STRESS AND FRAILTY IN MEDIEVAL PRUSSIA: INTERPRETATIONS FROM SKELETAL REMAINS AT BEZŁAWKI

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ABSTRACT

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Health is routinely studied in living populations using quantifiable measurements such as allostatic load and frailty. In recent years, particularly since the introduction of the osteological paradox, there has been increased interest among bioarchaeologists in how these concepts can be applied to the study of health in past populations. Although health is not directly observable in skeletal remains, assessment of frailty can be useful for understanding the implications of long-term exposures to stress on well-being and mortality. This study builds upon past research in this area by incorporating commonly observed indicators of physiological stress, such as dental disease and osteoarthritis, into a cumulative index that can be used to assess frailty in archaeological populations. A sample of 37 individuals (males, n=15; females, n=16; indet., n=6) between the ages of approximately 14 and 65 years from the Late Medieval site of Bezławki in northeastern Poland, were examined for evidence of 13 biomarkers of physiological stress related to nutritional deficiencies, growth disruption, infection, and trauma. These categories were chosen based on their potential to affect the lifestyles of individuals in the past and present. Following examination, each individual was assigned a frailty score, which was then compared across groups within the population. While results indicate no statistically significant variation in frailty between age and sex cohorts, biomarker prevalence reflects a population experiencing complex environmental change and social reform following a long period of colonization and conversion. Ongoing research will explore the relationship between frailty and lifestyle in Medieval Prussia, an area which currently has sparse historical records.

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1. INTRODUCTION

1.1 Stress and Frailty in Bioarchaeology

In bioarchaeological research, the concept of stress is central to the study of past health (Larsen, 2015). Continued stress over a lifetime can leave physical evidence in skeletal remains and has been shown to affect health, morbidity, and mortality (Marklein et al. 2016). Although health is a concept that is generally understood, it can be difficult to define and even more difficult to quantify, particularly in archaeological contexts (Reitsema and McIlvaine, 2014). Health encompasses several aspects of daily life, including human interactions with the environment, and the ability to effectively accomplish daily tasks (Reitsema and McIlvaine, 2014). The biocultural approach to paleopathology, the study of ancient disease, explores the interaction between the human body and its physical and socio-cultural environments, viewing human variation as the result of differential responses to environmental stimuli (Grauer, 2011).

Although biological stress itself is not directly observable in skeletal remains, the physiological effects of stress on the skeleton are measurable and quantifiable, allowing us to rely upon skeletal stress biomarkers as evidence of past health and disease (Klaus, 2014). In this way, skeletal biomarkers of stress serve as a useful proxy for health in bioarchaeological research (Reitsema and McIlvaine, 2014).

The ability to interpret the effect that stressors have on the human body facilitates our understanding of both past and future health (Marklein et al., 2016). Unfortunately, the relationship between stress and health is complicated. Health is a holistic concept of which physiological stress is only one aspect (Reitsema and McIlvaine, 2014). Recent biological research has expressed the need to incorporate interdisciplinary perspectives into the study of stress in order to facilitate the interpretation of health patterns from skeletal remains (Reitsema and McIlvaine, 2014; Temple and Goodman, 2014). In order to properly understand the physiological mechanisms of stress and disease, bioarchaeologists must incorporate knowledge from other disciplines rather than simply focusing on the prevalence of different skeletal stress indicators (Temple and Goodman, 2014). Pathophysiology, for example, examines the etiology of diseases and the mechanisms by which they develop and manifest as skeletal lesions, which can help us to interpret their broader meanings to anthropology (Klaus, 2014).

In living populations, health is commonly assessed using quantifiable measurements such as allostatic load and frailty. Allostatic load refers to the strain exerted on the physiological systems of the body as it reacts to repeated stressful stimuli (McEwen and Stellar, 1993). Similarly, frailty refers to an increased susceptibility to stressors as the result of cumulative physiological decline over time (Marklein et al., 2016). In recent years, there has been increased interest in how these concepts might be applied to bioarchaeological research. This has been especially true since the publication of the Osteological Paradox, which inspired paleopathologists to reconsider the way in which they interpret data gathered from skeletal samples (Wood et al., 1992). Application of the frailty concept to bioarchaeological research provides a mechanism through which comparisons can be made between living and past populations (Marklein et al., 2016). Advancements towards understanding the components of human frailty allow bioarchaeologists to begin to address issues of variations in frailty among members of past populations.

1.2 Assessing Frailty in Skeletal Remains

In 2016, Kathryn Marklein and colleagues published an article proposing the use of a Skeletal Frailty Index (SFI) that is designed after models of frailty that have been used to evaluate the health of living individuals. This method was created in order to provide researchers with a better way to better understand the overall health of past populations that is reliable and encompasses multiple aspects of health. The proposed index addresses 13 indicators of physiological stress that are commonly observed in human skeletal remains (Marklein et al., 2016). These indicators fall within one of four categories that are representative of different aspects of frailty: 1. growth disruption, 2. nutritional deficiency and infection, 3. activity, and 4. trauma. The index is used to score each individual based on the presence or absence of stress indicators in order to obtain a cumulative level of frailty for each person (Marklein et al., 2016) Individual frailty can then be compared across age and sex groups within the population to get a better overall understanding of population health. The methodology outlined by Marklein and colleagues in 2016 is an index of skeletal frailty that takes a multidisciplinary approach to the assessment of physiological stress markers that has been a much-needed addition to the field in recent years (Wood et al. 1992; Klaus 2014; Temple and Goodman 2014; Reitsema and McIlvaine 2014). This approach provides additional context for the interpretation of skeletal biomarkers of stress and has the potential to shed new light on the relationship between stress, health, and lifestyle by facilitating comparisons between contemporary and historical population groups. The research project being introduced here follows the methodology outlined by Marklein and colleagues in order to obtain a multidisciplinary perspective on physiological stress in Medieval Prussia.

1.3 The Research Sample

The primary goal of this project is to explore patterns of stress and frailty observed in the population represented by the human skeletal remains excavated from the Bezławki archaeology site, a Late-Medieval period (c. mid 1300- mid 1400) cemetery located in north-eastern Poland (Figure 1). This study does not aim to diagnose specific diseases, but rather to conduct an overall assessment of variation in patterns of stress and frailty between different age and sex cohorts within the Bezławki population. Finally, this research aims to make connections between frailty and lifestyle in Medieval Prussia, an area which currently has sparse historical records.

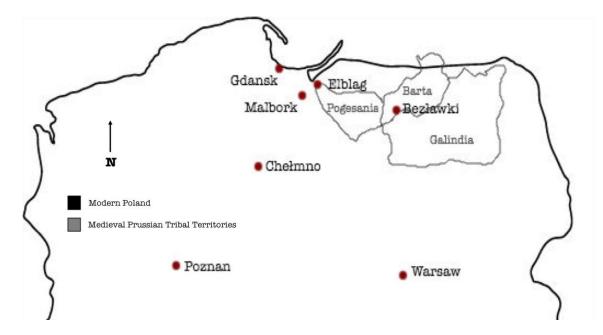


Figure 1: Map of Medieval Prussia, after Pluskowski (2013).

The sample included in this study is composed of 37 individuals between the ages of 14 and 65 years. Individuals were chosen from the larger Bezławki sample on the basis of preservation and representation of the necessary elements for analysis of frailty, as outlined in Chapter two of this thesis. A comprehensive macroscopic analysis of skeletal remains was conducted on each individual included in the study sample to assess growth disruption, nutritional status, pathological conditions, and trauma patterns. Individuals were then assigned a cumulative frailty score based upon the presence or absence of these factors.

1.4 Expected Outcomes and Hypotheses

The Bezławki research site is representative of an era of complex social and environmental reform surrounding the colonization of formerly Prussian lands by the Teutonic Order in the late 13th- mid 14th century (Pluskowski, 2013). During this period of history, the Prussian people were experiencing an influx of crusaders and settlers, largely from Germany and Poland, which resulted in a dramatic increase in population in the area by the beginning of the 14th century (Brown and Pluskowski, 2011). The Prussian Crusades were driven heavily by Christianization of the pagan Prussian tribes, and pagan traditions were outlawed following the establishment of the Teutonic Order in the region (Pluskowski, 2013). These factors, couples with changing agricultural practices, suggest an increase in environmental and social stressors that would likely have influenced the people of Medieval Bezławki. This research project explores the possibility that differences in frailty will be observed among age and sex cohorts within the Bezławki population due to these colonial pressures.

Among living populations, frailty is considered to increase with age, resulting in decreased resistance to stressful stimuli, increased susceptibility to disease, and higher mortality (Fried et al., 2001; Fulop et al., 2010). It is therefore hypothesized that analysis of the Bezławki research sample will reveal correlations between frailty and individual age-at-death. Previous bioarchaeological research has revealed correlations between prevalence of stress biomarkers and age-at-death, providing further support for this hypothesis (DeWitte, 2008; DeWitte and Bekvalac, 2010). In addition, it is expected that

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variations in frailty between sex cohorts will be observed within this population group. This hypothesis is based in previous clinical and bioarchaeological research studies, which suggest differential frailty between males and females in the populations studied (DeWitte et al., 2010; Fried et al., 2001; Yang, 2009).

1.5 Organization of Thesis

This thesis will be organized into five chapters. Chapter two will discuss literature relevant to this research that will familiarize the reader with the use of frailty in bioarchaeological and epidemiological research and with the unique history of medieval Prussia. Chapter three will further define the study sample, explore the methods used in this research study, and will outline differential diagnostic criteria used in this analysis. Chapter four details the results of data analysis. Chapter five is a discussion of the results and will conclude the thesis by exploring potential for future research and a summary of the project's relevance to applied biological anthropology.

REVIEW OF THE LITERATURE

2.1 The Complicated Nature of Health

The study of physiological stress is essential in bioarchaeology because of the complex nature of health. The definition of health asserts that it encompasses aspects of mental and social, as well as physical well-being (World Health Organization, 2001). It is a holistic concept in that it refers to factors relating to quality of life, the ability to complete daily tasks and interaction with society (Reitsema and McIlvaine, 2014). Individual health is influenced by the interaction of environmental, cultural, and physiological variables that are not always readily apparent in bioarchaeological assemblages (Marklein et al., 2016). Assessment of many of these factors, such as personal perceptions of well-being, require direct communication with individuals (Marklein et al., 2016). While these important components of human health prove difficult to quantify in clinical studies on living populations, they prove to be even more challenging to accurately interpret when working with archaeological samples (Reitsema and McIlvaine, 2014). In this sense, it is not possible to reliably interpret the health of past population groups from the skeletal remains examined in bioarchaeological research samples (Waldron, 2009).

While expressions of health that are commonly observed in clinical studies typically remain elusive to paleopathologists, certain biological changes associated with stress are readily apparent in human remains. Continued stress over a lifetime leads to chronic wear on the physiological systems of the body, which can affect its functional capacity over time (McEwen and Stellar, 1993). The disruption that occurs in response to stressful stimuli has the potential to leave behind quantifiable evidence in skeletal remains, making stress a useful proxy for health in bioarchaeological research (Reitsema and McIlvaine, 2014).

Modern paleopathology explores the relationships between human societies and pathological conditions in the past, and the influences that these interactions have had on human adaptation (Grauer, 2011). Paleopathology today is a multidisciplinary field that incorporates knowledge from subjects such as human biology and epidemiology, among others. If we are to properly interpret health by proxy of skeletal biomarkers of stress, this collaboration is essential in that it allows for a better understanding of the physiological and socio-cultural mechanisms of stress (Reitsema and McIlvaine, 2014; Temple and Goodman, 2014). The remainder of this chapter will explore definitions of stress, frailty, and allostatic load, and their applications to paleopathological research.

2.2 The Osteological Paradox

One of the greatest changes to paleopathological research came with the publication of the Osteological Paradox (Wood et al. 1992). The Osteological Paradox called attention to some inherent issues in paleopathology and bioarchaeology that affect the way that we interpret data collected from human skeletal remains. Wood et al. (1992) suggest that the interpretation of skeletal data is more complicated than originally thought. Issues such as demographic non-stationarity, selective mortality, and hidden heterogeneity in frailty complicate analysis and, according to the authors, must be accounted for in order to obtain reliable data. An understanding of these factors is essential to effectively analyzing patterns of stress in archaeological samples. These issues complicate analyses of skeletal assemblages in that they prevent the direct association of health and mortality from measures of average life expectancy and lesion frequencies (DeWitte and Stojanowski, 2015).

The first of these, demographic non-stationarity, refers to the idea that populations experience changes in population demographics over time, and that these changes are particularly susceptible to changing fertility rates (Wood et al., 1992). Most populations are non-stationary, meaning that their age distribution fluctuates as migration occurs or the population grows (Wood et al., 1992). Due to the fact that fertility has such a large effect on the age-at-death distributions observed in skeletal assemblages, Wood and colleagues assert that these measurements may be observations of changes in fertility rather than mortality (Wood et al., 1992).

Selective mortality refers to the idea that skeletal assemblages do not adequately represent population health because they are comprised of deceased individuals (Wood et al., 1992). While this observation may seem somewhat obvious, selective mortality is a significant issue in paleopathological research. The sample populations under study in bioarchaeological research are composed only of those individuals who died at any given age (Wood et al., 1992). This results in sample populations that are highly selective for skeletal biomarkers of stress that increase mortality risk, creating biased data that should,

in theory, overestimate true prevalence values of lesions within the sample population (Wood et al, 1992).

Finally, individuals vary in their susceptibility to death and disease as the result of different exposures and experiences over their lifetimes (DeWitte and Stojanowski, 2015). Differences in susceptibility to death and disease are referred to as heterogeneity in frailty (Wood et al., 1992). For example, an individual who has experienced trauma may be at higher risk of developing an infection or other complication than an individual who has not. Differential frailty makes it difficult to interpret individual risk of death from the population-level mortality patterns observed in bioarchaeological research, complicating comparisons between populations (Wood et al., 1992; DeWitte and Stojanowski, 2015). Wood and colleagues emphasize the need to better understand the causes of frailty and the relationship between frailty and mortality in order to combat the issues that arise in bioarchaeological interpretations of skeletal samples (Wood et al., 1992; DeWitte and Stojanowski, 2015). This would allow for models to be created that would estimate heterogeneity in frailty in past populations (DeWitte and Stojanowski, 2015). Additionally, they express the importance of understanding the underlying etiologies of skeletal biomarkers of stress, and variations in their expression (DeWitte and Stojanowski, 2015). In other words, skeletal biomarkers should be interpreted with regard to how they affect survival, rather than simply comparing prevalence values of lesions (Temple and Goodman, 2014).

Arguments against the Osteological Paradox have been made by researchers since its publication. Goodman (1993) suggests that Wood and colleagues ignore the contributions of culture in the development of skeletal lesions and misrepresent biological processes. However, it has also received an outpouring of support from other researchers in the field and has inspired a movement towards better approaches to evaluating skeletal remains (Grauer, 2011). Methods such as life-history studies and hazard models have been proposed and implemented within bioarchaeological research as a means of overcoming the issues outlined in the Osteological Paradox (Marklein et al., 2016).

