

**Skeletal taphonomy of *Sus scrofa* within a
burial environment in the South African
Highveld**

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DECLARATION

I Claudia Landsman declare that this dissertation is my own, unaided work. It is being submitted for the Degree of Masters of Science (Medicine) at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at any other University.



14 day of June 2022 in Johannesburg

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ABSTRACT

Taphonomy is the study of the environmental effects on the remains after body deposition to the time of recovery. It has become an important component in forensic investigations as it enables forensic anthropologists to estimate the postmortem interval and reconstruct the events leading up to and after death. This study focused on the skeletal taphonomy of buried remains after six years of internment to establish, if any, the relationships between the soil composition (pH and organic content) and burial environment (depth and grave positioning) with any taphonomic alterations observed on the bone.

The study was conducted at the Miertjie le Roux experimental farm located in the interior Highveld of Gauteng. The sample was made up of 39 pigs that had been buried six years prior as part of a previous study. The remains were excavated using standard archaeological techniques. Six taphonomic alterations were observed and scored, including depositional staining, adipocere formation, weathering, acidic soil corrosion, plant activity, and animal activity. Soil samples were taken from three separate levels in each grave (10 cm, at the level of the carcass and 10 cm below the carcass). Each soil sample was then analyzed for pH and the percent organic content.

The most common type of taphonomic alterations was depositional staining, followed by weathering, and plant activity, as these were observed on all the skeletal remains. Adipocere was also frequently observed as it was on 36 of the 39 skeletons. Animal activity was the least observed as it was only present on four pigs. There were significant correlations between the taphonomic relationships which included the skeletal completeness with depositional staining, adipocere formation, weathering, and plant activity. There were also significant relationships between depositional staining and acidic soil corrosion, adipocere formation and plant activity as well as weathering and acidic soil corrosion and weathering and plant activity. There were significant correlations between the soil composition and the taphonomic alterations which included pH with the skeletal completeness, depositional staining, adipocere formation and weathering and organic content with adipocere formation and weathering. The correlations with the taphonomic alterations and the burial environment included the depth with adipocere formation and weathering as well as the grave positioning with depositional staining, adipocere formation and plant activity.

Skeletal preservation was low due to the absence of many small bones. It was however increased by the presence of adipocere. The two most common colours were dark brown and brown. Dark staining was correlated with a more complete skeleton. The right side of the skeletons were the most darkly stained as they were in close contact with the cadaver decomposition island (CDI). In addition, the right sides had more adipocere formation than the left. The presence of adipocere also increased the plant activity which suggests the graves retained water for adipocere formation and plant growth. Results from the soil analyses indicated that in general the presence of the cadaver lowered the pH of the burial environment and increased the percentage organic content when compared to the controls. There was also a significant difference observed between the surface level pH and organic content compared to the soil at the level of the carcass due to the presence of the decomposing carcass. A basic pH was associated with a more complete skeleton and an increase in adipocere formation. Adipocere formation also increased by the depth of the grave. Additionally, less adipocere formation, plant activity and dark staining remains were associated with graves at the top of a naturally occurring slope.

This study confirms that there are relationships between the soil composition and burial environment with the taphonomic alterations. The soils in this study seemed to retain water and thus formed adipocere and preserved the remains especially on the right side. This further establishes that the micro-environment of the grave is important factor influencing the macroscopic appearance of skeletal remains.

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CHAPTER 1: INTRODUCTION

The study of taphonomy was originally rooted in the discipline of palaeontology (Ubelaker, 1997; Carter *et al.*, 2007; Dirkmaat *et al.*, 2008; Manifold, 2012; Nawrocki, 2016; Schotsmans *et al.*, 2017). The term was coined by Efremov (1940) who first defined it as the study of the transition (in all its details) of animal remains from the biosphere to the lithosphere or geological record.

Taphonomy can further be divided into two divisions: biotaphonomy and geotaphonomy. Biotaphonomy focuses on the decomposition of the remains and the environmental factors that influence the remains themselves, while geotaphonomy looks at how the remains influence the surrounding environment such as the soil chemistry (Nawrocki, 2016; Rattenbury, 2018). Bodies or carcasses (in animal models) form part of a much larger and more complex ecosystem (Nawrocki, 2016). Nawrocki (2016) identified three broad categories of the factors that affect decomposition and thus the taphonomy; intrinsic factors such as age-at-death and diseases; extrinsic or environmental factors that can either be biotic (scavengers, insects, and plants) or abiotic (temperature, presence of water and solar radiation); and even human culture or behaviour.

Behrensmeyer, (1975, 1978, 1980) was one of the first to study the effects of taphonomy to understand fossil and bone assemblages. Behrensmeyer's (1978) research also included the development of a scoring system that was correlated to a specific number of years to estimate the time-since-death/deposition based on the degree of bone weathering observed on surface-scatter remains. These earlier studies laid the foundation for understanding dry bone taphonomic alteration, creating an avenue to utilise the same methodology in understanding taphonomic alterations in other disciplines, including forensic anthropology (Janjua and Rogers, 2008; Ross & Cunningham, 2011)

Taphonomy has since become an integral part of forensic anthropological analyses (Vass *et al.*, 1992; Turner & Wiltshire, 1999; Hopkins *et al.*, 2000; Benninger *et al.*, 2008; Haslam & Tibbett, 2009; Jagers & Rogers, 2009; Tumer *et al.*, 2013; Nawrocki, 2016). Haglund and Sorg (1997: 3) defined forensic taphonomy as the “use of taphonomic models, approaches, and analysis in forensic contexts to estimate the time since death, reconstruct the circumstances before and after deposition, and discriminate the products of human behaviour from those created by the earth's biological, physical and geological subsystems.”

The incorporation of taphonomy into the forensic field has broadened the goals of the forensic anthropologist. The traditional emphasis of forensic anthropology has been victim identification (Dirkmaat *et al.*, 2008; Iscan & Steyn, 2013; Rattenbury, 2018). Forensic taphonomy has now enabled anthropologists to further estimate the time since death or postmortem interval (PMI), reconstruct the original position, location, and orientation of a body, and determine the role, if any, of human involvement with the remains (Dirkmaat *et al.*, 2008; Nawrocki, 2016; Rattenbury, 2018). Taphonomy also enables forensic anthropologists to broadly differentiate between peri- and postmortem trauma. Perimortem trauma in the field of anthropology refers to any trauma that occurs around the time of death or when the bone still reacts as “fresh” or “green” bone. During this time the bone still has a substantial amount of collagen and water and thus will bend and has some pliability if a force is applied. Postmortem trauma occurs after death when the bone is dry and less flexible (Cattaneo & Cappella, 2017). In some instances, differentiating between peri- and post-mortem trauma can be difficult as slight variations in the environmental conditions and context of the remains, as it relates to the manner of disposal or burial, may greatly affect the morphology of the fracture patterns observed (Symes *et al.*, 2014; Cattaneo & Cappella, 2017; Gent, 2020). For this reason, a greater understanding of the suite of possible affecting factors and how each contributes to the breakdown of bone becomes essential in reconstructing peri- and post-mortem events.

Burial environments, like other methods of body disposal (e.g., surface, fluvial environments), exposes remains to unique conditions that have shown to result in specific decomposition rates (Turner & Wiltshire, 1999; Forbes, 2008; Carter *et al.*, 2010; Simmons *et al.*, 2010; Schotsmans *et al.*, 2011), taphonomic alterations (Littleton, 2000; Ross & Cunningham, 2011; Pokines & Baker, 2014; Delannoy *et al.*, 2016; Buekenhout *et al.*, 2018; Pollock *et al.*, 2018) and changes to the immediate environment (Wilson *et al.*, 2007; Benninger *et al.*, 2008; Anderson *et al.*, 2013; Fancher *et al.*, 2017; Hamilton & Green, 2017; Junkins & Carter, 2017). Various studies have specifically analysed the influence of the burial environment on decomposing remains. However, these studies have focused on the changes to the microstructure of bone (Yoshino *et al.*, 1991; Bell, Skinner and Jones, 1996; Hedges, 2002; Ross *et al.*, 2018), soil chemical changes (Fiedler & Graw, 2003; Haslam & Tibbett, 2009; Van Belle *et al.*, 2009) and the microorganisms involved with decomposition within a burial environment (Hackett, 1981; Child, 1995a).

Research on the macroscopic taphonomical changes to the bone in a burial environment is limited as many focus on archaeological bone (Child, 1995b; Littleton, 2000; Janjua & Rogers, 2008; Huculak & Rogers, 2009; Cunningham *et al.*, 2011; Delannoy *et al.*, 2016). The development of scoring criteria for taphonomy related to burial environments have mainly focussed on using Behrenmeyer's (1987) and Galloway *et al.*'s (1989) criteria, however, these have been shown to be less effective in capturing and describing all the taphonomic alterations seen in buried skeletal remains as both methods are meant to be used for surface scatter remains (Buekenhout *et al.*, 2018). It has therefore been suggested that future research be focused on better understanding decomposition in a variety of micro-environments, including burial environments (Cunningham *et al.*, 2011; Delannoy *et al.*, 2016; Buekenhout *et al.*, 2018).

