error in general and potentially improve the method's accuracy in assessing activity levels and occupational type, as well as better estimate the full impact of other explanatory variables.

APPENDIX SECTION

Appendix A. Full List of TXSTDSC Individuals

| ID | SEX | AGE | SES | OCCUPATION | CLASSIFICATION |
|---------|--------|-----|-----------|-----------------------------|----------------|
| 01-2009 | Male | 49 | Middle | Mechanic | Manual |
| 08-2009 | Female | 53 | Middle | Education Consultant | Non-manual |
| 04-2010 | Female | 53 | Middle | Teacher | Non-manual |
| 08-2010 | Male | 67 | Lower | Steelworker | Manual |
| 09-2010 | Male | 63 | Middle | Aerospace Sys. Analyst | Non-manual |
| 10-2010 | Male | 32 | Low/Mid | Handyman | Manual |
| 12-2010 | Male | 54 | Middle | Tire Technician | Manual |
| 15-2010 | Male | 64 | Middle | Carpenter | Manual |
| 08-2011 | Male | 53 | Upper/Mid | Mechanic | Manual |
| 09-2011 | Female | 54 | Middle | Child Health Worker | Non-manual |
| 12-2011 | Female | 53 | Low/Mid | Medical Transcription | Non-manual |
| 19-2011 | Male | 56 | Low/Mid | Carpenter | Manual |
| 21-2011 | Female | 56 | Middle | Real Estate Agent | Non-manual |
| 06-2012 | Male | 58 | Middle | Construction Worker | Manual |
| 18-2012 | Male | 59 | Middle | Attorney | Non-manual |
| 19-2012 | Male | 18 | Middle | Student | Non-manual |
| 21-2012 | Male | 42 | Upper/Mid | Real Estate Agent | Non-manual |
| 27-2012 | Female | 58 | Upper/Mid | Computer Technician | Non-manual |
| 35-2012 | Female | 63 | Middle | Clerk | Non-manual |
| 36-2012 | Female | 42 | Low/Mid | Executive Assistant | Non-manual |
| 37-2012 | Male | 49 | Unknown | Minister | Non-manual |
| 38-2012 | Male | 50 | Low/Mid | Mechanic | Manual |
| 39-2012 | Male | 57 | Middle | Architect | Non-manual |
| 45-2012 | Male | 65 | Low/Mid | Mechanic | Manual |
| 48-2012 | Female | 64 | Middle | Teacher | Non-manual |
| 05-2013 | Male | 54 | Middle | Handyman | Manual |
| 06-2013 | Male | 68 | Low/Mid | Architect | Non-manual |
| 13-2013 | Male | 69 | Low/Mid | Carpenter | Manual |
| 15-2013 | Female | 55 | Upper/Mid | Office Administrator | Non-manual |
| 16-2013 | Male | 53 | Middle | Architect | Non-manual |
| 25-2013 | Male | 62 | Upper/Mid | Insurance Agent | Non-manual |
| 27-2013 | Male | 69 | Upper | Lawyer/Judge | Non-manual |
| 53-2013 | Male | 65 | Lower | Construction Worker | Manual |
| 55-2013 | Male | 57 | Low/Mid | Lumberjack | Manual |
| 57-2013 | Male | 54 | Lower | Factory Worker | Manual |
| 59-2013 | Male | 58 | Low/Mid | Electrical Engineer | Manual |
| 65-2013 | Male | 61 | Low/Mid | Handyman | Manual |
| 12-2014 | Female | 64 | Middle | Urban Planner | Non-manual |
| 21-2014 | Female | 23 | Upper/Mid | Student | Non-manual |

| Appendi | x A continue | ed. | | | |
|---------|--------------|-----|-----------|-----------------------|------------|
| | | | | Administrative | |
| 30-2014 | Female | 66 | Unknown | Assistant | Non-manual |
| 48-2014 | Male | 52 | Middle | Electrician | Manual |
| 49-2014 | Male | 56 | Middle | School Chair Director | Non-manual |
| 56-2014 | Female | 69 | Lower | Real Estate Agent | Non-manual |
| 57-2014 | Male | 59 | Middle | Mason | Manual |
| 60-2014 | Male | 59 | Low/Mid | Mechanic | Manual |
| 65-2014 | Male | 43 | Upper/Mid | Construction Worker | Manual |
| 14-2015 | Male | 70 | Middle | Computer Technician | Non-manual |
| 41-2015 | Female | 57 | Middle | Secretary | Non-manual |
| 60-2015 | Female | 49 | Middle | Teacher | Non-manual |
| 68-2015 | Male | 62 | Upper/Mid | Financial Officer | Non-manual |