2.3 Stress

2.3.1 What is Stress?

Stress is defined as the physiological changes that take place within the body in response to external stimuli (Larsen, 2015; Reitsema and McIlvaine, 2014). It is useful to think of stress as a continuous internal process in which the body responds to potentially harmful stimuli rather than as a single, isolated event (Marklein et al., 2016). The body attempts to respond to external stimuli by using various resources in an attempt to mitigate stress and maintain a stable internal environment. Stress is often viewed as a disruption to homeostasis, as external stimuli lead to responses that impact the capacity of the body to maintain stability (McEwen and Stellar, 1993; Larsen, 2015). It has been proposed that *allostasis* may be a better term for use in this area because the human body is rarely in a state of equilibrium (McEwen and Stellar, 1993). Rather, it is in a constant state of fluctuation in response to external stimuli (Sterling, 2004). Allostasis refers to the

body's ability to adapt to new challenges by altering its vital functions in such a way that it restores its internal environment to a new state of stability, different from before (McEwen and Stellar, 1993). Stress introduces the need to adapt the operating range of bodily functions, placing strain on certain systems in favor of others (McEwen and Stellar, 1993). If the body is unable to resist the multifactorial causes of stress, a physiological response can occur (Temple and Goodman, 2014).

The human body reacts to stress by initiating a series of adaptive responses that vary in their degree of permanence depending upon the duration of the environmental change experienced. (Kuzawa and Thayer, 2011). Acute stress is typically managed effectively by homeostatic systems, which work to maintain internal stability by altering physiological and behavioral systems (Kuzawa and Thayer, 2011). Chronic exposure to stress represents a more complicated issue. Homeostatic systems require energy reallocation, and typically cannot be sustained long enough to effectively manage longterm environmental or social changes (Kuzawa and Thayer, 2011). Although the body is able to adapt to stressful stimuli, a strain is placed on bodily systems over time (McEwen and Stellar, 1993). In periods of stress, the body must work harder in order to initiate a physiological response (McEwen and Stellar, 1993). This process of repeated strain building on the body as it continues to respond to repeated insults over time is called allostatic load (McEwen and Stellar, 1993). When stress is sustained over long periods of time and cannot be effectively managed by adaptive responses, the result may be disruptions to functional capacity in the form of diminished health, work capacity, or fertility, which can be detrimental to both the individual and their society (Temple and

Goodman, 2014). Furthermore, these disruptions to functional capacity may lead to biological or behavioral changes that can then initiate future episodes of stress.

To understand the mechanisms by which stress affects an individual, we must first investigate its primary causes. Stress occurs as the result of complex interactions that take place between environmental, cultural, and biological factors (Goodman et al., 1984; Larsen, 2015). The relationship between these components determine the nature of the stressor produced and the physiological response that will occur as a result (Larsen, 2015). The following sections present definitions for each of these categories, and present evidence for their importance in the development of physiological responses to stress.

2.3.2 Environmental Constraints

The environmental context in which individuals live in large part determines their susceptibility to infectious diseases and access to important nutrients (Piperata et al., 2014). Two categories of environmental constraints have been described, including limitation of available resources and direct stressors (Goodman, 1984). Humans rely upon their environment to provide resources that are essential to their survival, such as food and water. When environmental constraints such as extreme drought or food shortages lead to limited availability of essential resources, individuals find it more difficult to obtain the nutrients they need to survive, and the body initiates a response. Humans may also artificially create similar conditions by limiting access to resources for specific groups of people. An example of artificial environmental constraints is the famine caused when a combination of German blockades and freezing temperatures

limited the delivery of resources to the Netherlands during the Dutch Hunger Winter of 1944-1945 (Stein et al., 2004). This period of history is commonly cited as an example of the effect that malnutrition in early life can have on human development (Gowland, 2015)

An example of the consequences that limitations to resources can have on the development of stress in individuals and populations can be seen in the study on subadult remains from the Great Irish Famine of the mid-1800's (Gerber, 2014). The famine came as the result of a devastating disease that eradicated the region's potato crop, destroying the population's primary source of food. During this period, Ireland lost nearly a quarter of its population due to starvation and emigration (Gerber, 2014). A sample of 545 individuals excavated from the mass graves at the Kilkenny Workhouse were examined for evidence of physiological stress. The Kilkenny Workhouse served as a location for victims of the famine to take refuge but had ultimately seen overwhelming mortality in its inhabitants (Gerber, 2014). Results of the study indicate that children who perished during the height of the famine saw high prevalence of Harris lines (transverse lines that can be observed at the metaphyses that indicate periods of stress that occurred during growth) and growth retardation, suggesting significant periods of physiological stress (Gerber, 2014).

Additional sources of environmental stress come directly from ecological variables such as extreme fluctuations in temperature, severe weather patterns, or physiogeography (Larsen, 2015; Temple and Goodman, 2014). Direct environmental stressors may influence patterns of behavior and lead to changes in social structure. For example, several recent studies have investigated the relationship between extreme environmental conditions and interpersonal violence. Mares and Moffett (2016) analyzed data from 57 countries and discovered a positive correlation between rising temperatures and homicide rates (Mares and Moffett, 2016). A similar study was conducted on an archaeological sample population from San Pedro de Atacama, located in northern Chile (Torres-Rouff and Costa Junquiera, 2005). This particular population was living during the pre-Columbian occupation, a time of social and environmental change (Torres-Rouff and Costa Junquiera, 2005). A sample of 682 crania spanning four distinct periods of the area's history were analyzed for evidence of trauma (Torres-Rouff and Costa Junquiera, 2005). Results of this study indicate a positive association between resource availability and trauma prevalence. Changes were particularly apparent in the Late Intermediate period (AD 950-1400) population, whose members would have experienced a series of major droughts (Torres-Rouff and Costa Junquiera, 2005). Results of both of these studies indicate that periods of environmental stress are associated with increases in interpersonal violence, suggesting that bioarchaeologists might infer environmental deficiencies through evidence of trauma in skeletal remains.

2.3.3 Cultural Systems

When considering the underlying causes of stress in human populations, we must also consider cultural systems. In many of the examples of interpersonal violence in relation to resource availability presented above, researchers suggest that the effects of environmental stressors may have been exacerbated by social and economic inequality in the regions involved in the studies (Torres-Rouff and Costa Junquiera, 2005; Mares and Moffett, 2016). Mares and Moffet note that changing rates of youth unemployment are typically related to increased homicide rates (Mares and Moffett, 2016). In the case of the Irish Famine, the effects of institutionalization were considered as potential complicating factors of an already stressful situation (Gerber, 2014). Society and culture are important components of the larger human environment. While our cultural systems may exacerbate stressors, as in the case of the Irish Famine, they also have the potential to buffer the effects of environmental constraints (Goodman et al., 1984).

Take, for example, the differential anemia risk observed between participants in the Mexican Family Life Survey, as described in the 2014 study conducted by Piperata and colleagues. Results of this study indicate that children under the age of five and women of reproductive age are at a significantly higher risk of developing anemia than men from the same population, a trend that can be observed in other cultures as well (Piperata et al., 2014). Children, for example, are biologically at a higher risk of developing anemia due to a combination of factors; their small body size affects the amount of micronutrients that they are able to ingest, which is in direct conflict with their relatively high need to absorb these nutrients (Piperata et al., 2014). Additionally, young children are undergoing a period of dietary changes following weaning, which not only affects their immune system but also exposes them to potentially novel pathogens (Piperata et al., 2014). These factors, coupled with sociocultural practices, influence a child's risk of developing anemia. In some cultures, children are not prioritized at mealtimes, thus further decreasing the amount of nutrient-rich foods that they are able to intake and increasing their susceptibility to anemia (Piperata et al., 2014). In other parts of the world, women tend to decrease their own intake of nutrient-rich foods in times of nutritional stress in favor of ensuring that children receive the resources that they need to survive (Piperata et al., 2014). In this sense, cultural practices either buffer or exacerbate the effects of environmental constraints on population groups.

Cultural factors can also create situations in which increased stress is likely to occur (Goodman et al., 1984). For example, changing socio-political conditions in the Central Andes of Peru following Spanish colonization have been linked to increased physiological stress in an indigenous Mochica population from Mórrope (Klaus and Tam, 2009). This is evidenced by an increased prevalence of several non-specific indicators of stress, including porotic hyperostosis, periosteal new bone formation, decreased femoral growth velocity, and decreased female fertility in the post-contact population (Klaus and Tam, 2009). The authors suggest that Spanish colonization of the area led to an increase in population density in the area which would have contributed to poor sanitation and contamination of the water supply which would have in turn increased pathogen exposure (Klaus and Tam, 2009). These factors, coupled with stressful environmental conditions including water shortages, contributed to an increase in physiological stress (Klaus and Tam, 2009).

2.3.4 Host Resistance

Individuals vary greatly in both their biological and behavioral responses to stressful stimuli (McEwen and Stellar, 1993). This variation in the capacity to resist

stressors introduced by either the environment or cultural systems is referred to as host resistance (Goodman et al., 1984). Individuals within a population will, by nature, vary in their susceptibility to death and disease (Wood et al., 1992). This is a key component of evolutionary processes that shape life on Earth. An individual's genotype in part determines the range within which they are capable of responding to environmental insults (Gluckman, 2004), but genetic differences alone are insufficient to explain the variation expressed by humans in reaction to changes in the environment (McEwen and Stellar, 1993). Developmental plasticity, or the ability of the genome to exhibit a range of potential phenotypes in response to information received during early developmental periods, plays a key role in an individual's ability to respond to stress (Kuzawa and Thayer, 2011).

A variety of factors play important roles in determining individual host resistance. For example, developmental instability and exposure to stressors in early life can have significant impacts on the future well-being of individuals (Larsen, 2015). Signals received from the mother during particularly sensitive periods of growth and development influence the expression of phenotypes by relaying important information about the environment to their offspring (Kuzawa and Thayer, 2011). Growing evidence has suggested that environmental insults in the early stages of development can have substantial influence on susceptibility to pathological conditions in later life (Gillman, 2005). Continuing research in the field of epigenetics suggests that the biological effects of early environmental stress may also be passed down to the next generation of offspring (Kuzawa and Thayer, 2011). Furthermore, continued exposure to stress over the course of a lifetime may increase an individual's susceptibility to future health risks. Essentially, individuals will exhibit unique variations in frailty as a result of their age, sex, or life experience (Wood et al, 1992). This is an important concept to consider when interpreting patterns of stress from human skeletal remains. Individuals within population groups vary in their ability to combat stressful stimuli and therefore differ in their risk of morbidity and mortality. This idea was introduced above as *hidden heterogeneity*, a key component of the Osteological Paradox that is essential to understanding the study of stress in bioarchaeological research.

2.3.5 Physiological Stress Biomarkers

Paleopathologists are somewhat limited in their analysis of stress in the past because many causes of stressors leave no physical evidence in skeletal remains (Ortner, 2011). Additionally, skeletal and dental tissues are limited in the ways that they are able to respond to physiological stress (Brickley and Ives, 2008). Abnormalities that can be observed in the skeleton fall into one of five categories, each of which can occur either individually or in combination with others (Ortner, 2011). These categories include abnormal formation, destruction, density, size, or shape of bone (Ortner, 2011). At the microscopic level, these changes involve either the resorption or deposition of osteons, or an alternation of these two processes (Goodman et al., 1988). While many diseases leave characteristic diagnostic changes in skeletal tissues, many leave only generalized changes attributable to several etiologies, so called non-specific stress indicators (Goodman et al., 1988). Nonetheless, abnormal alterations to skeletal and dental tissues suggest the occurrence of some unusual stimulus (such as infection) regardless of ability to recognize a specific cause (Goodman et al., 1988).

While stress itself is not directly observable in skeletal remains, the physiological changes that it causes in bodily tissues are (Klaus, 2014). If individuals are unable to resist the effects of harmful external stimuli, then a disruption to biological systems is likely to occur, often resulting in physical changes to the body (Goodman and Armelagos, 1989). Bone is generally slower to respond to the effects of stress than soft tissues, so the presence of skeletal biomarkers indicate stressful stimuli that were either severe or present for a long period of time (Goodman et al., 1988). Chronic exposure to stressors exert a strain on biological systems and make it increasingly difficult for the body to function (McEwen and Stellar, 1993). This lends additional context to the interpretation of skeletal lesions and emphasizes the concept of stress as a continuous process.

While skeletal and dental biomarkers of stress may indicate the presence of pathological conditions in an individual, the effect that these conditions had on the individual's ability to function within society is less apparent (Ortner, 2011). This is where paleopathology benefits from collaboration with disciplines such as epidemiology and human biology (Klaus, 2014). The model of physiological stress proposed by Goodman and Armelagos in 1989 recognizes that, while skeletal biomarkers are representations of stress in the body, they are not the direct cause of functional impairment (Temple and Goodman, 2014). There are underlying causes to the lesions that can be observed in skeletal remains that are important to consider in our analysis of stress in past populations.

Analyses are further complicated by the fact that stressors tend to interact with each other in complicated ways, making analysis of the relationship between the environment and individuals somewhat difficult (Goodman et al., 1988). The use of multiple indicators of stress is common, as health is widely recognized as a complex phenomenon that incorporates aspects of nutrition and disease, among other factors (Larsen, 2015). The multiple indicator approach allows researchers to formulate a more holistic understanding of stress in the past (Larsen, 2015).

2.3.6 Stress in Bioarchaeological Research

From an evolutionary perspective, evidence of physiological stress in skeletal remains can be viewed as evidence of either human adaptation or maladaptation to harmful environmental circumstances (Grauer, 2011). Prolonged or severe periods of physiological stress can result in functional impairment that affects an individual's ability to interact effectively with their environment and society (Larsen, 2015). The biocultural perspective views stress as a product of the interaction between humans and their social and biological environments (Grauer, 2011). While skeletal biomarkers of stress are observed and diagnosed at the individual level, they reveal important insights into the ways in which society responds to these situations (Goodman et al., 1988). Tilley and Oxenham present an excellent example of this in their analysis of an individual with likely juvenile-onset quadriplegia from Neolithic Vietnam (Tilley and Oxenham, 2011). The individual involved in this study would have required a high level of care from his community due to his impairment. Evidence of survival to adult age, despite significant limitations, suggests that others were willing to provide advanced levels of care for the man (Tilley and Oxenham 2011). In this sense, we are able to learn a significant amount about the values and lifestyles of past populations by studying the health of a single individual.