In addition to understanding how different micro-environments may affect taphonomic alterations, it is also important to study the effects of these in different environmental conditions (Wilson *et al.*, 2007; Buekenhout *et al.*, 2018) as factors such as the pH of the soil, temperature fluctuations, types of microbial communities, and type of soil all influence the rate of decomposition and breakdown of the skeleton (Von Endt & Ortner, 1984; Collins *et al.*, 2002; Fiedler & Graw, 2003).

Studies that have been done within a South African context have analysed the soft tissue decomposition rates of both buried and surface remains as well as how clothing influences decomposition (Kelly, 2006; Sutherland *et al.*, 2013; Keough *et al.*, 2017; Marais-Werner *et al.*, 2017; Spies *et al.*, 2018; Finaughty, 2019; Spies *et al.*, 2020). Additionally, the studies done in the Western Cape provide different results than the studies conducted in the Highveld due to differences in the climate and ecology, emphasizing the need to establish region-specific data. These studies, however, did not include an analysis of skeletonized remains and primarily focused on soft tissue decomposition. Decomposition can, however, reach the extremes of skeletonization if the rate of decomposition is accelerated by environmental factors such as temperature or if the body is only recovered after an extended period. It is therefore necessary to also understand the full suite of alterations that may occur post-skeletonization.

To date, no South African studies, looking at the rate and effects of deposition on post-skeletonized remains in a medico-legal context, could be found. The aim of this investigation was, therefore, to study the interrelated relationships between dry bone taphonomic alterations, soil compositions and the burial environment on a sample of buried skeletonized domestic pig (*Sus scrofa*) in the South African Highveld.

To achieve the aim the following objectives were met:

- I. Assess the types, degree, and frequency of macroscopic taphonomic alterations present on the sample.
- II. Assess whether there is any correlation between the different types of taphonomic alterations observed.
- III. Assess whether there is any correlation between taphonomic alterations and soil composition (pH and organic content).
- IV. Assess whether there is any correlation between taphonomic alterations and the burial environment (depth and grave position).

CHAPTER 2: LITERATURE REVIEW

For the most part, taphonomic studies in forensic anthropology focus on the rate of soft tissue decomposition and its uses in the estimation of time-since-death or the post-mortem interval (PMI) (Galloway *et al.*, 1989; Mann *et al.*, 1990; Vass *et al.*, 1992; Turner & Wiltshire, 1999; Megyesi *et al.*, 2005; Myburgh *et al.*, 2013). The decomposition of remains is initiated immediately after death through the destructive processes of autolysis and putrefaction (Forbes *et al.*, 2005; Carter & Tibbett, 2008; Janaway *et al.*, 2009; Schotsmans *et al.*, 2011). Autolysis is the chemical breakdown of the body's tissues and is driven by hydrolytic enzymes from organelles known as lysosomes whereas putrefaction is dependent on microbes such as bacteria present in the gastrointestinal tract (Clark *et al.*, 1997; Janaway *et al.*, 2009; Forbes *et al.*, 2017).

Autolysis is initiated by the decrease in oxygen and the subsequent decrease in pH levels. It promotes decomposition by enzymatically breaking down cell membranes as well as macromolecules such as carbohydrates and proteins. The release of these nutrients will help stimulate the growth and reproduction of endogenous bacteria within the gastrointestinal tract that aid in putrefaction (Hamilton & Green, 2017). Most putrefactive bacteria are anaerobic and produce gas as well as aromatic organic compounds that produce the distinctive smell of decomposition. Putrefaction results in the liquification of the tissues and the release of molecules such as carbon dioxide, methane and nitrogen into the surrounding environment (Clark *et al.*, 1997; Janaway *et al.*, 2009; Forbes *et al.*, 2017).

These processes of decomposition are continuous; however, they proceed through a variety of defined stages that broadly include; fresh, bloating, active decay, advanced decay and skeletonization (Galloway *et al.*, 1989; Megyesi *et al.*, 2005). The sequence of these stages is similar in different environments but the time between each of these stages, and thus the decomposition rate, is influenced by the immediate environment and manner of deposition (Galloway *et al.*, 1989; Forbes *et al.*, 2005; Carter *et al.*, 2007; Schotsmans *et al.*, 2011; Buekenhout *et al.*, 2018). These stages become harder to distinguish as decomposition progresses into the later stages (Adlam & Simmons, 2007; Junod, 2013).

The skeletonization stage can be variable in a single body due to the differential decomposition of each region of the body (head and neck, trunk and limbs) (Galloway *et al.*, 1989; Janjua & Rogers, 2008). The bones of the abdomen and thorax, for example, are exposed to a higher degree of putrefaction due to the presence of intestinal bacteria compared to the bones of the

limbs and skull (Child, 1995b). Once skeletonization has been reached various factors will affect the structural integrity of the osseous tissue over time leading to dry bone weathering and breakdown (Junod and Pokines, 2014).

2.1. Factors affecting decomposition

2.1.1. Intrinsic

Human bone structure and type

Bone is made up of a combination of organic proteins and inorganic minerals. Collagen type I is dominant as it makes up almost 90% of the organic portion while the mineral component is primarily calcium hydroxyapatite (Child, 1995b; Collins *et al.*, 2002; Trueman & Martill, 2002; Baxter, 2004). Collagen and hydroxyapatite form complex bonds that ultimately provide bone with its structure, flexibility and strength (Von Endt & Ortner, 1984; Trueman & Martill, 2002). The decomposition or breakdown of a skeleton depends on the destruction of these two compounds (Child, 1995a; Trueman & Martill, 2002).

Bone is further made up of two types; cancellous and cortical bone which have different proportions of collagen and calcium hydroxyapatite (Nielsen-Marsh *et al.*, 2000). This will result in differential degradation of bone depending on the ratio of cancellous to cortical bone. Bones that have a thin cortical layer and a large portion of cancellous bone are more susceptible to breakdowns, such as the ribs or the epiphyses of long bones (Manifold, 2012), due to the numerous pores that are inherent in the cancellous bone structure. Pores in a bone will increase the surface area and thus more reactions can take place which will ultimately cause the rate of bone degradation to increase. Similarly, small bones have an increased surface area to volume ratio further increasing the rate of bone breakdown (Djurić *et al.*, 2011). Thus, small bones such as the carpals, tarsals and phalanges, degrade faster than larger bones such as the femur (Von Endt & Ortner, 1984; Manifold, 2012). Smaller bones also have a low representation due to the excavation process when the bones are often not identified and may be left behind or are easily dispersed by scavengers (Bello *et al.*, 2006; Djurić *et al.*, 2011; Manifold, 2012).

Age at death

In addition to the type of bone and its size, the degradation of the skeleton is also dependent on the age of the individual. Juvenile bones are smaller, have open epiphyses and the skeleton contains more cartilage when compared to adults (Bello *et al.*, 2006). Cartilage decomposition is more rapid than bone (Cunningham *et al.*, 2011; Manifold, 2012). Bello *et al.* (2006)

hypothesised that the preservation of a skeleton increases with age until all bones have fused. Senescent changes to bone (e.g., osteoporosis) in geriatric individuals can also lead to progressively porous bones which tend to rapidly degrade when compared to non-porous bones (Manifold, 2012). Similarly, chronic conditions can compromise the structural integrity and strength of an individual's skeleton thus leading to increased porosity and decreased mineralisation making a bone more susceptible to breakdown. Examples of chronic conditions can include; osteoporosis, rickets and cribra orbitalia (Manifold, 2012). Even though factors related to age could potentially also play a role in animal models when studying decomposition and diagenesis, these conditions will rarely present itself given the generally young age of animals used for such studies.

2.1.2 Extrinsic

Temperature is one of the most important environmental factors that affect the rate of decomposition, however, it is an extremely variable factor. The temperature of a region depends on its latitude as well as daily fluctuations, especially between day and night-time temperatures. As the temperature is so variable, decomposition studies in different areas could not be compared and thus a new method was developed; Accumulated Degree Days (ADD). ADD is used to compensate for differences in temperature between regions as it represents the heat units that are needed to drive various biological reactions during decomposition (Megyesi *et al.*, 2005; Adlam & Simmons, 2007; Simmons *et al.*, 2010).

The rate of decomposition increases with increasing temperature as dictated by Van't Hoff's rule. Van't Hoff's rule states that with each 10°C increase in temperature, the chemical reactions associated with autolysis and putrefaction increase by at least two times (Carter and Tibbett, 2008a; Vass, 2011). Once remains have reached the skeletonization stage of decomposition, the temperature still influences bone breakdown. An increase in temperature is associated with an increase in the loss of bone collagen (Von Endt & Ortner, 1984; Nielsen-Marsh *et al.*, 2000; Collins *et al.*, 2002; Hedges, 2002; Baxter, 2004). Collagen, in life, prevents the bone from breaking under tensile forces. However, in death, this collagen is destroyed making the bone more susceptible to damage such as cracking (Junod and Pokines, 2014).