Appendix B. Coimbra Method as Detailed by Henderson et al. in 2016

| Zone | Feature | Abbr. | Definition | Degrees of Expression |
|--------|----------------|-------|---|--|
| Zone 1 | Bone Formation | BF1 | See degrees of expression. Normal morphological smooth-rounded or mound-like (check by touching) margins, even if the margin is elevated, should be scored as 0. | 1 = distinct sharp demarcated new bone formation along the margin or other enthesophyte which does not meet the criteria for stage 2 in terms of size or extent 2 = distinct sharp demarcated new bone formation along the margin or other enthesophyte ≥1 mm in elevation and ≥50% of margin affected by new bone formation |
| | Erosion | E1 | Depressions or excavations of any shape and involving discontinuity of the floor of the lesion greater in width than depth with irregular margins. Only erosions >1 mm, where you can clearly see the floor, were recorded. This does not include pores (i.e. rounded margins). Score erosions if they occur on bone formation | 1=<25% of margin 2=≥25% of margin |
| Zone 2 | Bone Formation | BF2 | Any bone production from roughness of surface to true exostoses (e.g. distinct bone projections of any form, like bony spurs, bony nodules and amorphous bone formation). | 1 = distinct bone formation >1 mm in size in any direction and affecting 1 mm in size in any direction and affecting ≥50% of surface 2 = distinct bone formation >1 mm in size in any direction and affecting ≥50% of surface |
| | Erosion | E2 | Depressions or excavations of any shape (but not covered by the definition of macro-porosity) and involving discontinuity of the floor of the lesion greater in width than depth with irregular margins. Only erosions >2 mm were recorded. MPO or FPO occurring within an erosion should not be recorded separately. Bone formation is only scored if it exceeds the height of the depression (do not score woven bone). Score erosions if they occur on bone formation. | 1 =<25% of surface 2=≥25% of surface,50%e |

| Appendix B continued. | | | | | | | |
|-----------------------|-----------------|----|---|--|--|--|--|
| | Fine Porosity | FP | Small, round to oval perforations with smooth, rounded margins <1 mm. These should be visible to the naked eye and be in a localized area. Do not score if they are at the base of an erosion or if they occur as part of woven bone. | 1=<50% of surface 2=≥50% of surface | | | |
| | Macro Porosity | MP | Small, round to oval perforations with smooth, rounded margins about 1 mm or larger in size with the appearance of a channel, but the internal aspect is rarely visible. Do not score if they are at the base of an erosion. | 1=one or two pores 2=>2 pores | | | |
| | Cavitation | С | Subcortical cavity with a clear floor which is not a channel. The opening should be >2 mm and the whole floor must be visible. | 1=1 cavitation 2=>1 cavitation | | | |
| | Textural Change | TC | A non-smooth, diffuse granular texture (with the appearance of fine-grained sandpaper) | 1=covering >50% of surface | | | |

^{*}Absence of changes should be scored as zero.

Appendix C. Additional Photographs



L. humerus, m. subscapularis: Zone 1, Bone Formation score 0



L. humerus, m. subscapularis: Zone 1, Bone Formation score 1



L. humerus, m. subscapularis: Zone 1, Bone Formation score 1



L. humerus, m. subscapularis: Zone 1, Bone Formation score 1



L. humerus, m. subscapularis: Zone 1, Bone Formation score 2



L. humerus, m. subscapularis: Zone 2, Erosion score 2



R. humerus, m. subscapularis: Fine Porosity score 1



L. humerus, Common Extensor Origin: Zone 1, Bone Formation score 1



R. humerus, m. infraspinatus: Macro-porosity score 2, Cavitation score 2

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