The primary goal of stress research is to speak to the underlying interactions between biology, behavior, society, and the environment, rather than simply the description of lesion prevalence (Klaus, 2014). In recent years, increased collaboration with the fields of human biology, epidemiology, and pathophysiology have greatly increased our understanding of the effect that stress has on the human skeleton and the implications that this may have on health and well-being (Klaus, 2014). Interdisciplinary methods for assessing the effects of stress continue to be developed and refined. Measurements such as frailty and allostatic load are commonly used to assess levels of stress and health in living population groups and are beginning to find a place in bioarchaeological research (Marklein et al., 2016). The interpretation of physiological stress from skeletal remains is an important component of bioarchaeological research. Interdisciplinary approaches help to shed light on etiology of stress biomarkers and allow researchers to make broader connections between populations past and present (Marklein et al., 2016).

2.4 Frailty

2.4.1 Definition of Frailty

Epidemiological definitions of frailty refer to it as a state of decreased resistance to stressors as the result of progressive physiological decline (Fried et al., 2001). Factors commonly used to diagnose frailty in clinical research include musculoskeletal function, aerobic capacity, cognitive and integrative neurological function, and nutritional reserve (Campbell and Buchner, 1997). They may also include such factors as lean body mass, balance, and typical level of activity (Fried et al., 2001). These components of frailty reflect an individual's ability to interact effectively with their environment, which is key to the concept of frailty (Campbell and Buchner, 1997). If an individual is unable to effectively interact with their environment, then their level of frailty is higher than it would be if they were able to successfully perform these tasks (Campbell and Buchner, 1997).

Research has shown that frailty serves as a useful predictor of adverse outcomes such as falls, hospitalizations, impairment, and death in elderly clinical patients (Fried et al., 2001). Strong associations between frailty and several major chronic diseases, such as cardiovascular and pulmonary disease, have been observed as well (Fried et al., 2001). In these studies, frailty was higher when two or more pathological conditions were present (Fried et al., 2001). This study also found that women had a higher likelihood of frailty than men of the same age. The authors hypothesize that this may be due to women having lower lean-body mass and strength on average than men of the same age, making it easier for them to cross the frailty threshold (Fried et al., 2001).

2.4.2 Frailty in Bioarchaeology

Individual variation in frailty is a key argument made in the Osteological Paradox as to why health cannot be accurately assessed from skeletal remains (Wood et al., 1992). By working towards an understanding of the components that influence the development of frailty we begin to understand why some individuals might be more susceptible to stress than other members of their population group. Factors that are typically used to assess frailty in epidemiological research are generally not directly observable in skeletal remains. However, certain skeletal biomarkers of stress may be associated with frailty indicators that are utilized in clinical research (Marklein et al., 2016). This facilitates comparisons between past and present human groups and encourages collaboration between numerous fields of study in further understanding human frailty.

2.4.3 Frailty in Recent Literature

The concept of frailty has been central to several recent studies within bioarchaeology. Sharon DeWitte, for example, has incorporated measures of frailty into her work researching the Black Death in medieval European populations, particularly London. In 2008, DeWitte and Wood published an article exploring the possibility that the Black Death may not have killed indiscriminately, as has been commonly assumed. Results of the study indicated that the epidemic was selective in that it was more likely to affect individuals with pre-existing health conditions. Although the Black Death cemetery populations are a unique case in regard to their extremely virulent circumstances, this study nonetheless highlights the importance of considering underlying variation in frailty when interpreting mortality in bioarchaeological contexts (DeWitte and Wood, 2008).

Subsequent studies conducted by DeWitte and colleagues have explored potential underlying components of differential frailty. One study investigated the relationship between stature and increased mortality, revealing that the correlation between these two factors makes stature an effective biomarker of physiological stress (DeWitte and Hughes-Morey, 2010). Another study revealed differences in frailty between males and females from the East Smithfield Black Death Cemetery in London, suggesting that sex may be an important risk factor in the development of frailty (DeWitte, 2010). These studies are particularly important because all individuals involved in the study samples were victims of the Black Death, therefore eliminating any complicating variables that may have come from differences in cause of death. This resulted in data that is highly reflective of mortality risk associated with stress and frailty (DeWitte, 2010).

A study by Piperata and colleagues in 2014 explored the potential use of data collected from living sample population in further understanding the relationship between the environment and human health and well-being. The results of this study call into question the ability of bioarchaeologists to employ the use of skeletal biomarkers of stress (in this instance, cribra orbitalia and porotic hyperostosis) to infer larger conclusions regarding the environmental and social conditions in which individuals lived in the past (Piperata et al., 2014). The study by Piperata and colleagues (2014) also challenged the assumption that skeletal biomarkers of stress have noticeable impacts on individual perceptions of well-being and quality of life. This was an important contribution to the field because this particular component of human health is not directly observable in skeletal remains (Piperata et al., 2014).

The researchers initially hypothesized that individuals who shared identical environmental contexts (shared a household) would be at similar risk for developing anemia and that socioeconomic status would be an important predictive factor in determining anemia risk (Piperata et al., 2014). These are common assumptions in bioarchaeological research. However, although results of the study indicate that these hypotheses could both be supported, overall association between these factors was rather weak (Piperata et al., 2014). This particular study emphasizes the importance of incorporating measures of individual frailty into bioarchaeological analyses, despite the difficulty in doing so (Piperata et al., 2014). Piperata and colleagues argue that the concept of frailty should be expanded to include the larger social and cultural dynamics that can exacerbate biological risk factors (Piperata et al., 2014). From this project, we see how variations in frailty may begin to be understood through the use of biocultural approaches to the study of physiological stress (Piperata et al., 2014). Additionally, this study emphasizes the need to incorporate data from studies on living human populations in an attempt to understand differences in frailty in the past (Piperata et al., 2014).

2.5 The Prussian Crusades

An understanding of the biocultural impact of the stressors experienced by members of the Bezławki population must begin with an examination of the historical context of the region. Prior to the beginning of the 13th century, the region of Prussia was inhabited by 11 distinct tribal groups with a prominent military culture (Pluskowksi, 2013). Their religion was characterized by the worship of natural resources, such as groves of trees, bodies of water, and numerous animals (Gimbutus, 1963). Although much has yet to be discovered about the people of pre-Christian Prussia, archaeological evidence is beginning to piece together certain aspects of their society (Pluskowski, 2013). A significant amount of this information comes from Prussian burial sites, as treatment of the dead was an important component of Prussian culture (Pluskowski, 2013). Cemetery sites are relatively rare in the tribal regions of Warmia, Barta, Sasna, and Galindia as compared with Sambia and Natangia, possibly due to their smaller population densities (Pluskowski, 2013). The Bezławki research site involved in this study, located in what was once Prussian Barta, is of particular research interest for this reason.

The Medieval period of Prussian history saw significant development in social, economic, and political systems that can be characterized by the introduction of Christianity and changing political structures as a result of increased European influence (Brown and Pluskowski, 2011). From the beginning of the tenth century, Poland had been attempting to colonize neighboring regions in an effort to gain territory, spread

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Christianity, and establish churches (Pluskowski, 2013). Beginning in 1230, the Teutonic knights initiated a series of crusades on the Prussians that would continue over the next fifty years (Pluskowski, 2013). These wars would come to be known collectively as the Prussian Crusade (Pluskowski, 2013).

Crusades were wars that were officially sanctioned by the Pope in defense of Christianity and those who practice it (Pluskowski, 2013). While knights from across the continent came to participate in the Prussian Crusades, the Teutonic Order was the driving force behind the transformation of Prussia into a Christianized European state (Pluskowski, 2013).

2.5.1 1100-1200

The first official crusade on the southern Baltic was officially sanctioned by the Pope in 1147 (Pluskowski, 2013). This war, unlike others, began with the primary intention of converting pagan inhabitants of the area to Christianity (Pluskowski, 2013). At the time, there was little interest from the Pope in conversion, and so this initial crusading effort largely failed (Pluskowski, 2013). However, this effort was the first of many wars to against pagan societies in the region to follow (Pluskowski, 2013). In the following years, Polish and Danish leadership would launch military expeditions under the guise of crusades, although they were not officially sanctioned by the Pope (Pluskowski, 2013). These engagements would be met with Prussian aggression and eventually fail (Pluskowski, 2013).

2.5.2 1200-1300

The thirteenth century saw perhaps the most dramatic change in Prussian landscape and culture than any other period of history. In 1215, Christian, a Cistercian monk, was appointed Bishop of Prussia. His newly established church in the Kulmerland (near modern Cłemno), located in the far south-west region of Prussia, struggled due to tribal aggression and low recruitment, and in 1218 the Pope sanctioned crusades in their defense (Pluskowski, 2013). In his letter, the Pope encouraged crusaders to convert pagans without forcing them into servitude (Pluskowski, 2013). These endeavors were followed by Prussian rebellion and a raid on Pomerania, during which two major Pomeranian settlements were destroyed (Pluskowski, 2013). This series of events would contribute to Bishop Christian's eventual replacement by the Teutonic Order (Pluskowski, 2013).

The Teutonic Order's presence in Prussian society is largely due to Duke Konrad I of Masovia, one of several duchies that made up Poland during this time (Lukowski and Zawadski, 2006). Masovia was located along the border of Prussia and Poland, making it an important strategic location (Pluskowski, 2013). In the early 1220's, civil unrest in the area coupled with a desire to expand his territory led the Duke to request the assistance of the Teutonic Order in stabilizing the northern frontier of Masovia in return for territory within the Kulmerland (Pluskowski, 2013). This early-thirteenth-century call to arms against the Prussians was rooted in a desire to protect Christians living in the Polish-held Kulmerland from the perceived threat from their pagan counterparts (Pluskowski, 2013). The Duke had previously come to the aid of Bishop Christian by forming the Knights of

Dobrin, a small local military order who would eventually be absorbed by the Teutonic Knights (Pluskowski, 2013). The Duke had claims to the Prussian territory because he was their closest Catholic neighbor, and so was eager to revive the crusades which had failed to colonize the region in earlier centuries (Urban, 2003).

In 1226, the Holy Roman Empire granted permission to the Teutonic Knights to crusade in Prussia and bestowed upon them the rights to govern the lands offered by the Duke of Masovia and all other land that would be conquered in Prussia (Pluskowski, 2013). By spring of 1228, a small group of Teutonic Knights gathered on the Masovian frontier and initiated a raid on the Prussian-occupied Kulmerland in an attempt to gather information regarding its land and inhabitants (Pluskowski, 2013). In 1230, the Teutonic Order's requests to crusade in Prussia were officially sanctioned by the Pope (Pluskowski, 2013). This led to increased recruitment for the Prussian crusades and established the leadership of the Order (Pluskowski, 2013). Following the conclusion of the Polish civil war, which occurred as the result of the initial fragmentation of the Polish state (Lukowski and Zawadski, 2006), Polish Dukes joined the crusade and battles began taking place along the river Vistula (Pluskowski, 2013). Local Prussian nobles began to surrender their lands and join the crusades in response to early military successes by the Teutonic Order (Pluskowski, 2013). At this time, the Kulmerland was occupied by both pagan and Christian groups (Pluskowski, 2013). The population was small due to previous Polish invasions, which likely contributed to the Knights' success (Urban, 2003). The Prussian tribes were divided due to infighting and had difficulty uniting

against the influx of crusaders and, by 1232, the Teutonic Knights had occupied the Kulmerland (Pluskowski, 2013).

Following this victory, the armies largely dispersed to crusade in neighboring lands, leaving the Teutonic Order to secure their newly-won territory (Pluskowski, 2013). Groups of German and Polish knights and peasants were invited to colonize Prussia, many travelling from the Holy Roman Empire (Pluskowski, 2013). Native Prussian groups were forced into serfdom and required to perform strenuous agricultural and construction tasks for the Order (Pluskowski, 2013). Local resistance continued against the presence of the Teutonic Order, eventually culminating in an attack on Pomeralia and the abduction of Bishop Christian by the Prussians in 1234 (Pluskowski, 2013). In response, the Pope declared his endorsement of the Teutonic Order's control in the region and consented to launch an additional crusade two years later (Pluskowski, 2013).

In the following decades, the Order would request additional support from the Pope, eventually leading to the initiation of several more crusades in defense of Christian converts against remaining pagan Prussians (Pluskowski, 2013). In 1242, the Prussians gained the alliance of Duke Sventopelk of Pomerania and proceeded to overrun much of Warmia, Natangia, and Barta (Pluskowski, 2013). Prussian rebellion and the Pomeranian War against the Teutonic Order would continue until 1249, when a peace treaty was arranged (Pluskowski, 2013). Under this treaty, pagan Prussians swore to accept the Order's rule and abandon their religious practices in favor of Christianity (Pluskowski, 2013). One final attempt against the Order was made, which resulted in another peace treaty in 1253 (Pluskowski, 2013). By 1255, the Teutonic Order's rule spread east to Sambia, although they had been met with resistance. Sambian nobility swore allegiance to the Order, allowing the initiation of military expeditions into pagan Samogitia, which at the time was associated with Lithuania rather than Prussia (Pluskowski, 2013). A truce was arranged between 1257 and 1259, but upon its expiration, the Samogitians fought back against the Order, inspiring a series of uprisings against native groups in Lithuania (Pluskowski, 2013).

During this time, the Prussians were organizing a unified attack against the Teutonic Order that would come to be known as the Great Prussian Uprising (Pluskowski, 2013). The uprising, beginning in 1260, took place in Sambia, Natangia, Barta, and Warmia (Pluskowski, 2013). The Teutonic Order suffered substantial losses at the hands of the Prussians and, as the rebellion moved deeper into west Prussia, the Pope called for a new crusade (Pluskowski, 2013). Although the initial crusade against the Prussian army was successful, the Order's castles continued to fall (Pluskowski, 2013). Despite serious losses, the Teutonic Order held out until, in 1265, assistance in the form on large armies from Germany arrived (Pluskowski, 2013). In the 1270's, the Order launched a series of raids (Pluskowski, 2013).

In 1271, the Bartians set out to claim Pogesania, an area which remained highly contested (Pluskowski, 2013). Shortly after, an army of Sudovians and Lithuanians travelled to Kulm but failed to penetrate their defenses and the leader of the Bartian army was killed, causing his troops to disperse (Pluskowski, 2013). That winter, the crusades arrived in Natangia and the region was destroyed (Pluskowski, 2013). The uprising came to an end in 1274 and the Order began the process of rebuilding (Pluskowski, 2013). The

last decade of the crusades saw a third, minor uprising that ultimately failed. By 1283, the Prussian Crusades had come to a close and the Teutonic Order held nearly complete control of Prussia (Pluskowski, 2013). They established the castle at Malbork as their headquarters beginning in 1309 (Pluskowski, 2013).

2.5.3 1300-1400

The fourteenth century was characterized by colonization, expansion, and structural development of the Teutonic Order's administration in Prussia (Pluskowski, 2013). During this time, the Order was faced with the loss of key allies in central Europe and growing hostility from Poland, resulting in their increased reliance upon Prussian resources (Pluskowski, 2013).