Temperature fluctuations also contribute to the loss of moisture as well as the heating and cooling of a bone (Junod & Pokines, 2014). Primarily bone exposed to subaerial conditions experiences temperature fluctuations on the surface due to the increased exposure to the sun. As the temperature increases the bone expands but when it cools it contracts which over some

time will ultimately cause the bone structure to fail causing cracks (Von Endt & Ortner, 1984; Junod & Pokines, 2014). This forms one part of the weathering process. Weathering is defined by Behrensmeyer (1978) as the process by which the original micro-and macro-components of bone are separated from each other and destroyed by chemical (e.g., pH of the soil) or physical agents (e.g., solar radiation).

Temperature controls other factors such as insect and microbial activity (Campobasso *et al.*, 2001; Adlam & Simmons, 2007). Microbes such as bacteria perform optimally at specific temperatures. Most bacteria are active at 25° - 35° C (Campobasso *et al.*, 2001). Insects also increase their activity and their numbers in warmer temperatures. Decomposition proceeds much slower in winter resulting in decreased or absent populations of insects (Campobasso *et al.*, 2001).

Insects are the primary decomposers, and the rate of decomposition is established by their presence or absence, thus buried remains decompose slower as there is decreased insect access to the body (Simmons *et al.*, 2010). In 1968, Payne *et al.* analysed which insects were attracted to five buried pigs buried at depths between 50-100 cm. In the first stage of decomposition, the most prominent insects were ants followed by flies. However, recent studies suggest that most insects cannot reach bodies that are buried deeper than 30 cm (Campobasso *et al.*, 2001; Simmons *et al.*, 2010).

One of the insects that have been observed to alter buried bone would be termites. Termite tunnelling has been observed on buried skeletal remains but can often be mistaken for acidic soil corrosion (Pokines & Baker, 2014). Termite activity can be distinguished from acidic soil corrosion as the termite tunnels often form a stellate-shaped pattern and only sections of the bone will be affected whereas acidic soil corrosion affects almost all of the bone surface (Pokines, 2014; Pokines & Baker, 2014).

Additionally, species within the soil such as earthworms, moles, rabbits, meerkats, porcupines and mongoose can disperse the remains from their original location. This is especially significant for smaller bones such as the hand and foot bones such as the carpals, tarsals, metacarpals and -tarsals as well as the phalanges. This process is known as bioturbation (Pokines, 2014). These scavengers can also expose bones which can then be damaged further by gnawing. The gnawing of bone provides essential minerals and nutrients such as calcium, phosphate, sodium and potassium as well as fat. Rodents also use bone to grind down and sharpen their continuously growing incisors (Pokines, 2014).

Microbes refer to the combination of bacteria, fungi and protozoa (Trueman and Martill, 2002). In a burial environment, the microbial communities present in the soil are the primary catalysts for bone decomposition, as bone can provide these organisms with essential nutrients (Pokines & Baker, 2014). The Kingdom Fungi and bacteria are the two main microbes that damage bone (Hackett, 1981; Child, 1995; Jans, 2014). The damage is mostly microscopic in nature.

Microscopical focal destruction (MFD) is an example of this damage. Bacterial MFD are localised areas that have a grainy and porous appearance under the microscope. In these cases, the bacteria reorganise the bone material instead of destroying it (Hackett, 1981; Child, 1995; Jans, 2014; Pokines & Baker, 2014). Bacteria have also been observed to infiltrate the Haversian canals of bone and then destroy the bone from the inside out (Jans, 2014). Fungi create tunnels known as Wedl tunnels by absorbing bone collagen and hydroxyapatite (Hackett, 1981).

Bone is also a good source of nutrients for plants and due to the lattice structure of the trabecular bone, it can trap and store water. Thus, plant roots often grow around or through bone to absorb the nutrients, such as phosphate and nitrogen, as well as water. This process can be very destructive to bone (Gabet *et al.*, 2003). In addition, plant roots excrete acidic compounds that erode the surface of the bone to produce root etching. Root etching can be identified by the U-shaped and branching pattern that it leaves on the bone. However, root etching is not common in forensic cases and is most often found on archaeological bone (Dupras & Schultz, 2014).

Fungi can also create macroscopic changes as they can grow in large masses on the bone which can be observed. These masses also erode the bone surface from acidic compounds that fungi can produce thus making the bone surface appear rough and porous (Andrews, 1995; Pokines & Baker, 2014). Surface etching can also be caused by various soil fungi through their hyphae (Manifold, 2012; Dupras & Schultz, 2014; Pokines & Baker, 2014). However, fungi are often not present in burial environments as the majority of them are aerobic and a burial environment can become anaerobic very quickly (Dent *et al.*, 2004).

2.2. Methods of body disposal and their environmental influences

A perpetrator has a variety of options when discarding a body and the most common include deposition in subaerial (exposed on the surface), aquatic, or burial environments. Each of these environments has their own unique combination of factors that will influence the pattern and rate of decomposition.

Decomposition is significantly influenced by the presence of oxygen. Oxygen tends to increase the rate of decomposition which is widely available for surface remains and to some extent remains found in aquatic environments. However, in a burial environment the oxygen content is extremely limited and dependent on the type and texture of the surrounding soil (Dent *et al.*, 2004).

Buried remains tend to decompose about eight times slower than remains exposed on the surface (Rodriquez, 1997; Carter & Tibbett, 2008). The slower rate of decomposition in a burial environment is mainly attributed to lower ambient temperatures, as remains are protected from exposure to solar radiation, a lowered risk of scavenger activity and decreased insect access (Galloway *et al.*, 1989; Carter *et al.*, 2007; Forbes, 2008; Buekenhout *et al.*, 2018). However, the depth of the burial is also important as shallow burials (less than one metre) are more susceptible to temperature fluctuations and are more likely to be disturbed by scavengers and insects (Forbes, 2008; Manifold, 2012).

Naturally, soil is made up of various stratigraphic layers representing distinct soil compositions that have different chemical properties (Crow, 2008) For example, Zhang *et al* (2019) studied the difference in pH levels at three different soil levels in a hilly region in southwestern China. They determined that the top horizon was more acidic than the lower two layers due to ions such as calcium leaching down and out of the top and into the bottom soil layers. This top layer's pH was influenced by the climate and the topography. The lower two layers' pH was found to be affected by the type of rock from which the soil originated. In general, the topmost or surface layer has the highest organic content and as the depth of the soil gets deeper the soil generally becomes more clay-like with increased moisture and then finally the deepest layers consist of weathered rock and a rock layer. There also may be additional water layers beneath the soil layers (Fitzpatrick, 2008). However, this varies between regions due to the specific climatic conditions and geology found in the area. In addition, a grave would not conform to these stratigraphic layers due to backfilling as the soil layers get mixed forming a unique layer.

2.2.1. Formation of a cadaver decomposition island (CDI)

Soil is an extremely dynamic medium that can respond to any changes within the environment including the introduction of a decomposing cadaver (Carter & Tibbett, 2008). Decomposition of a body provides a flow of nutrients into the soil which results in the formation of a cadaver decomposition island (CDI) or grave soil. A CDI is associated with high microbial biomass, microbe activity, as well as changes in soil chemistry which include the increase of soil pH and

nutrient concentrations such as nitrogen, carbon and phosphorus (Carter *et al.*, 2007; Wilson *et al.*, 2007; Benninger *et al.*, 2008; Breton *et al.*, 2016). The CDI's chemistry and physical properties are not static and there is a change over time. At first, the fluids from the body destabilises the surrounding ecosystem which can be associated with the death of nearby vegetation (Carter *et al.*, 2007; Breton *et al.*, 2016). However, as decomposition progresses there is an increase in soil carbon and nitrogen which subsequently will promote the growth of new pioneer vegetation. The roots of this vegetation can grow in and around a skeleton which over time can cause damage to the bone (Pokines and Baker, 2014). Benninger *et al.* (2008) established that the change in soil chemistry from a cadaver could still be detected at least 100 days post-mortem whereas Fancher *et al.* (2017) indicates at least 5 years or more. These soil changes have also been utilised for estimating the PMI (Vass *et al.*, 1992; Benninger *et al.*, 2008; Anderson *et al.*, 2013; Fancher *et al.*, 2017).

The extent of the CDI is dependent on the mass of the cadaver and the soil texture (Carter & Tibbett, 2008; Breton *et al.*, 2016). Towne, (2000) observed the effect of different sized ungulates decomposing in a grassland ecosystem in Kansas. The presence of the adult and juvenile remains created barren areas around the carcass as the decomposition products would destroy any existing vegetation. However, the size of these patches was depended on the mass of the animal as these barren areas were larger around the adult carcasses. In addition, after two years there were still areas devoid of vegetation where the adult remains decomposed but there was no evidence of a disturbance near the juvenile areas (Towne, 2000). This is due to the adults having an increased volume of decomposition materials that can be produced and absorbed into the soil (Towne, 2000).