Beginning in 1304, the Order sent military expeditions for elite members of society into Lithuania, providing young men of high social status the opportunity to impress their peers and setting the stage for crusading as an expression of nobility (Pluskowski, 2013). The primary goal of these attacks was the discouragement of further attacks from Lithuanians on Christian lands and, while some of these raids focused on strategic locations, many were simply destructive in nature (Pluskowski, 2013). As the raids continued and the population of local villages declined, Prussia became separated from southern Livonia by an expanse of wilderness (Pluskowski, 2013). This wilderness came to be an important source of timber for the eastern Prussians but made conquest of Lithuanian lands more difficult for the Order (Pluskowski, 2013).

In 1309, the Order's territory expanded further with the annexation of Gdańsk and Pomeralia (Brown and Pluskowski, 2011). This same year, the Teutonic Order relocated their headquarters to Malbork castle (Pluskowski, 2013). As colonization in Prussia continued to expand under the Teutonic Order, native Prussians were relocated to villages where they were recruited to work in agriculture despite the destruction of many crops that had occurred during the crusades (Pluskowski, 2013). Women and children were removed from villages and boys were sent to Germany to be educated as Christians (Pluskowski, 2013). Meanwhile, small numbers of Polish Knights living in Kulm held lands belonging to the Bishop of Kuyavia, with rights to their natural resources (Pluskowski, 2013). The Prussian territories saw a series of attacks from Lithuania beginning in 1311. This continued until 1370, when a failed attack on Sambia led Vytautas, son of the Grand Prince of Lithuania, to agree to convert to Catholicism in return for support of the Order against his brother (Pluskowski, 2013). This allowed the Crusades to press deeper into Lithuania (Pluskowski, 2013). Shortly thereafter, civil war broke out in Lithuania and military incursions from Prussia intensified in the region (Pluskowski, 2013). In 1398, a peace treaty surrendered Samogitia to the Teutonic Order, although military action would continue into the fifteenth century (Pluskowski, 2013). Following the initiation of peace, colonists were recruited from Germany and Poland to settle the newly won Prussian lands (Pluskowski, 2013).

2.5.4 1400-1500

In 1409, rebellion rose against the Teutonic Order in Samogitia (Pluskowski, 2013). Attempts to secure peace in the region failed, and the combined Polish and Lithuanian forces met the Teutonic Order's army on the battlefield between Tannenberg and Grunwald. The Order suffered substantial losses in the battle, including the death of its Grand Master (Pluskowski, 2013). While neither side emerged victorious, the battle marked the beginnings of the decline of the Teutonic Order's reign in Prussia (Pluskowski, 2013). Eventually, the Order's administrative state began to disintegrate due to a combination of inexperienced officials, disobedient ranks, and decline in financial resources as the result of a lack in targets for crusading (Pluskowski, 2013).

In 1440, a new wave of opposition to the Teutonic Order began with the formation of the Prussian League (Pluskowski, 2013). The League ultimately allied with Poland and launched a series of uprisings against the Order, resulting in the fall of several of the Teutonic Order's major settlements and the beginning of the Thirteen Years War (Pluskowski, 2013). In 1462, a major battle took place between the Teutonic Order and the Prussian League and, by the following year, the Order's army had been completely destroyed (Pluskowski, 2013). A peace treaty was signed in 1466 that required the return of West Prussia and Kulm to Poland along with several castles, including Malbork (Pluskowski, 2013). The Teutonic Order relocated to Königsberg, where they reorganized, and local Prussian governments gained increasing control over their lands (Pluskowski, 2013). A significant reason for the decline of the Teutonic Order in the fifteenth century was likely the disappearance of crusading culture (Pluskowski, 2013). A lack of interest and available targets for crusades resulted in a lack of funding for the Order (Pluskowski, 2013). The Teutonic Order came to Prussia to crusade and so, with the loss of crusade culture in the region, the Order no longer had a purpose there (Pluskowski, 2013).

2.5.5 A Note on Changing Environmental Conditions

The colonization of Prussia, as with other areas of Europe, during the crusades saw an increased tendency of people to alter their environments in response to changing socio-economic needs (Pluskowski et al., 2011). Economic expansion in the region required an increased production of produce and, therefore, an expansion of agriculture (Brown and Pluskowski, 2014). The Prussian crusades had a tremendous impact on the natural environment. Prior to colonization, the religion of Pre-Christian Prussia had emphasized the protection of sacred natural resources, forbidding the cutting of trees from certain groves (Gimbutus, 1963). Following the establishment of the Teutonic Order in Poland, the landscape began to change dramatically (Pluskowski, 2013). Pollen recovered from Baltic peat bogs and lakes have revealed evidence of woodland clearance and increased production of cereal grains (Brown and Pluskowski, 2011; Brown and Pluskowski, 2014). Additionally, in the years following the crusades, population saw a dramatic increase, reaching approximately 220,000 by the 14th century (Brown and Pluskowski, 2011).

2. METHODS

3.1 The Study Sample

This research aims to explore patterns of stress and frailty in an area that currently has sparse historical records. Specifically, this study will explore variations in average frailty scores between age and sex cohorts within the Bezławki population. A sample of 37 individuals (males, n=15; females, n=16; indet., n=6) between the ages of approximately 14 and 65 years from the Bezławki medieval cemetery in northeastern Poland, were examined for evidence of physiological stress and scored according to Marklein et al.'s (2016) Skeletal Frailty Index criteria (Table 1). The Bezławki site, a Late-Medieval period (c. mid AD 1300- mid AD 1400) cemetery located in north-eastern Poland (see map), is representative of an era of complex social and environmental reform surrounding the colonization of formerly Prussian lands by the Teutonic Order in the late 13th century (Pluskowski, 2013). This particular region of Prussia was inhabited by the Barta tribe prior to the arrival of the Teutonic Knights, and is one of few cemeteries from the area that have been discovered and excavated thus far (Pluskowski, 2013).

The cemetery at Bezławki sits atop a hill near the local church. The post-glacial soil is sandy and well-drained, resulting in overall good preservation of burials despite minor bioturbation due to moles and substantial root growth in some areas of the cemetery. To date, approximately 205 (males >13 years, n=29; females>13 years, n=27; indet.>13 years, n=23) individuals have been excavated from the cemetery site at

Bezławki. The Bezławki cemetery site was in use for approximately 100 years,

suggesting that the individuals excavated from the site may represent several generations. Cemetery reuse over time resulted in newer burials often cutting through the older ones, further complicating the analysis of burial chronology. Furthermore, following the arrival of the Teutonic Order, individuals from Germany and Poland were invited to colonize the area. Therefore, the Bezławki cemetery may contain individuals from multiple population groups. At this point, differentiating between the different groups is not possible, although these challenges may be addressed by future research. Instances of both single and multiple inhumations were observed. The burials exhibit syncretism, including both Christian and Pagan burial traditions. Following typical Christian practices, the burials were laid in an east-west orientation and hands were placed such that the right arm crossed the chest and the left crossed the pelvis (Koperkiewicz, 2011). These practices, coupled with the Pagan tradition of a coin placed in the mouth, reflect growing Christian influence in formerly Pagan territories (Koperkiewicz, 2011; Pluskowski, 2013). In a few cases, Teutonic-ware grey pottery was discovered near the hip of individuals (Koperkiewicz, 2011).

Individuals were included this study on the basis of preservation and representation of adequate skeletal elements for the analysis of all biomarkers included in the SFI. Sub-adults were further excluded from analysis if accurate measurements of maximum femoral length and femoral head diameter could not be accurately obtained due to inadequate development of the epiphyses.

3.2 Research Methods

3.2.1 Sex Estimation

Sex was estimated non-metrically through the assessment of features of the skull and pelvis. Pelvic features involved in estimation of sex include: width of subpubic angle, presence of ventral arc, width of greater sciatic notch; width of medial aspect of the ischio-pubic ramus, and presence of subpubic concavity (Phenice, 1969; White et al., 2012). Skull features involved in the estimation of sex include: nuchal crests, mastoid processes, supraorbital margins, glabella, mental eminence, and gonial angle (Acsádi & Nemeskéri, 1970; White et al., 2012).

3.2.2 Age Estimation

The individuals included in this study sample ranged in age from approximately 14 to 65 years. Adult age was estimated based on the morphology of the pubic symphysis (Brooks and Suchey, 1990; Katz and Suchey, 1989) and auricular surface of the pelvis (Lovejoy et al., 1985). Sub-adult age was estimated based upon epiphyseal fusion (Schaefer et al., 2009) and dental eruption (Ubelaker, 1979). Average age estimates were determined based on the cumulative age range produced by the scoring methods. Age categories were then defined based upon the mean age estimates. These categories (<25 years, 25-35 years, 35-45 years, >45 years) were chosen so that ages could be effectively separated by decade, providing a simple means of dividing the sample.

3.3 The Skeletal Frailty Index

The Skeletal Frailty Index (SFI) proposed by Marklein and colleagues (Marklein et al. 2016) is designed after models of frailty that have been used to evaluate the health of living individuals. This particular model incorporates several different biomarkers of skeletal stress that are commonly found in bioarchaeological samples. The SFI recognizes frailty as a condition in living human populations rather than as a measurement of increased mortality, allowing for the acknowledgement of skeletal lesions as indicators of both ongoing physiological stress and survival from past stress events (Marklein et al, 2016). Furthermore, this method seeks to relate these indicators of skeletal stress to aspects of frailty in living people to facilitate comparisons between archaeological and living populations (Marklein et al. 2016). This method was created in order to provide researchers with a way to better understand the overall health of past populations in a reliable way that encompasses multiple aspects of health. The proposed index addresses thirteen indicators of physiological stress that are commonly observed in human skeletal remains. These indicators fall within one of four categories that are representative of different aspects of frailty: growth disruption, nutritional deficiency and infection, activity, and trauma (Marklein et al. 2016). The index is used to score each individual based on the presence or absence of stress indicators in order to obtain a cumulative frailty score for each person. Individual frailty can then be compared across age and sex groups to get a better overall understanding of population health.

3.3.1 Scoring Criteria

The Skeletal Frailty Index provides an overall assessment of frailty for each individual within a population by assigning a score to each individual biomarker being observed. For each biomarker, an individual can receive either a score of "0" or "1", resulting in a maximum possible cumulative frailty score of "13". Biomarkers are quantified relative to frailty risk. The stage at which each biomarker contributes to highest risk of frailty receives a score of "1". For most biomarkers included in this study, scoring is based on "presence" versus "absence". The exception to this is the metric biomarkers: maximum femoral length and femoral head diameter. Scoring criteria for each biomarker are outlined in Table 1. The following sections explain in detail differential diagnosis criteria for each biomarker as well as their etiology. This is included to provide context regarding the relationship of the biomarker to frailty.

Skeletal Stress Biomarker	Scores and	Frailty Score 1
	Measurements	·
Maximum Femoral Length	Lengths in Quartiles	Lowest Quartile
Femoral Head Diameter	Diameters in Quartiles	Lowest Quartile
Linear Enamel Hypoplasia	Present, Absent	Present
Porotic Hyperostosis; Cribra	Present/Active,	Present/Active
Orbitalia	Present/Healing, Absent	
Periostitis/Osteomyelitis	Present, Absent	Present
Dental Disease	Present, Absent	Present
Scurvy	Present, Absent	Present
Neoplasms	Present, Absent	Present
Osteopenia/Osteoporosis	Present, Absent	Present
Osteoarthritis	Present, Absent	Present
Intervertebral Disk Disease	Present, Absent	Present
Rotator Cuff Disorder	Present, Absent	Present
Trauma	Present, Absent	Present

Table 1: Outline of SFI Scoring Criteria (Adapted from Marklein et al., 2016)

3.3.2 Growth Disruption

Stressors that are experienced in early childhood can have a direct effect on future morbidity and mortality outcomes (Gowland, 2015). These stressors often manifest in skeletal and dental remains as disruptions to normal growth patterns (Marklein et al., 2016). For example, short stature and abnormal enamel thickness may be indicative of stressful events that occurred during early life. Although any number of factors can influence patterns of growth in humans, stressful stimuli resulting from nutritional deficiencies and infection are most often cited (Larsen, 2015). To assess patterns of growth disruption, stature and robusticity measurements were taken for each individual within the Bezławki sample. Consistent with the Skeletal Frailty Index outlined by Marklein and colleagues (2016), maximum femoral length was taken as a proxy for stature and femoral head diameter was taken as an indication of skeletal robusticity. All metric data were collected following the standards outlined by Langley and colleagues (2016). Individuals were considered to be at higher risk of frailty if they fell within the lowest quartile of measurements for their age and sex cohorts. Patterns of growth and development vary between population groups (Marklein et al., 2016). Therefore, metric quartiles were determined based upon the distribution of measurements taken for this study sample. Linear enamel hypoplasia (LEH), the sole non-metric biomarker of physiological stress included in the growth disruption category, was scored based on presence versus absence criteria. The presence of linear enamel hypoplasia was considered indicative of higher frailty, which will be discussed in more detail below.

<u>3.3.2a Stature – Maximum Femoral Length</u>

Stature has been shown to be positively correlated with increased risk of mortality (DeWitte and Hughes-Morey, 2012). Studies conducted on living populations have shown a strong correlation between growth disruption during childhood due to adverse environmental conditions and terminal adult stature (Larsen, 2015). Following the methodology outlined above, stature was determined by taking maximum femoral length measurements for each individual within the study sample. For the majority of the sample, maximum femoral length measurements were taken from both the right and left side and averaged. Figure 2 provides an example of maximum femoral length measurements taken for this research. In cases where one femur was unavailable or unable to be measured accurately due to damage or pathology, the remaining side was

measured. Individuals for whom a maximum femoral length measurement could not be accurately determined were excluded from the sample. Stature was compared within sex cohorts to control for variation related to sexual dimorphism, as outlined by Marklein and colleagues (2016). Metric quartiles of maximum femoral length were determined for each age and sex cohort using Excel version 16.10. Individuals of indeterminate sex were considered as a separate group, regardless of age.



Figure 2: Example of Maximum Femoral Length Measurement. Bezławki, Poland. Photo by K. Gaddis (2017).

Short stature is not itself directly indicative of increased frailty. Genetic factors play an important role in the determination of adult stature and should be considered (McEvoy and Visscher, 2009). However, because stature may reflect stress in early life and therefore serves as a useful non-specific indicator of physiological stress, it was included in this research study. Individuals were considered to be at higher risk of frailty if their average maximum femoral length measurement fell within the lowest quartile for their age and sex cohort. Individuals who fell within the lowest quartile received a score of "1" for this category. All other individuals received a score of "0".

<u>3.3.2b Robusticity – Femoral Head Diameter</u>

In living populations, walking performance, physical activity, and strength are often used to assess frailty (Fried et al., 2001; Marklein et al., 2016). For this research study, skeletal robusticity (indicated by femoral head diameter) served as a proxy for these factors, as they cannot be directly observed in skeletal remains (Marklein et al., 2016). The femoral head is often well preserved in bioarchaeological assemblages, making it an effective indicator of skeletal robusticity that is likely to be present for comparison between population groups (Marklein et al., 2016). Femoral head diameter measurements were taken for each individual included in the study sample. Wherever possible, both the right and left femoral head diameters were taken and averaged. If either femur was unavailable, damaged, or affected by pathology, the opposite measurement was taken alone. Figure 3 provides an example of femoral head diameter measurements taken for this research study. Individuals for whom a femoral head diameter measurement could not be accurately obtained were excluded from the study sample. Metric quartiles were determined for each age and sex cohort using Microsoft Excel version 16.10. Individuals of indeterminate sex were considered as a separate group, regardless of age.



Figure 3: Example of Femoral Head Diameter Measurement. Bezławki, Poland. Photo by K. Gaddis (2017).

Criteria for scoring skeletal robusticity are similar to those outlined previously for stature. Individuals were considered to be at higher risk of frailty if they fall within the lowest quartile of femoral head diameter for their sex cohort. Therefore, individuals who fell within the lowest quartile of femoral head diameter for their age and sex cohort will receive a score of "1", while all others received a score of "0". Again, quartiles were compared only within sex cohorts to avoid complications due to sexual dimorphism.

3.3.2c Linear Enamel Hypoplasia

Enamel hypoplasia can be defined as abnormal deficiencies in enamel thickness that disrupt the normally smooth surface of dental crowns (Hillson, 1996). During amelogenesis, the process of enamel formation, ameloblasts work to secrete enamel matrix that will then mature to form the hard coating that covers the crowns of teeth (Hillson, 1996; White et al., 2011). During this process, stressful stimuli may disrupt ameloblast function, resulting in less matrix secretion and sections of reduced enamel thickness (Goodman and Rose, 1991; Hillson, 1996). Enamel defects often appear as thin bands of enamel deficiency that run the circumference of the crown parallel to the cemento-enamel junction (Hillson, 1996; Aufderheide and Rodriguez-Martin, 1998, Larsen, 2015). Figure 4 provides an example of linear enamel hypoplasias. In these instances, we refer to the defect as a linear enamel hypoplasia (LEH). While particularly prominent on the incisors and canines, linear enamel hypoplasia can also be observed on the cervical halves of premolars and molars (Hillson, 1996). The mandibular canines and maxillary incisors are most typically affected, as most instances of enamel hypoplasia occur during the period of life during which these teeth are forming (Aufderheide and Rodriguez-Martin, 1998).

Linear enamel hypoplasia, has been of particular use in bioarchaeological and paleopathological research due to the chronological nature of their distribution and their prevalence in skeletal assemblages (Goodman and Rose, 1991; Hillson, 1996; Ritzman et al., 2008). Enamel defects occur during development, allowing their location on the tooth surface can be read as a timeline of individual stress occurrences (Ritzman et al., 2008; Larsen, 2015). For example, stress events occurring earlier in an individual's life will be evidenced by enamel defects that are located closer to the occlusal surface than those representing stressors that occurred later in life. Enamel hypoplasias are particularly useful biomarkers of stress because, unlike skeletal tissues, enamel does not remodel and thus contains an essentially permanent record of stress events over the course of development (King et al., 2005). Although the particular source of stressor that results in linear enamel hypoplasias remains a topic of study, they serve as important non-specific indicators of stress in bioarchaeological research (Goodman and Rose, 1991; Larsen, 2015; Marklein, 2016; Ortner, 2011).

Linear enamel hypoplasias are typically observable macroscopically upon examination of dental surfaces under good lighting conditions (Figure 2; Yaussy et al., 2016). During analysis for this research, LEH were scored as being either present or absent. For this research study, linear enamel hypoplasias were indicated as being present if they were observable through macroscopic analysis on at least one tooth. Individuals scored as present for linear enamel hypoplasias were considered to be at higher risk of frailty and therefore received a score of "1" for this category of the Skeletal Frailty Index. Individuals with no enamel hypoplasias were assigned a score of "0".



Figure 4: Linear Enamel Hypoplasia in a sub-adult. Bezławki, Poland. Photo by K.

Gaddis (2017)

3.3.3 Nutrition/Infection

Nutrition and infection are considered together in this category of the Skeletal Frailty Index because of the inherent relationship that exists between them. Nutritional deficiencies may increase an individual's susceptibility to infectious disease, while disease may make it more difficult for the body to absorb essential nutrients (Larsen, 2015). The biomarkers included in this adapted SFI are indicators of nutritional deficiencies and infection that are often observed in bioarchaeological research: periostitis/osteomyelitis, osteopenia/osteoporosis, dental disease, cribra orbitalia / porotic hyperostosis, scurvy, and neoplasms (Marklein et al., 2016). Scurvy and dental disease were not included in the original Skeletal Frailty Index (Marklein et al., 2016), but were chosen for this adapted SFI due to their prevalence in the Bezławki sample population. The following sections explain the etiology, diagnostic criteria, and scoring criteria for each of the included biomarkers.

3.3.3a Periosteal New Bone Formation (Periostitis)/Osteomyelitis

Etiology:

Periosteal new bone formation serves as an important indicator of both specific and non-specific stress to the periosteum, the thin membrane that covers the outer surface of bones (Ortner, 2011; Waldron, 2009). When the periosteum is irritated by stressful stimuli, the underlying osteoblasts begin to create new, disorganized bone along the periosteal surface (Marklein, 2016). Essentially, periosteal new bone forms as a healing mechanism in response to harmful stimuli (Weston, 2008). In paleopathological literature, the term periostitis is used interchangeably to describe both a specific disease process (primary periostitis) and a lesion category associated with multiple other infectious diseases or traumatic injury (secondary periostitis) (Ortner, 2011). Nearly any type of insult to the periosteum can stimulate the production of new bone, regardless of inflammatory response (Weston, 2008). For these reasons, many paleopathologists have advocated for the use of the term periosteal new bone (PNB) formation in place of periostitis (Weston, 2008; Waldron, 2009).

Osteomyelitis describes an infection of the bone and bone marrow as a result of the introduction of an infectious agent into bone following localized infection, systemic infection spread through the bloodstream, or trauma (Waldron, 2009; Ortner, 2011). In nearly all cases, the infection is caused by the bacteria *Staphylococcus aureus*, which produces pus and causes localized skin infections that can spread to other organs and skeletal tissues (Waldron, 2009; Ortner, 2011). Once bacteria are introduced into the medullary cavity, they multiply and stimulate an inflammatory response that eventually leads to increased pressure within the cavity due to the production of pus (Waldron, 2009). Eventually, the bone may expand, causing periosteal reactions, localized areas of bone necrosis, and the formation of cloacae, which act as drainage channels for pus to exit the medullary cavity (Waldron, 2009). Although, it is possible for osteomyelitis to occur at any age, it is most commonly observed in children (Aufderheide and Rodriguez-Martin, 1998; Waldron, 2009). Acute osteomyelitis can prove fatal under unfavorable

circumstances, although untreated osteomyelitis can persist for several years before complications arise (Aufderheide and Rodriguez-Martin, 1998; Waldron, 2009).

Diagnostic Criteria:

Non-specific periosteal lesions are among the most commonly observed biomarkers of physiological stress observed in archaeological assemblages (Ortner, 2011). In their active form, they resemble patches of unorganized woven bone (Weston, 2008; Larsen, 2015). They are commonly observed on the diaphysis of the tibia, possibly because it is closer in proximity to the skin surface, putting it at higher risk of exposure to infectious pathogens and trauma (Ortner, 2011). Periosteal new bone formation can occur either unilaterally or bilaterally (Ortner, 2011). As they heal, the lesions will begin to resemble lamellar bone (Weston, 2008).

Characteristic properties of osteomyelitis include the presence of sequestra, cloacae and involucrum (Ortner, 2011). Sequestra are areas of bone necrosis, surrounded by living skeletal tissue, that can be observed either radiographically or through particularly large cloacae (Waldron, 2009). Involucrum occur when periosteal new bone formation occurs to such an extent that the new bone forms a thick layer around the infected bone (Waldron, 2009). Unless a cloaca or sequestra are present, osteomyelitis is difficult to differentiate from osteitis or periostitis in skeletal remains (Ortner, 2011). Osteomyelitis is most commonly observed in the long bones, particularly the femur and tibia (Aufderheide and Rodriguez-Martin, 1998).

Scoring Criteria:

Recent research in bioarchaeology has indicated that the presence of healed periosteal lesions may suggest higher rates of survivorship as compared to active lesions, suggesting that individuals with healed lesions were generally less frail than individuals with active lesions (DeWitte, 2014). This study did not attempt to associate periosteal new bone formation with any particular pathological condition, but rather used it as an indication of non-specific infection. In accordance with the Skeletal Frailty Index and previous research, only active lesions were scored as evidence of higher frailty (DeWitte, 2014; Marklein et al., 2016). If active periosteal new bone formation was observed on any skeletal element, the individual received a score of "1" for this category. Figure 5 provides an example of active periosteal lesions in an adult male from the Bezławki sample population. Individuals with either healed periosteal lesions, or no periosteal lesions, were assigned a score of "0" for this category. Furthermore, if a cloaca or sequestra were observed, indicating the presence of osteomyelitis, a score of "1" was received.



Figure 5: Active periosteal lesions, Adult Male. Bezławki, Poland. Photo by K. Gaddis (2017).

3.3.3b Dental Disease

The original Skeletal Frailty Index included periodontal disease as a biomarker of physiological stress related to nutritional deficiencies or infection (Marklein et al., 2016) This category has been expanded for this analysis to also include dental caries and abscesses. This expanded category has been designated as "dental disease". Oral health may reflect the general and systemic health of individuals, making dental pathologies such as caries, abscesses, and periodontal disease effective proxies for health in bioarchaeological research (DeWitte and Bekvalac, 2011). The observation of these conditions in skeletal remains suggest periods of stress related to nutritional deficiencies or infection and were thus included in the adapted SFI. For example, dental caries are a common source of oral pain and tooth loss (Waldron, 2009), and previous research has shown a relationship between caries prevalence and increased risk of mortality (DeWitte and Bekvalac, 2011). Painful caries may also limit food intake by affecting normal eating

processes, resulting in nutritional deficiencies (DeWitte and Bekvalac, 2011). Abscesses typically form following an infection of the dental pulp (Waldron, 2009). The following sections further define each of these commonly observed pathologies and explain how they were scored within the SFI.

Etiology:

The term periodontal disease typically refers to a chronic inflammatory condition involving the periodontal tissue, including the alveolar processes of the mandible and maxilla, the periodontal ligament, cementum, gingiva, and mucosa (Hillson, 1996; Aufderheide and Rodriguez-Martin, 1998). Periodontal disease has a rather complex etiology, involving genetic, environmental, dietary, and social factors such as hygiene (Hillson, 1996). Periodontal disease begins as gingivitis, during which the gums become inflamed in response to the presence of pathogenic bacteria in the dental plaque (Hillson, 1996; Waldron, 2009). The lesion will continue to develop until it penetrates deeply enough to involve the other periodontal tissues, at which point it is referred to as periodontitis (Hillson, 1996). If the lesion continues to progress, alveolar bone loss will eventually occur, undermining the support system of the affected teeth (Hillson, 1996; Ortner, 2003). At this stage, pockets can form between the gums and the tooth roots, which can fill with bacteria causing pain, discomfort, and eventual tooth loss (Waldron, 2009). Periodontal disease has been shown to be related to serious systemic diseases such as cardiovascular disease and diabetes (Waldron, 2009).

Dental caries are areas of dental tissue destruction resulting from acid production by the bacteria in dental plaque (Hillson, 1996). Certain variable characteristics of dentition, such as the presence of enamel defects or occlusal surface attrition may predispose individuals to dental caries (Ortner, 2003; Larsen, 2015). Additionally, factors such as exposure to infectious disease, diet, age, and genetic factors may further complicate individual susceptibility to dental caries (Larsen, 2015). In the early stages of demineralization, the process can be reversed through increased consumption of calcium, phosphate, or fluoride (Waldron, 2009). If allowed to continue, demineralization will result in the formation of caries (Waldron, 2009). While caries are typically observed on enamel, they can also form on root surfaces that have been exposed due to periodontal disease (Hillson, 1996). Once caries progress through either the enamel or cementum coatings, they spread into the dentine (Hillson, 1996). If caries remain untreated, they can lead to eventual loss of the dental crown or root, exposing the pulp chamber and leaving the individual more susceptible to infection (Ortner, 2003). If caries are deep enough to expose the root canal, infection can lead to the formation of a periapical abscess or granuloma (Waldron, 2009).

Abscesses and granulomas form following the death of dental pulp as the result of infection introduced into the pulp chamber via dental caries (Hillson, 1996). Following pulp death, the infection will continue through the apical foramen and initiate an inflammatory response in the periapical tissues (Hillson, 1996). The formation of granulomas or abscesses depend upon the severity of periapical infection and the host's response to that infection (Waldron, 2009). The primary response of periapical tissues to

infection is the formation of a granuloma, which develops as a smooth-walled cavity surrounding the apex of a tooth (Waldron, 2009). If pus collects in the granuloma cavity, an abscess will develop (Waldron, 2009). Acute abscesses will typically drain pus through the surrounding bone, resulting in the development of a large cloaca around the root apex (Hillson, 1996). While granulomas and chronic abscesses tend to be only mildly painful, acute abscesses can cause general malaise and severe pain (Waldron, 2009). In archaeological populations, dental abscess may be related to severe attrition rather than caries (Ortner, 2003).

Diagnostic Criteria:

In skeletal remains, periodontitis is evidenced by the destructive remodeling of the alveolar processes, resulting in exposure of the root surface of teeth (Ortner, 2003). Additionally, the alveolar margin may display pitting or new bone formation in response to inflammation (Waldron, 2009). Resorption can either be local or generalized (Ortner, 2003). Periodontal resorption must be distinguished from resorption as the result of abscess (Ortner, 2003). In dry bone specimens, periapical abscesses are typically diagnosed based by the large cavity created as pus drained from the periapical cavity (Hillson, 1996). This will result in exposure of the root cavity through the alveolar processes of the mandible or maxilla. Figures 6 and 7provide examples of periodontal disease and periapical abscesses, respectively. Note the recession of the alveolar bone in Figure 6, as indicated by the arrow.



Figure 6: Periodontal disease in an adult female. Bezławki, Poland. Photo by K. Gaddis

(2017).



Figure 7: Periapical abscess in an adult female. Bezławki, Poland. Photo by K. Gaddis (2017).

Dental caries vary in appearance depending on their severity (Hillson, 1996). Caries are generally observed in the early stages as white or brown spots on the surface of enamel (Waldron, 2009). As caries spread into the dentine, progressively more dental tissue is destroyed, and cavities begin to form (Waldron, 2009). Caries at the tooth apex are also common (Waldron, 2009). Dental caries are most often observed on the molars, followed by the premolars and then canines and incisors (Hillson, 1996). Caries prevalence increases with age (Waldron, 2009). Figure 8 provides an example of dental caries.