In soil, the skeleton is broken down by either the soil's chemical composition or by the microbes and plants that are present (Collins *et al.*, 2002; Jans *et al.*, 2004; Kendall *et al.*, 2018)(Collins *et al.*, 2002; Jans *et al.*, 2004; Kendall *et al.*, 2018) . In addition, but to a lesser extent, insects also contribute to skeletal damage. The chemical properties of the soil (such as pH, the presence of groundwater and the ion concentrations) have a larger impact on the dissolution of the hydroxyapatite while microbes enzymatically break down collagen (Nielsen-Marsh *et al.*, 2000; Trueman & Martill, 2002; Kendall *et al.*, 2018).

Collagen is usually insoluble but it, and its peptide bond to calcium hydroxyapatite, can be broken down through the process of hydrolysis which is primarily catalysed by microbial collagenases (Collins *et al.*, 2002; Forbes, 2008). Collagen breakdown can also be increased

with an increase in temperature and the extremes in pH (Gordan & Buikstra, 1981; Von Endt & Ortner, 1984; Collins *et al.*, 2002; Trueman & Martill, 2002). However, mineralized bone has extremely small pores that restrict the larger microbes from entering the bone to access the collagen which requires the dissolution of the hydroxyapatite through the process of diagenesis (Child, 1995; Nielsen-Marsh *et al.*, 2000; Collins *et al.*, 2002; Forbes, 2008). Diagenesis refers specifically to the chemical alteration of bone which involves the exchange of ions between the bone itself and the surrounding environment which can either be subaerial, burial or aquatic (Simmons & Cross, 2013).

2.2.2. Soil pH

The natural pH of a soil is regulated by several variables such as the geographical location and regional climate, the composition of the parent rock, the volume and type of organic content and its decomposition as well as the number of ions leaching out of the soil (Zhang *et al.*, 2019). The pH of soil can also change due to the effects of acid rain and groundwater, especially from pollution. Each layer or horizon of soil can have different pH values (Bloom & Skjellberg, 2012; Zhang *et al.*, 2019). A soil's pH constantly fluctuates. In the presence of a decomposing cadaver, soil pH can rapidly change as it progresses through the various stages of decomposition and degradation (Vass *et al.*, 1992; Gill-King, 1997; Carter *et al.*, 2007; Aitkenhead-Peterson *et al.*, 2012).

There have been contrasting results around how decomposition may alter the pH of the soil. Most studies that have analysed pH changes during decomposition have noted that the soil's pH initially increases and becomes alkaline (Vass *et al.*, 1992; Carter *et al.*, 2007; Wilson *et al.*, 2007; Aitkenhead-Peterson *et al.*, 2012; Anderson *et al.*, 2013). This is thought to be due to the increase in ammonia which is a product of putrefaction and the ammonification of nitrogen. This is followed by a subsequent decrease in pH after cessation of soft tissue decomposition (Wilson *et al.*, 2007; Carter *et al.*, 2010). In a study conducted by Haslam and Tibbett (2009), they suggest that each soil type has its own original microbial profile and such the pH response brought on by decomposition within each soil type (acidic, neutral and alkaline) will differ. There was an initial increase in the pH of all the soils but at varying degrees. The acidic soil had the largest change from a pH of 4.6 to 8.2 whereas the alkaline soils started at a pH of 7.1 and reached a pH of 8.2. The pH of the soils after 4 weeks then decreased to the starting pH. They hypothesised that soils became more acidic due to the increase in certain acidic compounds such as acetic acid and fatty acids which are decomposition by-products. In general, it was noted that the rate of soft tissue decomposition

was increased almost three times in sterile soils that were originally acidic compared to alkaline soils (Haslam & Tibbett, 2009).

In contrast, Tumer *et al.* (2013) noted that in loamy and organic soils the soil pH decreased, and it was suggested that this was due to the conversion of ammonia to ammonium which produces an increase of hydrogen ions into the soil. Soil composition before body deposition and the process of decomposition, therefore, needs to be considered.

The pH of the soil governs the preservation of calcium hydroxyapatite and to a lesser extent collagen. If the soil is acidic there are an increase in the hydrogen (H^+) ions which results in the breakdown of the hydroxyapatite mineral in bone (Nielsen-Marsh *et al.*, 2000). Thus, acidic soils are one of the major destructors of bone in a burial environment whereas neutral and alkaline soils tend to preserve it (Pokines & Baker, 2014; Buekenhout *et al.*, 2018).

Acidic soils can alter the macroscopic appearance of bone by a process of acidic soil corrosion. This causes the bone to have an irregular, pitted and porous appearance and it can sometimes form larger foramina or “windowing” at the thinner margins of the cortex (Pokines & Baker, 2014). This is due to acidic soils first degrading the hydroxyapatite creating pores and thus allowing microbial attack of the collagen (Nielsen-Marsh *et al.*, 2000; Manifold, 2012). The breakdown of collagen produces acidic compounds and thus decreasing the surrounding environments pH which further promotes the dissolution of hydroxyapatite (White & Hannus, 1983; Crow, 2008)

The pH of the soil regulates the type of microbial decomposers within the soil. Each microbe and its activity have an optimum pH range. Generally, acidic and alkaline soils are dominated by fungi whereas near-neutral soils give bacteria a competitive advantage (Haslam & Tibbett, 2009). Thus, the change in soil pH due to decomposition could render the original soil's microbes inactive while new microbes are introduced into the soil from the body (Breton *et al.*, 2016), which will further impact on the rate and degree of bone breakdown.

2.2.3. Soil texture and type

Soil texture can be broadly classified into three types: sand, silt and clay (Tumer *et al.*, 2013). The classification is established by the soils' particle sizes which also controls the soil's moisture content. Sandy soils are coarse-textured and consist of large particles. These soils are often well drained due to the large pores between the particles that allow for gas diffusion and water movement (Owens & Rutledge, 2005). The movement of the moisture also increases the mobility of gases and nutrients within the soil. These soils are generally associated with an

increased rate of decomposition (Carter & Tibbett, 2008). In extreme cases, a coarse textured soil can become very dry as water can easily escape, and this may result in desiccation of soft tissue (Fiedler & Graw, 2003). In other instances, this soil can promote the formation of pseudomorphs – these are the silhouettes of the cadaver outline imprinted in soil (Forbes, 2008). Pseudomorphs form due to the presence of phosphorus which is a by-product of decomposition. In coarse textured acidic soils phosphorus can attract metals in the soil such as manganese which may stain the outline of the remains (Dent *et al.*, 2004).

In contrast, clay soils are comprised of very small or fine particles comprising small pore sizes. Water, gases and nutrients cannot move freely in these soil types, which can lead to an increase in the moisture content and to the limited mobility of oxygen (Owens & Rutledge, 2005). This creates an anoxic environment, and the soil becomes dominated by anaerobic decomposers. Anaerobic decomposers are less efficient than aerobic decomposers which leads to a decrease in the rate of decomposition and can reach the extent of preserving soft tissues (Forbes *et al.*, 2005).

Decomposition in clay may often lead to the formation of adipocere, also referred to as grave wax, which tends to preserve remains (Fiedler & Graw, 2003; Forbes *et al.*, 2005; Carter & Tibbett, 2008; Schotsmans *et al.*, 2011; Breton *et al.*, 2016). Adipocere is formed by the hydrolysis and hydrogenation of free fatty acids from the body into saturated fatty acids which seeps into the surrounding tissue (Forbes *et al.*, 2005). It can appear as a white or grey, soft substance, but can become hardened over time (Fiedler & Graw, 2003). The basic factors that are needed for adipocere formation include any environment that promotes anaerobic bacteria activity. Bacterial enzymes, in a burial environment, are the catalysts for the essential reactions to produce adipocere. The optimal environment has been established to be a warm, moist, anaerobic and slightly alkaline environment (Takatori, 1996; Forbes *et al.*, 2005; Ubelaker & Zarenko, 2011). It was first thought that adipocere could only be produced if a body was deposited in water, but further studies proved that the body itself can contain sufficient moisture for adipocere formation (Forbes, 2008; Moses, 2012). Moses (2012) experimentally proved that adipocere could even form on de-fleshed dry bone as long as there were minimal fatty acids present and that the bone was in an environment that allowed for the growth of bacteria (Forbes *et al.*, 2005; Ubelaker & Zarenko, 2011). Adipocere, once formed, is highly resilient to degradation and thus aids in the preservation of tissues, found on cadavers and skeletons that are hundreds of years old (Mayer *et al.*, 1997; Fiedler *et al.*, 2009; Henderson *et al.*, 2015). The process of adipocere decomposition has not been fully established, however, it was determined

that it requires sufficient oxygen and possibly gram-positive bacteria (Pfeiffer *et al.*, 1998; Fründ & Schoenen, 2009).

The presence of water in the soil is an important influential factor for the decomposition of both buried soft tissue and bone. Water in soils stabilizes the temperature and provides some buffering capacity against the changes in pH, whilst acting as a solvent (Vass, 2011), therefore allowing for the dissolution and ion exchange (Nielson-Marsh *et al.*, 2000; Hedges, 2002; Jagers & Rogers, 2009). Microbes are essential decomposers in soil and their activity is dependent on the movement of water (Carter *et al.*, 2010; Delannoy *et al.*, 2016).