Figure 8: Large dental caries in an adult male. Bezławki, Poland. Photo by K. Gaddis (2017).

Scoring Criteria:

Individuals were diagnosed as having dental disease if caries, abscesses, or periodontal disease were observed. The observation of any of these three factors were considered evidence of increased frailty in individuals within this study sample. All available teeth were examined for dental disease. Caries were considered present if observed on at least one tooth. Abscesses were considered present if at least one abscess was observed. Periodontal disease was diagnosed according to the criteria presented above. Individuals with observed evidence of dental disease were assigned a score of "1" for this category of the Skeletal Frailty Index. All other individuals were assigned a score of "0".

3.3.3c Porotic Hyperostosis/ Cribra Orbitalia

Etiology:

Porotic hyperostosis and cribra orbitalia are commonly used as indicators of nutritional deficiencies in past populations. Although their specific etiology is still widely debated, both lesions are most often attributed to different forms of anemia (Ortner, 2003). Anemia is a condition characterized by a deficiency in either red blood cells or hemoglobin as the result of blood loss, impaired red blood cell production, increased red blood cell destruction, or some combination thereof (Walker et al., 2009). Anemic conditions can be either genetic or acquired following episodes of blood loss, nutritional deficiencies, or disease (Walker et al., 2009; Ortner, 2003).

While anemia is generally accepted by paleopathologists as a source of porotic hyperostosis and cribra orbitalia, there has been some debate as to the specific type of anemia that causes these lesions. In paleopathological literature, porotic hyperostosis and cribra orbitalia are most often attributed to iron-deficiency anemia (Aufderheide and Rodriguez-Martin 1998; Ortner, 2003; Waldron, 2009; Walker et al., 2009). Other researchers have suggested that megaloblastic anemia resulting from B-12 insufficiencies is a more likely explanation (Walker et al., 2009), although this claim has been met with resistance from proponents of the iron-deficiency hypothesis (Oxenham and Cavill, 2010; McIlvaine, 2013). Porotic hyperostosis and cribra orbitalia have also been recorded in relation to certain infectious, neoplastic, and metabolic diseases (Ortner, 2003).

Diagnostic Criteria:

Porotic hyperostosis and cribra orbitalia are two of the most commonly observed skeletal lesions in the archaeological record (Walker et al., 2009). Both can be described as areas of abnormal porosity on the skull (Ortner, 2003). In general, the term porotic hyperostosis is used to describe these lesions as they occur on the outer cranium, while cribra orbitalia is used to describe these lesions on the orbital roofs (Ortner, 2003).

Porotic hyperostosis is typically distributed symmetrically across the outer surfaces of the frontal and parietal bones (Aufderheide and Rodriguez-Martin, 1998). In fully developed lesions, the affected areas are thickened, and the outer table of the bone is completely resorbed, permitting vascularization and coarsening of the exposed diploë (Aufderheide and Rodriguez-Martin, 1998). In less extreme or healing cases, the outer table tends to be incompletely resorbed and the lesions present as multiple small perforations or take on an orange peel appearance (Aufderheide and Rodriguez-Martin, 1998; Mann and Hunt, 2005). Porotic hyperostosis typically develops in childhood and is often seen in the healing stages in adult individuals (Aufderheide and Rodriguez-Martin, 1998).

Cribra orbitalia is similar in appearance to porotic hyperostosis but occurs in smaller patches on the orbital roofs, almost always bilaterally (Aufderheide and Rodriguez-Martin, 1998). Although frequently observed together, cribra orbitalia occurs more commonly than porotic hyperostosis and is therefore often considered to be a more sensitive biomarker of stressful stimuli (Aufderheide and Rodriguez-Martin, 1998). Figure 9 provides an example of cribra orbitalia.



Figure 9: Healed cribra orbitalia in an adult female. Bezławki, Poland. Photo by K. Gaddis (2017).

Scoring Criteria:

Despite the debate surrounding the specific etiology of porotic hyperostosis and cribra orbitalia, they remain effective biomarkers of physiological stress and are therefore included in this analysis as indicators of nutritional deficiencies and infection (Marklein et al., 2011). Porotic hyperostosis and cribra orbitalia may indicate prolonged exposure to harmful stimuli, as expansion of the cranial vault only occurs in response to anemia after less-costly responses have failed to mitigate the effects of the stressor (Walker et al., 2009). Therefore, individuals in this study will be considered to be at higher risk of frailty if evidence of either porotic hyperostosis or cribra orbitalia are observed. Individuals with either cribra orbitalia or porotic hyperostosis were assigned a score of "1" for this category of the SFI. All other individuals were assigned a score of "0".

3.3.3d Scurvy

Deviating slightly from the original methodology proposed by Marklein and colleagues, I employed scurvy as a representation of malnutrition in early childhood, rather than rickets/osteomalacia (Marklein et al., 2016). Rickets has not been observed in the Bezławki population to date, although lesions likely associated with scurvy have been observed in a few sub-adult individuals. Shifting dietary patterns in the region at this time may have been conducive to the development of scurvy. Following colonization by the Teutonic Order, cereal grain production increased in the region and traditional ways of gathering food began to change (Brown and Plusowski, 2014; Pluskowski, 2013). This may have led to a reduction in available fresh foods that would have previously been consumed.

Etiology:

Scurvy is well known to be the result of prolonged Vitamin C deficiency (Ortner, 2011). Unlike most mammals, humans are unable to produce their own vitamin C and thus must obtain it through the food that they consume (Brickley and Ives, 2008). Vitamin C is commonly found in a variety of fresh fruits and vegetables, as well as milk, meat, and fish (Brickley and Ives, 2008). The regular consumption of fresh foods is essential to maintain healthy levels of vitamin C, as it cannot be stored in the body

(Brickley and Ives, 2008). Cases of infantile scurvy are often attributed to weaning, as children of this age acquire their vitamin C from breastmilk (Brickley and Ives, 2008). Adult scurvy has been associated with the introduction of agriculture in some societies as individual diets changes in response to new methods of food production (Brickley and Ives, 2008). Scurvy is also often seen in groups of people who have recently migrated to a new part of the world, where available foods differed from their traditional diets (Brickley and Ives, 2008).

The essential contributions that vitamin C makes to various physiologic systems can be summarized as two primary functions: assisting in maintaining effective immune responses and the production of protective antioxidants (Brickley and Ives, 2008). Vitamin C also plays an important role in blood formation and iron metabolism, meaning that individuals who are deficient in vitamin C are also more likely to develop anemia (Brickley and Ives, 2008). Additionally, vitamin C is an essential factor in the formation of collagen, a major component of organic bone matrix (Ortner, 2011). Scurvy, therefore, often results in bone loss and arrested bone formation, leading to the development of osteopenia, increased susceptibility to fracture, and growth disruption in juveniles (Brickley and Ives, 2008; Ortner, 2011).

Symptoms of scurvy can begin to present within 29-90 days of vitamin C deprivation (Pimentel, 2003). These will vary in severity depending upon the age of the affected individual and the length of deficiency (Brickley and Ives, 2008). In living individuals, the most characteristic feature of scurvy is tissue bleeding, which can happen either spontaneously or secondary to minor trauma (Brickley and Ives, 2008; Ortner,

2011; Pimentel, 2003). Untreated scurvy inevitably results in sudden death or death secondary to infections (Pimentel, 2003).

Diagnostic Criteria:

Skeletal manifestations of scurvy are seen most prominently in infants (Ortner, 2011). Adolescents and adults show very minor evidence by comparison (Ortner, 2011). Adult manifestations of scurvy are typically limited to fracture at the osteocartilaginous joints of the ribs and inflammatory changes in alveolar bone (Ortner, 2011). Abnormal porosity of the alveolar margins may be differentiated from similar lesions associated with periodontal disease because scurvy will cause porosity that extends further beyond the alveolar processes (Brickley and Ives, 2006). Osteoporosis may also be indicative of scurvy in adult individuals (Waldron, 2009). Finally, cranial porosities and PNB, particularly on the frontal and parietal tubers and greater wing of the sphenoid are diagnostic criteria for scurvy in adult skeletal remains (Ortner, 2011; Waldron, 2009). No individuals in this research sample were diagnosed with scurvy. Therefore, no photos are available.

Scoring Criteria:

Scurvy was employed as a biomarker of nutritional stress in the observed sample. Therefore, the presence of scurvy in adult human skeletal remains was considered to be indicative of higher frailty. Individuals who could be diagnosed with scurvy were assigned a score of "1" for this category of the SFI. This proved particularly difficult for this research sample due to the ages of the individuals involved. However, evidence of scurvy discovered in previous investigations of the site suggested that this category should be included as a potential biomarker of stress. Individuals for whom a confident diagnosis of scurvy could not be determined were assigned a score of "0" for this category.

3.3.3e Neoplasms

Etiology:

Neoplasms are localized areas of abnormal new growth within the body, often referred to as tumors (Aufderheide and Rodriguez-Martin, 1998; Waldron, 2009). The tumors found in skeletal tissues originate as the result of uncontrolled growth of bone, cartilage, fibrous tissue, or blood vessels (Ortner, 2011). Tumors characterized by localized areas of controlled growth will well-defined (sclerotic) borders are classified as benign, whereas tumors that consist of poorly differentiated (lytic) regions of uncontrolled growth are classified as malignant (Aufderheide and Rodriguez-Martin, 1998; Ortner, 2011; Waldron, 2009). Benign tumors are by far more common than malignant tumors (Waldron, 2009). Malignant tumors may spread through the bloodstream to other areas of the body, resulting in the formation of metastatic tumors (Ortner, 2011). Metastatic bone tumors are encountered much more frequently in skeletal assemblages than malignant primary tumors, which originate within the tissues in which they are found (Ortner, 2011). Several important factors influence the prevalence of bone neoplasms (Ortner, 2011). Environmental factors, such as excessive exposure to the ultraviolet radiation produced by sunlight, may contribute to the determination of both tumor type and prevalence (Aufderheide and Rodriguez-Martin, 1998; Ortner, 2011). In modern human populations, chemical agents have also been shown to be a contributing factor to the development of cancerous tumors (Aufderheide and Rodriguez-Martin, 1998). There is also some evidence to suggest that viruses may play a role in the development of metastatic tumors by disrupting normal regulation of cell growth (Aufderheide and Rodriguez-Martin, 1998). Regardless of cause, the presence of neoplasms in skeletal remains represent the body's exposure to some period of physiological stress (Marklein et al., 2016). While many benign tumors would likely be asymptomatic, the metastatic tumors that indicate various cancers would have had a serious effect on an individual's ability to function normally in their society by restricting their ability to perform daily tasks (Marklein et al., 2016).

Diagnostic Criteria:

There are many different categories and classifications of tumors that are potentially observable in human skeletal remains. Different types of tumors vary in their location and appearance, and definitive diagnosis from dry-bone specimens is often challenging (Aufderheide and Rodriguez-Martin, 1998; Ortner, 2011; Waldron, 2009). Generally, neoplasms can be observed as areas of abnormal bone growth or abnormal bone resorption, depending upon the category of tumor represented (Aufderheide and Rodriguez-Martin, 1998; Marklein et al., 2016; Ortner, 2011; Waldron, 2009). None of the individuals within this sample population were diagnosed with neoplasms of any kind. Therefore, no photos are available.

Scoring Criteria:

Neoplasms were considered present if at least one neoplasm was observed anywhere in the skeleton. For the purposes of this study, cysts and benign tumors were not considered as biomarkers associated with physiological stress. Only metastatic tumors were considered to be associated with higher risk of frailty for the purposes of this research study. Individuals with neoplasms that fit these criteria were given a score of "1" for this particular category. Individuals with no neoplasms were assigned a score of "0".

3.3.3f Osteopenia/Osteoporosis

Etiology:

Osteoporosis is a metabolic condition characterized by abnormal bone loss accompanied by an increased risk of fracture from minor trauma (Brickley and Ives, 2008). Osteopenia is the precursor to osteoporosis, defined as bone loss without increased fracture risk (Brickley and Ives, 2008). Osteopenia can, however, also occur in relation to osteomalacia, hyperparathyroidism, cancer, and severe malnutrition (Ortner, 2011). Osteopenia and osteoporosis occur when there is an imbalance in the process of bone remodeling, resulting in an overall greater increase in bone loss relative to bone formation (Brickley and Ives, 2008; Ortner, 2011). These conditions naturally occur with age and are commonly attributed to menopause in women, although they may also occur secondary to infectious disease, nutritional imbalance, or trauma (Brickley and Ives, 2008).

Multiple factors contribute to the development of age-related osteoporosis. By the end of adolescent growth, individuals have achieved their peak bone mass, determined by environmental factors, childhood health, nutritional quality, physical activity, and genetics (Brickley and Ives, 2008). Age initiates imbalances in the bone remodeling process that contribute to progressive losses in skeletal tissue (Brickley and Ives, 2008). If an individual fails to attain adequate peak bone mass during the growth period, bone loss is likely to result in osteoporosis-related structural changes sooner (Brickley and Ives, 2008). Since males typically reach peak bone mass later in life than females, they tend to accumulate more skeletal tissue overall and are therefore less likely to develop age-related osteoporosis (Brickley and Ives, 2008). The loss of estrogen during menopause further exacerbates imbalances in the remodeling process, putting women at an even higher risk of developing osteoporosis during their lifetime (Brickley and Ives, 2008). Furthermore, mechanical loading plays an important role in the maintenance of bone strength (Brickley and Ives, 2008). As individuals become less active, they become more susceptible to osteopenia and osteoporosis (Brickley and Ives, 2008).

Secondary osteopenia may occur in response to injury, pathology, or nutrient deficiency (Brickley and Ives, 2008). Injury or subsequent infection and pain may result in decreased mobility or an inability to bear weight, ultimately leading to localized bone loss in response to disuse (Brickley and Ives, 2008; Ortner, 2011). The healing process

frequently requires the immobilization of the injured area, which may quickly lead to the development of secondary osteopenia (Brickley and Ives, 2008). Infectious disease and congenital and developmental conditions may similarly limit the normal daily functions of individuals, limiting their mobility and requiring temporary disuse of limbs (Brickley and Ives, 2008). Other pathological conditions may themselves cause severe bone destruction or alter the structure of skeletal tissues, resulting in the onset of secondary osteopenia (Brickley and Ives, 2008). Finally, dietary insufficiencies such as scurvy can dramatically increase an individual's risk of developing osteopenia (Brickley and Ives, 2008).

Diagnostic Criteria:

Reductions in the amount of skeletal tissue are not themselves indicative of osteoporosis unless structural changes result in an increased risk of fracture (Brickley and Ives, 2008). Prior to the onset of fracture, structural changes related to osteoporosis are not observable macroscopically in archaeological specimens (Brickley and Ives, 2008). The effects of age-related osteopenia and osteoporosis are most commonly observed in the spine, ribs, sternum, pelvis, and femoral neck (Ortner, 2011). Osteopenia and osteoporosis compromise the structural integrity of the skeletal system, putting individuals at risk of fracture from even minor trauma (Ortner, 2011). No photos are provided for this category, as it is difficult to capture evidence of osteopenia/osteoporosis through photographs.