The rate at which dissolution, ion exchange and microbial activity occur is determined by optimum moisture content. If the soil is too wet there is low gas diffusion and a decrease in microbial activity. Similarly, if the soil is too dry there will be no medium for microbes to absorb nutrients and the body alone may not provide sufficient moisture (Carter *et al.*, 2010).

A study conducted by Jagers, and Rogers (2009) aimed to determine how moisture may affect the rate of bone and manner (morphological alterations) of degradation. Bones were buried in two contrasting sites; one had soil with a high moisture content and the other had a low moisture content, although both had a near-neutral pH. It was found that bones that were buried in the soil with high moisture had an increase in bone mass loss. This was suggested to be caused by the dissolution of minerals as well as the exchange of ions (Jagers & Rogers, 2009). However, Jagers and Rogers (2009) discovered that there were no morphological differences between the bones buried at the two sites after five months. The only variable that differed was bone mass (Jagers & Rogers, 2009).

The presence of groundwater is essential for the breakdown of bone as the majority of the processes and reactions take place in water. For example, the bone's incorporation of ions and organic content as well as the dissolution of hydroxyapatite and the removal of collagen (Hedges & Millard, 1995). The concentration of ions in the soil solution is one of the determining factors for the rate of bone dissolution. If a soil solution is saturated with calcium and phosphate ions this will inhibit hydroxyapatite dissolution. The surrounding groundwater and the bone will always establish an equilibrium in terms of ion movement. Additionally, an increase in the groundwater movement will increase the rate of dissolution due to the constant removal of ions from the surrounding soil as well as will erode the bone surfaces and increase bone porosity (Hedges & Millard, 1995).

2.2.4. Colour and depositional staining

Bone is naturally an off-white yellowish colour due to the presence of lipids, proteins and blood (Dupras & Schultz, 2014). Numerous environmental conditions can alter the colour of the bone; however, compared to other taphonomic processes the studies on bone staining in burial environments are limited (Huculak & Rogers, 2009; Pollock *et al.*, 2018).

Bone may first be stained by soft tissue decomposition, especially from the breakdown of red blood cells that can stain bone a reddish-brown colour through the process of haemolysis (Huculak & Rogers, 2009; Dupras & Schultz, 2014). Additionally, bones that are deposited on the surface are exposed to the sun and eventually become bleached white (Behrensmeyer, 1978) as compounds that give bone its colour, namely lipids, blood and proteins, are destroyed by the UV radiation. Only hydroxyapatite will remain which is naturally white (Dupras & Schultz, 2014). Thermal destruction, adipocere formation and marine water can also give the bone a white appearance (Dupras & Schultz, 2014).

A bone's porosity enables it to absorb tannins from the surrounding environment and can assist in deposition reconstruction (Dupras & Schultz, 2014; Pollock *et al.*, 2018). For example, a study conducted by Pollock *et al.* (2018) aimed to determine if bone staining could assist in reconstructing the environment for remains that have unknown origins as well as differentiating between modern and possible forensic cases from historical cemetery graves. The study established that the colour staining of the bones could be used to differentiate between different environments such as exposed or buried as well as if the remains had been exposed to wood and plant material. Surface remains were observed to have sporadic staining compared to the more uniform staining seen on the buried bones. Additionally, bones that had a uniform dark brown staining were from wooden coffins and are most probably of historic significance, but bones with sporadic soil staining or staining from plant material could potentially be of a forensic nature (Pollock *et al.*, 2018). Similarly, a study done by Huculak and Rogers (2009) attempted to determine if the colour of a bone could be used to distinguish whether the bone had been buried and then exposed on the surface versus first being on the surface and then subsequently buried. They looked at the colour of both the surface of the bone and in the cross-section. Decomposition influenced the surface colour but did not penetrate the cortex in both scenarios. The only difference between the two was seen in cross-section. Bones that were buried then exposed had a pattern of a lighter surface and a darker cortex while bones that were first exposed had a darker surface but a lighter cortex or a pattern of dark, light, dark. However, they suggested that more studies need to be done as they did not compare the colours of only

burial and only surface to these two settings. They did; however, notice that many of the bones that were exposed and then buried had evidence of fungal growth which were not seen on the bones that were buried and then exposed (Huculak & Rogers, 2009).

The colour of the soil depends on the amount of organic content; the darker the soil the more organic content there is within the soil and thus the darker a bone will stain (Dupras & Schultz, 2014). Organic content is not only dependent on a grave's or site's geographical region but it also differs with depth as the top layers have more organic material that is both fresh and decomposing that can stain bone (Fitzpatrick, 2008). Metal may also be present in the soil such as copper or iron which corrode and stain bone. Iron, if exposed to moisture and oxygen, will rust and if it encounters bone can turn the bone various orange, red and brown colours, whereas copper typically stains bone green. Bones may also appear green if there is presence of algae or moss (Dupras & Schultz, 2014; Pokines & Baker, 2014).

2.3. South African studies

The studies that have been done within a South African context have analysed the soft tissue decomposition rates of primarily surface remains and to some extent buried remains (Kelly, 2006; Myburgh *et al.*, 2013; Sutherland *et al.*, 2013; Marais-Werner *et al.*, 2017; Spies *et al.*, 2018; Finaughty, 2019; Spies *et al.*, 2020).

Studies done in the Western Cape have focused on the surface remains in the area of the Cape Flats. Studies have included the effects of scavengers on the rate of decomposition (Spies *et al.*, 2018); how clothing influences the decomposition rate (Spies *et al.*, 2020) as well as establishing the baseline data for decomposition rate in the area (Finaughty, 2019). From these studies, it was established that the only vertebrate scavenger in the area is the Cape Grey mongoose which accelerated decomposition and caused the bloat phase to be skipped (Spies *et al.*, 2018; Finaughty, 2019; Spies *et al.*, 2020). Clothing was seen to decrease the rate of decomposition which was attributed to the reduced scavenger activity as the clothing restricted their access (Spies *et al.*, 2020). The primary decomposition agents are insects, particularly *Calilliphora vicina*. Additionally, Cape town, unlike the rest of South Africa, has hot and dry summers which can produce natural mummies (Finaughty, 2019). Entomological studies have also been conducted in the Free State (Kelly, 2006). The study in the Free state observed the decomposition and insect succession of cadavers with clothes and wounds. There was insect colonisation on all the remains (clothed, wounded and controls) and the only difference was observed at the end of the study where the clothed remains had less skin (Kelly, 2006). In

Gauteng, the studies have aimed to determine the effect of specific factors on decomposition and calculating the PMI. Myburgh *et al.* (2013) focused on the use of total body score (TBS) and ADD for estimating the PMI of surface remains using the methods provided by Megyesi *et al.* (2005). It was determined that this method is not suitable in a South African context as decomposition was shown to be highly dependent on environmental factors other than temperature and that aspects related to regional seasonal variability should be accounted for (Myburgh *et al.*, 2013). Sutherland *et al.* (2013) also analysed surface remains and the influence of body size on the rate of decomposition. They determined that small and large bodies follow a different pattern of decomposition as smaller bodies decompose faster. There has been one study on the decomposition of buried remains and the stages of soft tissue decomposition to determine the post-mortem interval (Marais-Werner *et al.*, 2017).

Marais-Werner *et al.* (2017) conducted a study that aimed to assess and analyse the soft tissue decomposition patterns and rates of buried remains and compare them to previous surface studies (Myburgh *et al.*, 2013). The study took place over ten months starting in September 2014 and ending in June 2015. It involved the burial of 40 pig carcasses which were obtained from a local farmer and were buried – without any coverings such as coffins, plastic, or clothing - within 24 hours of their deaths. They were placed into separate graves of which the average dimensions and depth were 2.25m x 1.22m x 0.75m and 3m between each grave to prevent any cross-contamination between the carcasses.

Five pigs were routinely excavated over five different periods after burial: one week, two weeks, one month, three months and six months. During each excavation, the pig carcass left *in situ* (original position), without displacing or removing the carcass. Each pig carcass was photographed, observed and scored using the total body score (TBS) method (Megyesi *et al.*, 2005; Marais-Werner *et al.*, 2016).

Pigs numbered 2; 5 and 18 were scavenged by either domestic dogs, black-backed jackals or suricates; pigs 37 to 39 were waterlogged; and pigs 8 to 12 may have had compromised burials due to excessive rainfall before the date of excavation (Marais-Werner, 2016). As such these graves were not excavated. In total, 25 pigs were analysed and included in the study of which 20 were males and five females.

The first interval was 7 days and the average TBS for the five pigs was 9.6. The carcasses were in the early stages of decomposition which included bloating, purging of decomposition fluids and skin slippage. The limbs of two pigs showed signs of bloating which was in contrast with

the Megyesi *et al.* (2005) study. There were however differences in colour as two of the pigs were still pink with no discolouration while the other three had a darker purple discolouration. This suggested that the microenvironment of a grave is important and that there can be differences in decomposition within a single area.