Scoring Criteria:

For the purposes of this study, osteoporosis was considered present only if associated fractures were observed. Osteopenia was diagnosed on the basis of unusually lightweight bones, but only in instances where taphonomic damage could be confidently ruled out. Individuals with observed osteoporosis or osteopenia were considered to be at higher risk of frailty and therefore assigned a frailty score of "1" for this category of the Skeletal Frailty Index. Individuals without evidence of osteoporosis or osteopenia were assigned a score of "0".

3.3.4 Movement

Exposure to physiological stressors can present as conditions that may limit an individual's mobility. Physical activity places strain on the body over the course of a lifetime that can result in restrictions to mobility and a reduced capacity for movement (Marklein et al., 2016). This can lead to impairment of an individual's ability to complete tasks associated with normal activities of daily life, affecting their ability to function effectively in society (Marklein et al., 2016). This is a commonly used indication of frailty in living human populations (Fried et al., 2001). Biomarkers that will be used to indicate diminished mobility include neoplasms, osteoporosis, osteoarthritis, intervertebral disc disease, and rotator cuff disorder.

3.3.4a Osteoarthritis

Etiology:

Osteoarthritis (OA) is a commonly observed pathological condition in both living and historical populations (Ortner, 2011). Generally slow to progress, osteoarthritis involved the initial destruction of the articular cartilage, followed by reactive changes in the subchondral bone (Ortner, 2011; Waldron, 2009). While the specific cause of osteoarthritis is still being researched, several factors are thought to contribute to its development, including age, sex, race genetics, obesity, trauma, and activity patterns (Waldron, 2009). Movement is a clear component in the development of osteoarthritis, as only joints that allow movement develop the condition (Waldron, 2009). The association between mechanical loading and the development of osteoarthritis has led many researchers to consider its use as a tool in the reconstruction of past lifestyles, although this has been met with some degree of hesitation due to the multifactorial etiology of the condition (Larsen, 2015; Weiss, 2007).

Clinical manifestations of osteoarthritis involve primarily pain and joint stiffness (Burt et al., 2013). This may be accompanied by swelling around the joint, crepitus, and restricted range of motion (Burt et al., 2013). Often, individuals who are diagnosed with osteoarthritis report no discomfort and, as such, skeletal evidence of osteoarthritis does not necessarily indicate that the individual experienced significant symptoms during life (Burt et al., 2013; Waldron, 2009). However, because osteoarthritis and associated conditions have the potential to restrict an individual's range of motion, which likely

would have influenced their ability to participate in essential daily activities, it has been included as an indicator of increased frailty (Marklein et al., 2016)

Diagnostic Criteria:

Osteoarthritis primarily affects the articular cartilage, which breaks down as the disease progresses, slowly exposing the underlying subchondral bone (Burt et al., 2013; Ortner, 2011; Waldron, 2009). Subsequent skeletal changes involve the production of new bone and pitting at the joint surface, osteophyte formation at the joint margins, changes to the normal shape of the joint, and eburnation (Waldron, 2009). Of these changes, eburnation is the most important diagnostic criteria of osteoarthritis in human skeletal remains as it is clearly distinguishable from all other joint diseases (Waldron, 2009). Figure 10 provides an example of spinal osteoarthritis from the Bezławki population.



Figure 10: Spinal OA in an adult female. Note the osteophytes present at the margins of the vertebral bodies. Bezławki, Poland. Photo by K. Gaddis (2017).

Scoring Criteria:

Osteoarthritis affects anywhere from a single joint to many joints (Waldron, 2009). Joints commonly affected by osteoarthritis include the interphalangeal joints of the hands, the base of the thumb, the knee, the hip, and apophyseal joints of vertebrae (Ortner, 2011). In archaeological samples, osteoarthritis may be considered present if either eburnation or at least two of the following criteria are observed: marginal osteophytes, new bone at the joint surface, pitting at the joint surface, or changes to the joint contour (Waldron, 2009). These criteria were employed in this research study to diagnose OA. Osteoarthritis was considered present if evidence was observed anywhere in the skeleton. Individuals with observed osteoarthritis were considered at higher risk of frailty and assigned a frailty score of "1" for this category of the SFI. Individuals with no evidence of OA were assigned a score of "0".

3.3.4b Intervertebral Disc Disease

Etiology:

Intervertebral disc disease refers to a degenerative condition that affects the intervertebral discs of the spine, which allow for movement within the vertebral column (Burt et al., 2013; Waldron, 2009; White, 2011). The intervertebral discs are composed of an outer collagen layer called the annulus fibrosus and an inner gelatinous later called the nucleus pulposus (Waldron, 2009). Progressive degeneration of the intervertebral discs leads to increased contact between the vertebral margins, which causes irritation that stimulates the production of osteophytes (Aufderheide and Rodriguez-Martin, 1998). Additionally, damage to the vertebral endplate following disc degeneration may cause vertical herniation of the nucleus pulposus into the adjacent vertebral body, resulting in a small, circular lesion known as a Schmorl's node (Plomp et al., 2012; Waldron, 2009). Although recent research has shown that Schmorl's nodes are frequently asymptomatic, they are often considered to represent the effects of biomechanical stress in bioarchaeological research (Burt et al., 2013; Kyere et al., 2012). Eventually, compression of the vertebral bodies may result in damage to neurological, mechanical, and vascular systems, which may cause functional impairment (Marklein et al., 2016; Waldron, 2009).

Diagnostic Criteria:

Intervertebral disc disease is common in archaeological samples and can be diagnosed based on the presence of both marginal osteophyte formation and pitting on the surfaces of the vertebral bodies (Waldron, 2009). Ultimately, fusion may occur between osteophytes of adjacent vertebrae as the vertebral central continues to deteriorate, as shown in figure 11 (Marklein et al., 2016). The presence of Schmorl's nodes further indicate a diagnosis of intervertebral disc disease (Figure 12).



Figure 11: Intervertebral disc disease in an adult male. Bezławki, Poland. Photos by K.

Gaddis (2017).

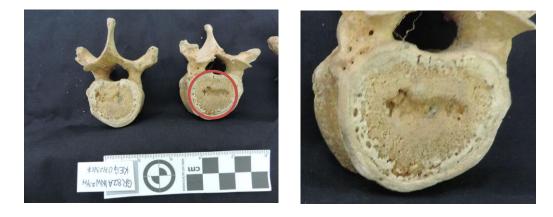


Figure 12: Schmorl's nodes in an adult male. Bezławki, Poland. Photos by K. Gaddis

(2017).

A major complicating factor of this particular frailty index is differentiation between pathological changes associated with intervertebral disc degeneration and those that occur as a result of osteoarthritis. While many of the changes commonly seen in spinal osteoarthritis are also observed in cases of intervertebral disc disease, changes specific to intervertebral disc degeneration can be useful in differentiating between the two conditions. Intervertebral disc disease involves marginal osteophyte formation and pitting on the vertebral body surfaces but does not include pitting or osteophyte formation on the vertebral facets (Ortner, 2011). Changes associated with the synchondroses alone cannot technically be considered arthritis, as they do not involve synovial joints, and thus represent intervertebral disc disease rather than osteoarthritis (Ortner, 2011). For the purposes of this research, osteoarthritis will be considered present if changes to the vertebral facets is observed, regardless of presence of changes to the vertebral bodies. If only changes to the vertebral bodies are observed, intervertebral disc degeneration will be the diagnosis.

3.3.4c Rotator Cuff Disorder

Etiology:

Rotator cuff disorder is a degenerative condition associated with overuse of the shoulder (Waldron, 2009). It is a common source of shoulder pain and disability in contemporary populations (Roberts et al., 2007; Waldron, 2009). The rotator cuff is composed of four muscles, which act to stabilize the humeral head in the glenoid fossa. (Marklein et al., 2016). These muscles include the subscapularis, supraspinatus,

infraspinatus, and teres minor (Marklein et al., 2016; Waldron, 2009). Over time, these muscles experience strain and eventually weaken (Marklein et al., 2016).

Diagnostic Criteria:

Skeletal changes as a result of rotator cuff disorder (RCD) are typically observable on the acromion, coracoid process, acromioclavicular joint (ACJ), and bicipital groove (Waldron, 2009). Diagnostic criteria for rotator cuff disorder in skeletal remains involves pitting, abnormal shape, or enthesophyte presence in any of these areas (Waldron, 2009). New bone formation is also occasionally present (Waldron, 2009). Figure 13 provides an example of rotator cuff disorder in an adult individual from the Bezławki research sample. Although manifestations of rotator cuff disorder are similar to those expressed in cases of osteoarthritis, rotator cuff disorder typically affects insertion points of the rotator cuff muscles, whereas osteoarthritis will affect the joint capsule itself (Marklein et al., 2016). These criteria will be used to differentiate between the two during diagnosis.



Figure 13: Abnormal porosity of the acromion associated with rotator cuff disorder in an adult male. Bezławki, Poland. Photo by K. Gaddis (2017).

Scoring Criteria:

Rotator cuff disorder was considered present if the above criteria could be observed on the available skeletal elements. For a diagnosis of RCD to be considered, the humerus, scapula, and clavicle had to be analyzed. Individuals considered to have rotator cuff disorder were assigned a score of "1" for this category. Individuals who did not exhibit signs of RCD, or for whom an accurate diagnosis was not possible due to a lack of available skeletal elements, were assigned a score of "0".

3.3.5 Trauma

Identification of traumatic lesions in human skeletal remains provides important information regarding the social and environmental conditions of past population groups (Aufderheide and Rodriguez-Martin, 1998). Strain is placed on normal bodily functions throughout the healing process following a traumatic event (Marklein et al., 2016). Evidence of trauma in skeletal remains may come as the result of accidental occurrences, intentional violence, or cultural cosmetic or surgical practices (Ortner, 2011). In general, trauma takes four primary forms in skeletal tissues, including partial to complete bone breaks, displacement or dislocation, disruptions to nerve or blood supply, or abnormal changes in shape (Ortner, 2011).

In human skeletal remains, trauma is typically assessed through the analysis of fractures. Fractures are cracks in bone that may or may not accompany injury to the surrounding soft tissue (Aufderheide and Rodriguez-Martin, 1998). Fractures are commonly observed in skeletal assemblages and can take a variety of different forms depending upon the source of the injury. Risk of fracture may increase as the result of developmental or pathological conditions (Aufderheide and Rodriguez-Martin, 1998). Fractures may also lead to complications that impair an individual's ability to interact effectively within their society to accomplish essential daily tasks, including restricted movement, delayed healing, secondary infections, and nerve damage (Aufderheide and Rodriguez Martin, 1998).

Scoring Criteria:

All available skeletal elements for each individual included in the research sample were analyzed for evidence of trauma. If trauma was observed, regardless of cause or severity, the individual received a score of "1" for this category of the SFI. Multiple occurrences of trauma were given the same score as single occurrences. If no trauma was observed, the individual received a score of "0" for this category. Figure 14 provides an example of trauma in an individual from the Bezławki research sample.



Figure 14: Clavicle fracture in an adult male. Bezławki, Poland. Photo by K. Gaddis

(2017).

4. RESULTS

The null hypotheses for this research study assert that there will be no association between frailty and age, and frailty and sex among members of the Bezławki population. Alternative hypotheses state that there will be statistically significant correlations between frailty and age and frailty and sex. The following chapter details the statistical analyses conducted to determine the relationship between frailty and these two independent variables and the results achieved from these assessments. Results of this study report an average skeletal frailty score of 3.0 (standard deviation 1.20) for the total sample population (n=37), with individual frailty scores ranging from 1 to 6.

4.1 Frailty and Sex

Using Excel version 16.10, an F-test revealed equal variance between male (n=15) and female (n=16) mean frailty scores (P>0.05). The subsequent T-test did not reveal a significant difference between the sexes (P>0.05). Those individuals for whom sex could not be accurately determined (n=6) were not included in this test. Results of this analysis revealed that there is no statistically significant correlation between sex and frailty for this population group. The threshold of statistical significance was set at .05 for this analysis. Frailty score distributions by sex are summarized in Table 2.

Table 2: Mean	frailty	scores	by s	sex.
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Sex	Mean Frailty Index	
Female (n=16)	2.88 (1.15)	
Male (n=15)	3.07 (1.33)	

4.2 Frailty and Age

Ideally, mean frailty scores for each of the four age cohorts described in this study would have been compared with the scores of the other cohorts. For the purposes of this analysis, the two older age cohorts were combined (n=18), as were the two younger age cohorts (n=19), in order to increase the available sample sizes. Therefore, the younger age cohort included individuals between the ages of approximately 14-35 years, and the older cohort included individuals between the ages of approximately 35-65 years. An ordinary least squares regression analysis and one-way Analysis of variance (ANOVA) revealed that there was not a statistically significant relationship between mean age estimate and frailty score (n=37, R²=0.01, ANOVA F=0.272, P=0.61). This pattern remained insignificant when data were analyzed separately for females and males. The results of an F-Test indicated that the variances between individuals <35 years (n=19) and individuals 35+ years (n=18) were not significantly different (P>0.05); therefore, a T-test for two-samples assuming equal variances was performed. The results of the T-test [T (1.39) < t Critical two-tail (2.03), P>0.05] indicate that the mean frailty score of the younger age cohorts (2.74) was not significantly lower than the mean frailty score of the

older age cohorts (3.28) (P>0.05) Frailty score distributions by age are summarized in table 3.

Age Category	Mean Frailty Index (Standard Deviation)	Female Means (Standard Deviation)	Male Means (Standard Deviation)
<25 Years (n=10)	2.90 (1.20)		
25-35 Years (n=9)	2.56 (1.01)	3.33 (0.58)	2.17 (0.98)
<35 Years (n=19)	2.74 (1.10)		
35-45 years (n=14)	3.50 (1.29)	3.20 (1.03)	4.25 (1.71)
>45 Years (n=4)	2.5 (1.00)		
>35 Years (n=18)	3.28 (1.27)		

Table 3: Mean frailty scores by age.

4.3 Biomarker Frequencies

The most commonly observed pathologies among the sample from the Bezławki population were linear enamel hypoplasia (50% females, 80% males) and dental disease (88% females, 47% males). Porotic hyperostosis and cribra orbitalia were observed to increase in frequency with age, perhaps indicating nutritional deficiencies in older individuals. Osteoarthritis and dental disease increase in frequency with age, but neither are noted in the oldest age cohort (>45 years), although this may be due in part to the small sample size (n=4). Furthermore, no instances of either scurvy or neoplasms were

reported in this sample population. An absence of scurvy is not entirely unexpected, as skeletal manifestations are generally subtle in adult individuals (Ortner, 2011). Finally, all age and sex groups exhibited signs of trauma, perhaps suggesting that all individuals were at equal risk of suffering fracture due to accidental injury, intense labor, or violence. Tables 4 and 5 provide a summary of biomarker prevalence values observed in the Bezławki research sample.