At 14 days the average TBS was 13.4 as there was the sagging and collapse of the abdomen which formed a secondary depression. The only pig that showed evidence of intestine protrusion was pig 25 during this time interval. In addition, pig 21 had evidence of maggot activity and an increased burial temperature (Marais-Werner *et al.*, 2016). An average TBS of 17.4 was reached at 33 days after burial. It was noted that the rate of decomposition was beginning to slow down at this phase. There was also evidence of adipocere, especially around the abdominal region with black soil. It was noted that adipocere slows the rate of decomposition and can lead to the preservation of the remains. Ninety-two days after burial there was partial skeletonisation of the heads, leathery skin and disarticulated hooves which provided an average score of 20.4. The lungs and intestines of pigs 28 and 32 also seemed to have been preserved while the other three pigs' organs had liquified. The last excavation took place 183 days after burial when the remains had reached the advanced decomposition stage. All the pigs had developed adipocere and pig 7 had evidence of insect activity. The remains never reached the extremes of skeletonisation. The greatest change of decomposition, for the buried remains, was during the seven- and 14-day intervals which coincided with early decomposition and then began to slow after a month (Marais-Werner *et al.*, 2016). These changes were initially most prominent in the head and neck.

These scores were then compared to the surface study by Myburgh *et al.* (2013). Both surface and burial decomposition were noted to follow a sigmoidal curve where there was a fast rate of decomposition during early decomposition followed by a plateau phase. However, the rate and the TBS values were much higher for the surface when compared to the buried remains. This establishes that the rate of decomposition of the surface remains was faster than the buried remains but that the stages of decomposition between the two were similar (Marais-Werner *et al.*, 2017). Additionally, the buried remains were always in a state of moist decomposition whereas the surface remains became desiccated and almost reached skeletonisation

CHAPTER 3: MATERIALS AND METHODS

3.1. The site

The study was conducted at the Forensic Anthropology Body Farm on the Miertjie le Roux Experimental Farm, Gauteng Province (25°47'20.2''S; 28°32'34.3''E). The farm is about 560 hectares in size and belongs to the Faculty of Natural and Agricultural Sciences of the University of Pretoria.

It is located on the central Highveld plateau of South Africa. This region is classified as warm and temperate with dry, short, cold winters and hot, long summers (Kottek *et al.*, 2006). The average temperatures range from 28°C in the summer to 12°C in the winter. The average rainfall is around 691 mm per year. The rainfall occurs mostly during September through February which correlates to Spring and Summer. The vegetation consists of primarily sour grassland and bush veldt (Turner & Wiltshire, 1999; Marais-Werner, 2016).



Figure 1: Map of the site. The green block indicates the exact location of the cemetery

3.2. The sample

Sus scrofa domesticus (domestic pigs) were used as the sample for this study. Pigs have been widely used as a human analogue in decomposition studies as pigs - like humans - are omnivorous, their skin resembles that of humans, they have analogous fat distribution and anatomy, and they have similar intestinal flora (Goff, 1993; Campobasso *et al.*, 2001; Schoenly *et al.*, 2006; Schultz *et al.*, 2006; Reeves, 2009).

The pig remains that were used in this study were buried and analysed in a separate study that was conducted by Marais-Werner *et al.* (2017). The 39 pigs were donated by two farmers after

they had died of natural causes. They were then buried within 24 hours after the death of which the dates are known. The average weight of the pigs ranged from 45 – to 80 kg.

Each pig was placed into a burial pit at a depth of 0.75 m and each pit was separated by 3 m to ensure that there was no cross-contamination between the pigs (Marais-Werner, 2016). The pig carcasses were excavated and observed *in situ* during the original study, the carcass was not disturbed or moved, and the grave was backfilled immediately after scoring was done. The pigs remained buried until excavation started for this study in 2021. Ethical clearance to use the remains and transport them was obtained from both the University of Witwatersrand Animal Research Ethics Committee (Ethical clearance certificate numbers: 2020/06/07/O; Appendix) and the University of Pretoria (Ethical clearance certificate numbers: 543/2020; Appendix).

3.3. Excavations

The skeletal remains were excavated using standard archaeological techniques which included the use of brushes, hand shovels and trowels (Dirkmaat & Adovasio, 1997; Hunter & Cox, 2005; Dupras *et al.*, 2006). Systematic excavation of soil layers at 20 cm intervals was done. This ensured that additional features, such as faunal or floral activity, that might have had an impact on the taphonomic alterations, could be fully documented.

The remains were exposed and observed *in situ* to allow for the detailed documentation of the surrounding gravesite features, which included soil staining, bioturbation, any effects on the burial environment or remains caused by insects, rodents or carnivores and possible intruding vegetation. Scaled colour photographs were taken of the grave before excavations as well as the skeleton *in situ*. The skeletal remains were then removed, sided on site, and placed into sealable plastic bags. Each bag was labelled with the respective grave number. The remains were then transported to the University of the Witwatersrand for analysis.

Due to the potential contact with pathogens, safety precautions were taken. Each person that excavated were required to wear PPE – personal protective equipment (double gloving, dust mask with filter, and eye protection). After each day, the equipment was disinfected with bleach. Additionally, COVID-19 protocols were put in place which included sanitising all the equipment before handling and maintaining social distancing during excavation.

3.4. Skeletal taphonomy analysis

Soft tissue decomposition is typically analysed using a variety of stages reflecting the fresh, early, advanced and skeletal phases (and variations of these) associated with decomposition

(Galloway *et al.*, 1989; Wilson *et al.*, 2007). Morphological characteristics associated with each stage are used to identify the specific decomposition stage, however, the body of an individual or pig does not decompose uniformly which necessitates the separate scoring of each region (head and neck, trunk, limbs) and thus the total body score (TBS) system was developed (Megyesi *et al.*, 2005). The TBS method assigns points to the three regions (head and neck, trunk, limbs) depending on the decomposition characteristics present, using a scoring system. The points are then added together to provide an overall TBS for that individual (Megyesi *et al.*, 2005; Adlam & Simmons, 2007; Cross & Simmons, 2010; Myburgh *et al.*, 2013; Marais-Werner, 2016).

In this study, a modification of the TBS system was used to analyse the taphonomy of each pig. A modification of this method was required as the original method primarily scores the soft tissue decomposition to indicate the progression of decomposition and their association with the PMI. This could not be applied to the skeletal remains in this study as the progression of bone breakdown is not sequential and testing for potential correlation between TBS scores and PMI was not included in this study. The skeletal breakdown is heavily dependent on the environmental conditions rather than time lapsed therefore requiring amendment of this scoring method.

The body regions of the TBS method were used in this study. These regions include the head and neck (skull and cervical vertebrae), trunk (shoulder and pelvic girdles, ribs, thoracic and lumbar vertebrae), forelimbs (humeri, ulnae, radii, and foot bones) and hindlimbs (femora, tibia, fibula and foot bones). Each skeleton was first inventoried to establish the skeletal completeness. Initial observation of the remains indicated the presence of six taphonomic alterations which included depositional staining, adipocere formation, bone weathering, acidic soil corrosion, plant activity and animal activity. The presence and degree of each of these six taphonomic alterations were scored for each of the TBS regions. To get the overall TBS for each carcass, the most common score from each region for each taphonomic alteration was used.

3.4.1. Skeletal preservation

A full skeletal inventory was recorded to establish the skeletal completeness as well as the general preservation of the remains. Each bone was scored as being absent, and where present scored into one of five categories; 1: present and complete, 2: present and fragmentary; 3: present and unfused; 4: present, unfused, and fragmentary; 5: postmortem absent.

A percentage of skeletal completeness was estimated to get an idea of the preservation. This required a total bone count per skeleton which was simplified by counting the skull (which consists of several skeletal elements e.g., individual cranial bones and the mandible) as one element and excluding epiphyses in the overall count. The sample was comprised of juvenile pigs and thus there were multiple unfused epiphyses that were not included in the overall count. Rather the presence and degree of completeness of the diaphysis were used to record the completeness and general preservation of the limbs. This was done to account for unnecessary inflation caused by individually fragmented or missing skeletal elements. To calculate the percentage completeness of each pig the total skeletal count was divided by 202 as an average pig skeleton has around 202 skeletal elements (Aspinall *et al.*, 2020).

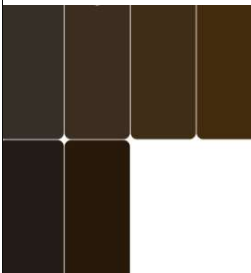
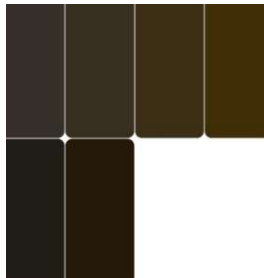
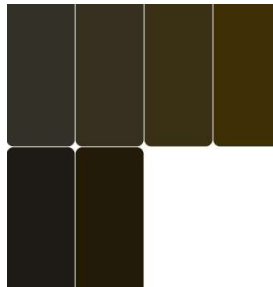
3.4.2. Depositional soil staining

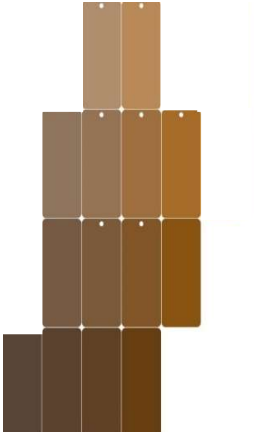
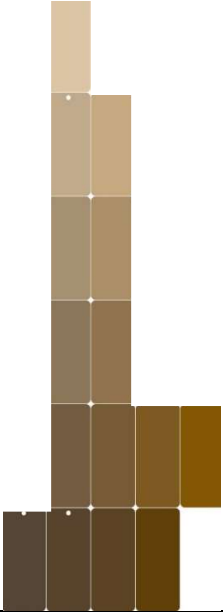
The perception of colour is subjective which necessitated the development of a standardised method such as the Munsell® Colour Chart when describing and scoring colour differences. The standard analysis for soil colour is the Munsell®'s Colour Chart (Jaggers & Rogers, 2009; Dupras & Schultz, 2014; Bloch *et al.*, 2021). This chart uses three values to describe a colour which is the hue, value and chroma. The hue is the overall colour and is red, yellow, blue, green, and purple with a variety of combinations. The value indicates how dark or light the colour is and the chroma indicates the colour's intensity (Bloch *et al.*, 2021). This chart includes a chart developed specifically for soil colour analysis called the Munsell® Soil Colour Chart which has nine separate hue pages that incorporate the red, yellow and grey hues.

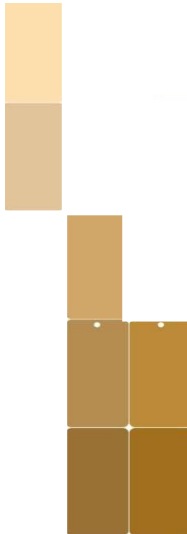
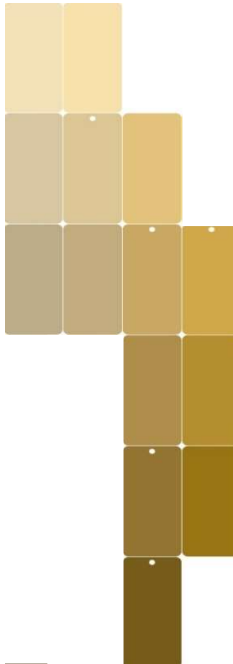
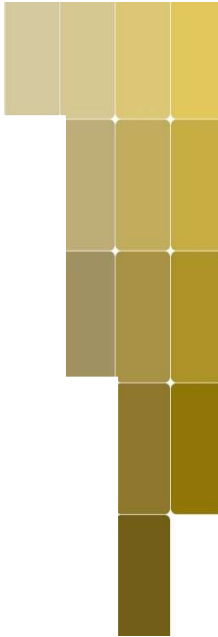
A Munsell Color Chart app (version 1.0.1.1) that was developed by KGSc and is available for free on the Google Play Store for Android was used to score the skeletal remains. The only hues that were observed were 7.5 YR, 10YR, 2.5 YR and 5Y. Unfortunately, there are no associations between the Munsell® Soil Colour Chart's common colour names and the Munsell Color Chart application. Thus, for this study, and in an attempt to simplify the scoring and capturing of the data the following colour categories were grouped together; black, dark brown, brown, yellowish-brown, yellowish-grey and greyish brown and were assigned a score from one to six. Each colour category had a corresponding range of colours and codes unique to the Munsell Soil Color Chart application (Table 1). These categories and the associated colour chips from the Munsell Soil Color Chart application was established by the investigator as they were the most common colours observed in this sample.

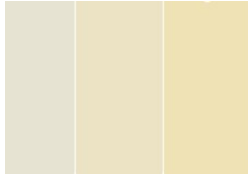
Each skeletal region (head and neck, trunk, upper and lower limbs) was scored with an overall colour from one of the six categories based on the most prominent colour seen on the bones of a particular region. The TBS was established using the most common colour throughout the regions.

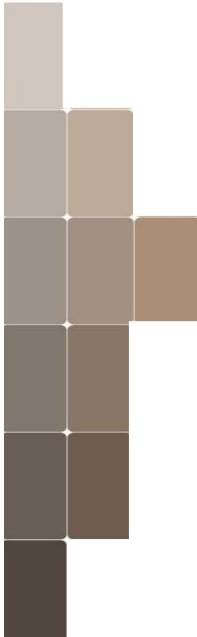
Table 1: Munsell colour codes from the app and the scores particular to this study (Munsell colour chart, 2016)

Score	Associated Munsell values			
	7.5 YR	10 YR	2.5 YR	5Y
1: Black				
	N1	N1	N1	N1
2: Dark brown				
	2/1; 2/2; 2/3; 2/4 1/1; 1/2	2/1; 2/2; 2/3; 2/4 1/1; 1/2	2/1; 2/2; 2/3; 2/4 1/1; 1/2	2/1; 2/2; 2/3; 2/4 1/1; 1/2

3: Brown				
	$6/4; 6/6$ $5/3; 5/4; 5/6; 5/8$ $4/3; 4/4; 4/6; 4/8$ $3/2; 3/3; 3/4; 3/6$	$8/3$ $7/3; 7/4$ $6/3; 6/4$ $5/3; 5/4$ $4/3; 4/4; 4/6; 4/8$ $3/2; 3/3; 3/4; 3/6$	$6/3; 6/4$ $5/3; 5/4$ $4/3; 4/4$ $3/3; 3/4; 3/6$	$7/3$ $6/3$ $5/3; 5/4$ $4/3; 4/4$ $3/3; 3/4$

4: Yellowish- brown				
	7/3; 7/4	9/4 8/4 7/6 6/6; 6/8 5/6; 5/8	9/3; 9/4 8/3; 8/4; 8/6 7/3; 7/4; 7/6; 7/8 6/6; 6/8 5/6; 5/8 4/6	8/3; 8/4; 8/6; 8/8 7/4; 7/6; 7/8 6/4; 6/6; 6/8 5/6; 5/8 4/6

5: Yellowish grey				
		9/2 8/1; 8/2	9/1; 9/2 8/1; 8/2 7/2 6/2	9/1; 9/2; 9/3

Greyish brown				
	8/1 7/1; 7/2 6/1; 6/2; 6/3 5/1; 5/2 4/1; 4/2 3/1	7/1; 7/2 6/1; 6/2 5/1; 5/2 4/1; 4/2 3/1	7/1 6/1 5/1; 5/2 4/1; 4/2 3/1; 3/2	8/1; 8/2 7/1; 7/2 6/1; 6/2 5/1; 5/2 4/1; 4/2 3/1; 3/2

3.4.3. Adipocere

The scoring method for adipocere formation, acidic soil corrosion, plant and animal activity could not be found and thus the scores were created for this study. Adipocere is a wax-like substance that can form during cadaver decomposition. It can appear as greyish white on both soft tissues and the bone (Fiedler & Graw, 2003; Moses, 2012). Adipocere in this sample was noted as a yellowish-grey colouring on the bone. Each skeletal region was scored as 1: absent; 2: coverage on less than half of the region; 3: coverage on more than half of the region.

Each region is made up of multiple skeletal elements for example the forelimb is made up of the humerus, radius, ulna, carpals, metacarpals, and phalanges. Each bone can develop adipocere and thus each bone was assessed individually. Each bone was scored as either being devoid of adipocere (Fig. 1), presenting with partial adipocere formation (Fig. 2) or excessive adipocere formation (Fig. 3). A score was then assigned to the entire region, for example, if only the humerus had evidence of partial adipocere formation a score of 2 was given as the humerus makes up less than half of the region's skeletal elements. A score of 3 was provided if there was excessive adipocere formation present on multiple bones of that region.



Figure 2: Adipocere is absent.



Figure 3: Partial adipocere formation as indicated by the green circles.



Figure 4: Excessive adipocere formation on multiple bones.

3.4.4. Bone weathering

Bone weathering includes the chemical and mechanical changes to the bone which often leads to the bone's physical destruction. This can be observed as bone-cracking, warping or general erosion (destruction of a bone's surface layers) (Pokines & Baker, 2014). Bone weathering was scored into one of six categories following a modification of Behrensmeyer's and Ross and Cunningham's (2011) scoring criteria. Behrensmeyer's (1978) stages are the standard for scoring the subaerial weathering of skeletal remains (Junod & Pokines, 2014), however, these

stages are not applicable for buried remains (Ross & Cunningham, 2011; Buekenhout *et al.*, 2018). Ross and Cunningham (2011) did not observe Behrensmeyer's stages in their buried sample and thus they created their own stages for their sample. These stages were still not fully applicable to this study as the stages were associated with long PMIs of up to 30 years, nevertheless descriptions from Ross and Cunningham (2011) were used for the purposes of this study but were amended slightly for a more detailed description of this pig sample.