Biomarker	Population Prevalence (n=37)	Male Prevalence (n=15)	Female Prevalence (n=16)
Linear Enamel Hypoplasia	0.68	0.80	0.50
Periosteal New Bone/ Osteomyelitis	0.14	0.20	0.06
Dental Disease	0.65	0.47	0.88
Porotic Hyperostosis/ Cribra Orbitalia	0.16	0.27	0.06
Scurvy	0.00	0.00	0.00
Neoplasms	0.00	0.00	0.00
Osteopenia/Osteoporosis	0.11	0.07	0.13
Osteoarthritis	0.24	0.20	0.31
Intervertebral Disc Disease	0.11	0.20	0.06
Rotator Cuff Disorder	0.03	0.07	0.00
Fractures/Trauma	0.38	0.47	0.38

Table 4: Biomarker prevalence by sex.

Biomarker	Population Prevalence (n=37)	<25 Years (n=10)	25-35 Years (n=9)	35-45 Years (n=14)	>45 Years (n=4)
Linear Enamel Hypoplasia	0.68	0.80	0.78	0.50	0.75
Periosteal New Bone/ Osteomyelitis	0.14	0.30	0.00	0.14	0.00
Dental Disease	0.65	0.40	.056	0.93	0.50
Porotic Hyperostosis/ Cribra Orbitalia	0.16	0.10	0.11	0.21	0.25
Scurvy	0.00	0.00	0.00	0.00	0.00
Neoplasms	0.00	0.00	0.00	0.00	0.00
Osteopenia/Osteoporosis	0.11	0.20	0.00	0.14	0.00
Osteoarthritis	0.24	0.10	0.11	0.50	0.00
Intervertebral Disc Disease	0.11	0.00	0.22	0.14	0.00
Rotator Cuff Disorder	0.03	0.00	0.00	0.07	0.00
Fractures/Trauma	0.38	0.20	0.44	0.36	0.75

Table 5: Biomarker prevalence by age.

4. DISCUSSION

The skeletal remains included in the Bezławki research sample represent a population undergoing complex environmental change and social reform following a long period of colonization and conversion in the region (Pluskowski, 2013). The average frailty score for the population (3.0) indicates that stress was present in the population, although no statistically significant differences in mean frailty between age or sex cohorts were observed. These results may indicate that all older juveniles and adult individuals within the Bezławki sample were equally susceptible to the particular stressors experienced during this time period, which likely involved a rapid population increase and environmental changes following the arrival of the Teutonic Order. This sample size is small and further analysis may provide beneficial supplementary data. However, the results of this study provide important information regarding stress and frailty in Medieval Prussia.

This period of history is characterized by the colonization of formerly Prussian territories by the Teutonic Order, who chose to invite immigrants from nearby Germany and Poland to occupy the region (Pluskowski, 2013). As a result of this change, the cemetery at Bezławki may also include the remains of colonizers. Future research at the Bezławki site will involve investigating potential variations in biomarker frequencies that might indicate distinct groups within the Bezławki research sample. In the absence of ability to differentiate colonizers from native groups, all individuals within the sample population were considered together.

5.1 Frailty and Age

The statistical analysis of average frailty scores among members of the Bezławki population did not indicate a statistically significant (p>0.05) correlation between frailty and age. These results were unexpected considering the relationship between frailty and ageing commonly observed in living human population groups (Fried et al., 2001). The statistically insignificant variation in mean frailty scores between age cohorts may be reflective of the small sample size (n=37). A larger sample would be ideal to accurately represent relationships between frailty and age in the greater Bezławki population. However, statistically insignificant variation in mean frailty scores between age cohorts may be indicative of the substantial environmental and political changes taking place within the region at the time. Individuals may have been exposed to stress, regardless of age.

Although there does not appear to be a statistically significant correlation between age and frailty, some interesting general patterns were observed. An initial decline in mean frailty scores between the youngest and younger-middle age cohorts reflects observations seen in previous bioarchaeological research on frailty (Marklein et al., 2016). Individuals who experience high degrees of stress early in life are subject to increased mortality and are therefore more likely to die at a younger age. This results in an older age cohort composed of individuals with lower frailty who exhibit fewer skeletal biomarkers of stress. Cumulative stress should then continue to build over time, resulting in progressively higher mean frailty scores with age following the initial drop between the two youngest age cohorts. While a rise is frailty is exhibited between the two middle age cohorts, resulting in the older-middle age cohort displaying the highest mean frailty score relative to the other cohorts, mean frailty score dropped dramatically again between the two oldest age cohorts. This discrepancy is potentially due to the fairly small sample representing the oldest age cohort (n=4) compared with the older-middle age cohort (n=15).

Of particular concern to this research is the potential that physiological stress and the presence of stress biomarkers may affect the ability to properly estimate the age of individuals. The aging methods involved in this research study estimate age based on patterns of wear observed on the pubic symphysis and auricular surfaces. Pathological conditions may exacerbate wear to these surfaces, resulting in a potential over-estimation of age. Furthermore, certain pathological conditions may affect the fusion of various epiphyses, which may result in an under-estimation of age. To address this potential issue, multiple aging methods should be considered.

5.2 Frailty and Sex

Research involving living population groups has revealed common variations in frailty between the sexes (Berges et al., 2009; DeWitte, 2010; Fried et al., 2001; Puts et al., 2005). However, no significant correlation was observed between sex and frailty within this sample population based on the statistical analyses described above. These results may again be reflective of the small sample size available for analysis. The sample

included in the analysis of the relationship between frailty and sex is particularly small (n=31) because the six individuals for whom sex could not be accurately determined were excluded. These results may again, however, reflect the environmental and social changes occurring at this time. Both men and women were displaced and required to perform strenuous labor under the Teutonic Order (Pluskowski, 2013). With this in mind, a statistically insignificant difference in mean frailty index between males and females is not entirely unexpected.

5.3 Biomarker Prevalence

Variation in biomarker prevalence was observed between age and sex cohorts. Males, for example, were more likely to exhibit linear enamel hypoplasias (80% prevalence) than females (50% prevalence). However, females were more likely to exhibit signs of dental disease (88% prevalence) than males (47% prevalence). This may indicate that, while males were more likely to experience nutritional stress early in life than females, females were more likely to have poor dental health in general. Similar patterns showing higher caries rates among women are commonly reported in bioarchaeological research (Lukacs, 2008). Recent studies have indicated that increased prevalence among females may be related to behavioral or dietary differences between the sexes (Temple, 2010). It has been suggested that this difference in caries prevalence between males and females may be associated with female reproductive physiology, life history patterns, and pregnancy (Lukacs, 2008; Temple, 2010). Bezławki males also exhibited a higher prevalence of porotic hyperostosis and cribra orbitalia (0.20) than females (0.06), which may indicate that dietary differences may have contributed to higher caries prevalence among females. Although the exact nature of increased female caries prevalence among members of the Bezławki research sample is unclear, this observation presents an interesting opportunity for future research.

Furthermore, the observed lack of osteoarthritis in the oldest age cohort was unexpected. Osteoarthritis tends to increase in prevalence with age (Ortner, 2011). Therefore, a higher prevalence of OA in the oldest cohort would be expected. As discussed above, the observed lack of osteoarthritis in this age category may be reflective of the small sample size (n=4). However, it is also possible that lifestyle differences may have contributed to the absence of osteoarthritis. The Bezławki research sample includes individuals from multiple generations. The individuals included in the oldest age cohort may have lived prior to the introduction of strenuous agricultural activity in the region, resulting in overall lower OA prevalence. Unfortunately, it is difficult to determine to what generation these individuals may have belonged. An additional explanation for the absence of OA in this particular age group may lie in the influx of colonizers from Germany and Poland into Prussia that was occurring during this period of history. It is possible that the individuals included in the Bezławki research sample represent multiple population groups. During this period of time, native Prussians were required to perform strenuous agricultural tasks under the Teutonic Order. Individuals from Germany or Poland who were invited to settle in the area may not have been subject to the same heavy labor, and therefore would have been at less risk of developing osteoarthritis than their Prussian neighbors. Although it is not currently possible to distinguish between

population groups within the Bezławki research sample, future isotopic analysis may contribute to knowledge of lifestyle differences that would have contributed to variations in OA prevalence.

5.4 Limitations of Study

5.4.1 Skeletal Frailty Index Limitations

While the Skeletal Frailty Index presents a useful method for evaluating frailty in bioarchaeological research samples, there are several limitations to the applicability of the model that should be addressed here. These limitations include issues with regard to skeletal preservation, biomarkers included, and lack of appropriate weighting procedures for the different biomarkers. While these challenges may prove problematic with regard to effective application of the Skeletal Frailty Index to different sample populations, they nonetheless present opportunities for future research and further development of an otherwise useful tool for assessing frailty in past populations.

5.4.2 Skeletal Preservation and Sample Size

The first issue that was encountered in conducting data analysis for this research project was that efforts to apply the full 13-biomarker SFI resulted in a serious reduction in sample size. In order to be considered for analysis, enough skeletal elements needed to be present for each of the biomarkers to be reasonably evaluated. This requires a mostly complete skeleton that is largely unaffected by taphonomic processes that might obscure evidence of pathology. For this particular study, the primary reasons that individuals were excluded from the research sample were lack of skulls and lack of intact femora to obtain maximum femoral length for stature indications. Femoral damage was largely due to a modern powerline that cut through the cemetery site (A. Koperkiewicz, personal communication, July 2017).

Marklein and colleagues address the issue of preservation in their initial proposal of the SFI (Marklein et al., 2016). They have recently issued a follow-up analysis of their original sample using modified SFIs with varying numbers of biomarkers. Results of this study indicated that an SFI with as few as 6 biomarkers produced statistically significant results (Marklein et al., 2017). Additionally, the authors report that in the absence of adequate means to collect metric data, SFIs using only non-metric biomarkers may be implemented, although they will not produce results that are as specific as those containing both metric and non-metric data (Marklein et al., 2017).

While it would certainly be possible to adjust the number of biomarkers used in analysis to accommodate additional individuals, it was decided that preliminary analysis of frailty in the Bezławki population should include as many biomarkers as possible. Use of multiple indicators in frailty analysis provides a broader, more detailed understanding of frailty by encompassing various contributing factors. Application of an SFI with fewer biomarkers to obtain a larger sample size may be an opportunity for future research in Bezławki.

5.4.3 Biomarkers Included

In addition to issues of skeletal preservation, the original SFI does not accommodate several categories of pathological conditions that may serve as effective biomarkers of stress. For example, while the SFI accounts for instances of osteoarthritis and intervertebral disk degeneration, it does not account for occurrences of other forms of arthritis that may have impacted an individual's range of motion. Similarly, the original SFI accounts for periodontal disease but not other forms of dental disease such as caries and abscesses, which could certainly have influenced frailty. Following their proposal of the Skeletal Frailty Index, Marklein and colleagues suggest adapting SFI criteria to individual samples (Marklein et al., 2016). For the purposes of this study, for example, caries and abscesses were included in the SFI due to their high prevalence in the sample. However, this process complicates comparisons of frailty between population groups and thus is not an ideal solution.

One of the greatest strengths of the Skeletal Frailty Index is that it creates a standardized mechanism by which comparisons between population groups are made possible, allowing for greater understanding of frailty in the past and providing opportunities for future comparative work between living and past human groups. If SFI criteria were expanded to encompass broader categories of stress biomarkers, a more holistic understanding of frailty could be achieved. While development of such criteria may prove problematic in the short term, these efforts would eventually allow for better comparisons between population groups and increased sample size, providing more interesting data.

5.4.4 Weighting the SFI:

The final issue that will be addressed here concerns the binary nature of the Skeletal Frailty Index scoring system. While the presence versus absence scoring system provides a simple and standardized means for analysis, it fails to adequately take into account differences in severity of the various biomarkers of stress. This again becomes problematic regarding comparison both within and between sample populations. For example, within the SFI as it currently stands, a fractured metacarpal would be given the same weight as a fractured tibia. Additionally, early evidence of osteoarthritis would be given the same score as fusion of the vertebral bodies resulting from severe osteoarthritis or DISH. While all of these provide evidence of physiological stress, some likely had a larger impact on overall functional capacity than others.

Ideally, the Skeletal Frailty Index would account for variation in the degree to which stress may have been experienced by individuals in the past as evidenced by the severity of the stress response exhibited. Although, it would be difficult to properly ascertain the degree to which particular stress responses would have impacted individuals, there may be a way to weight the SFI such that some degree of variation in response is represented. This would provide a more detailed review of individual frailty that can be compared more accurately within population groups. If frailty is considered as a decrease in resistance to stressful stimuli as the result of cumulative functional decline, then a weighted SFI would more accurately consider individual stressors in regard to their actual risk to morbidity and mortality (Marklein et al., 2016). Better comparisons could be made between individual frailty scores if the Skeletal Frailty Index were better able to account for differential stress severity.

5.4.5 Applicability of the Skeletal Frailty Index

Overall, the Skeletal Frailty Index is a useful tool for assessing frailty in bioarchaeological research samples. The assessment of frailty in bioarchaeology is an important step towards a more comprehensive understanding of health and well-being in past societies. Despite several challenges presented by this new methodology, it provides opportunities for future research and important data on stress and frailty in past human groups. Collaboration with researchers in the fields of epidemiology, human biology, and pathophysiology may aid in further development of the SFI, improving upon its usefulness in making comparisons between living people and past populations.

CONCLUSION

The comprehensive macroscopic analysis of human skeletal remains for evidence of physiological stress is a common method by which bioarchaeologists begin to infer patterns of physiological health in the past. Bioarchaeological research benefits greatly from the incorporation of interdisciplinary methods from fields such as epidemiology and human biology into the study of stress because they provide insight into how commonly observed skeletal biomarkers relate to human health and well-being. The integration of concepts such as allostatic load and frailty, which are commonly used in clinical research to assess health, facilitate comparisons between past and present population groups to provide a means by which it may be possible to track patterns of physiological health over time. Furthermore, methods such as the Skeletal Frailty Index (Marklein et al., 2016) assess both individual and population frailty, which allows for direct comparison both within and between population samples.

This research study aimed to interpret variations in frailty between age and sex cohorts in a sample population from the Bezławki research site. Although the results of the statistical analyses were not significant, there were interesting trends in the data, which revealed opportunities for future studies and demonstrated the applicability of the SFI. Future opportunities to expand upon this thesis research would involve the exploration of the limitations presented above, including an adaptation of the Skeletal Frailty Index to include additional biomarkers, and the introduction of a weighted SFI. Ongoing research will also involve the assessment of a larger sample size and will explore the possibility of adjusting the Skeletal Frailty Index to account for variations in severity of stress responses. Medieval Prussia has a unique history that represents an era of complex environmental and social change from which we could learn much about physiological health in the past, and it is therefore deserving of further exploration.

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