Behrensmeyer (1978) scored the bones using the most extensive stages that were present on more than 1 cm² of bone. In this study, all the bones of a region were analysed and given a score from one to six and the highest value was used to score the entire region. To establish the TBS per pig the most common stage between the regions was used. For this study the following scoring criteria were utilised:

- 1: No weathering, the bone was intact with a smooth surface. This score was not observed on any of the remains.
- 2: Rough bone surface as cortical bone begins to break down with delamination of the cortical bone which is when the top layers of bone lift and flake. "Marbling" of the bone surface may be present which is the flaking of cortical bone, especially on the diaphysis creating very fine cracks in a marble pattern (Figs. 4 & 5).
- 3: Weathering penetrates the inner cavities with partial exposure of the trabecular bone and longitudinal cracking may be present (Figs. 6 & 7).
- 4: Extensive exposure of the trabecular bone which is beginning to break down (Figs. 8 & 9).
- 5: Bone is extremely fragile and fragmented (Figs. 10 & 11).
- 6: Complete disintegration/bone shadow, which was noted during excavation.



Figure 5: Stage 2. Bone has a slightly roughened surface with minimal cortical bone exposure.



Figure 6: Stage 2. The delamination of the cortical bone indicated by the red box. The bone is lifting from the bone surface with some flaking.



Figure 7: Stage 3. Longitudinal cracking indicated by the red arrow with partial trabecular bone exposure at the distal end of the radius.



Figure 8: Stage 3. Partial trabecular bone exposure within the red circle.



Figure 9: Stage 4. Extensive trabecular bone exposure of both the proximal and distal ends of the femur.



Figure 10: Stage 4. Extensive exposure of the trabecular bone which has started to break down.

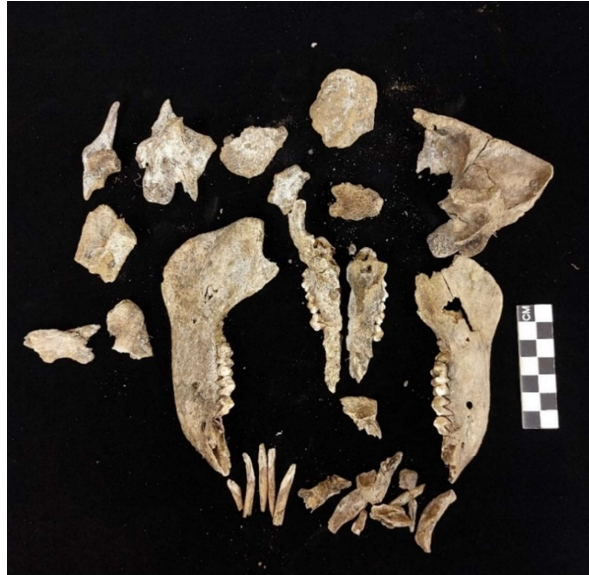


Figure 11: Stage 5. The skull is extensively fragmented.



Figure 12: Fragmented ribs indicating score 5.

3.4.5. Acidic soil corrosion

Acidic soil may corrode bone, giving it a pitted and porous appearance (Pokines & Baker, 2014). The main characteristics of acidic soil corrosion are described by Pokines and Baker (2014) and were used as the basis for the scoring method used in this study.

Acidic soil corrosion was scored as either; 1: absent; 2: partial acidic corrosion was identified as a porous appearance on less than half the bone (Fig. 12) 3: extensive acidic corrosion was identified as a porous appearance on more than half the bone in addition to the presence of “windowing” (Figs 13 & 14). TBS per pig was established using the most common score.



Figure 13: Partial acidic soil corrosion. The bone only has a scooped and pitted appearance on the proximal end as indicated by the red circle.



Figure 14: Extensive acidic soil corrosion as more than half of the bone has a pitted and porous appearance.



Figure 15: Example of acidic soil causing "windowing" of a rib.

3.4.6. Plant activity

Roots often affect bone surfaces in the form of root staining and etching and may even penetrate the cortical bone to enable growth into the trabecular bone (Pokines & Baker, 2014). Plant activity was scored into one of four categories. These were created from descriptions from Pokines and Baker (2014) and ranged from absent to severe destruction. Plant activity per region was scored as either being 1: absent, 2: root etching/staining (Fig 15 & 16); 3: present with partial macroscopic damage (Fig. 17A); 4: present with extensive macroscopic damage that could have caused complete destruction (Fig. 17B). TBS for each pig was established using the most common score per region.



Figure 16: Root etching on the proximal end of the femur as indicated by the red circle. U shaped grooves with a branching pattern (Pokines and Baker, 2014).



Figure 17: Root staining/etching with branch like patterns on the internal surface of the rib.

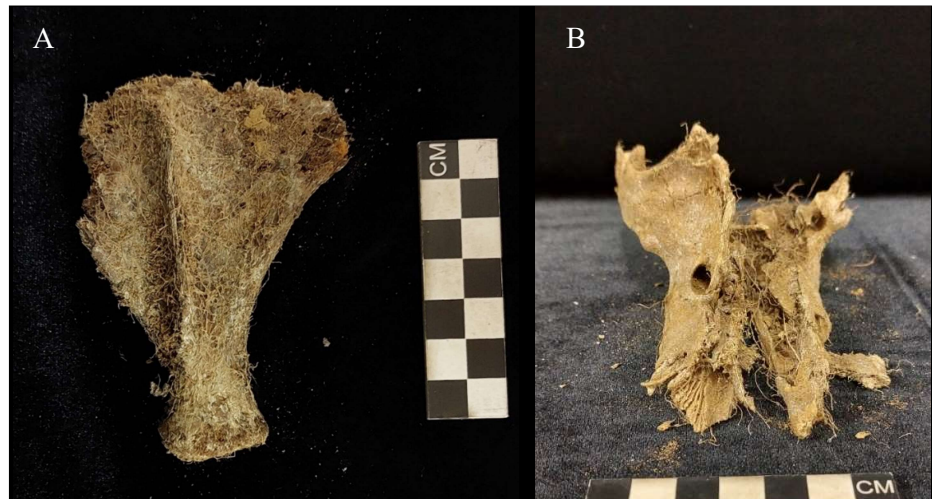


Figure 18: Plant root damage. A) Plant roots cover the surface of the bone. The roots are attached to the bone surface. B) Plant roots have broken the cranium in half.

3.4.7. Animal activity

Any form of animal activity was noted as either 1: absent or 2: present. This included any insect activity such as termite damage, scavenger activity (illustrated as bone gnawing), and bioturbation which was determined present depending on the location of the remains and if they move from the original margins of the gravesite (Pokines, 2014). Displaced bones were noted, and their position was documented by measuring the distance of the remains from their original anatomical position within the grave matrix.

3.5. Soil analysis

3.5.1. Soil sample collection

Soil samples were collected in sterile sample tubes from each grave from three different levels: 10 cm from the surface, at the level of the carcass, and 10 cm below the carcass. Each sample was analysed for soil pH and the percentage organic content. All soil analyses were done at the University of Pretoria's Department of Geography, Geoinformatics and Meteorology.

The first 10 cm samples were collected to determine how backfilling and the mixture of the natural occurring stratigraphic layers would influence soil composition. The samples taken at the level of and 10 cm below the carcass would indicate the effects that decomposition might have on the soil composition. Control samples were collected 10 m from the burial site. A trench of 90 cm was dug, and the samples were taken 10 cm from the surface, 70 cm which was the average depth of all the burials and at 80 cm.

3.5.2. pH analysis

The pH of the soil was determined by creating a soil slurry which requires mixing soil with distilled water in a 1:1 ratio. The soil was first sieved in a 2 mm sieve and 5 g of the sieved sample was collected and added to a clean container. Using a clean glass pipette, 5 ml of distilled water was added to the soil. The solution was stirred for 10 seconds and allowed to stand for 20 minutes. During this time the pH meter (Eutech instruments Ecoscan pH 6) was calibrated using buffer solutions of pH 4.00 and pH 7.00. The pH of the soil slurry was then determined by placing the electrodes into the solution at the soil-water interface and stirring constantly until the reading stabilised (Soil and Plant Analysis Council, 2000).

3.5.3. Organic content analysis

The organic content of the soil was determined using the loss-on-ignition method which involves burning the soil at high temperatures and comparing the weights of the soil before and after burning. The organic content of the soil will burn away and the difference in weights can be used to calculate the percentage organic content. A soil sample of 10 g was weighed out and placed into a crucible that was able to withstand 1500 °C. The sample was heated to 105°C in a drying oven for four hours. The sample was then weighed to the nearest 0.01 g and then subsequently transferred to a muffled furnace to be ashed at 400°C for four hours, after which of the sample was weighed again to the nearest 0.01 g. The loss in weight between the two measurements was used to determine the percentage organic content in the sample. The following formula was used in calculating the percentage value: $\% OC = \frac{[(W_{105} - W_{400}) \times 100]}{W_{105}}$ where W_{105} is the weight of the soil at 105°C and W_{400} is the weight of soil at 400°C (Soil and Plant Analysis Council, 2000).

The depth of each grave was determined using a measuring tape at the level of the trunk. The tape cylinder was placed at the level of the surface and then stretched down until it touched the ribs of the carcass and the resulting measurement read off. For grave positioning, a score was created depending on the grave's position within the cemetery. The cemetery was divided into four quadrants and the score for each grave depended on its location in these quadrants (Fig. 18). In addition, vegetation changes and grass species were noted and identified that were growing above the surfaces of the graves.

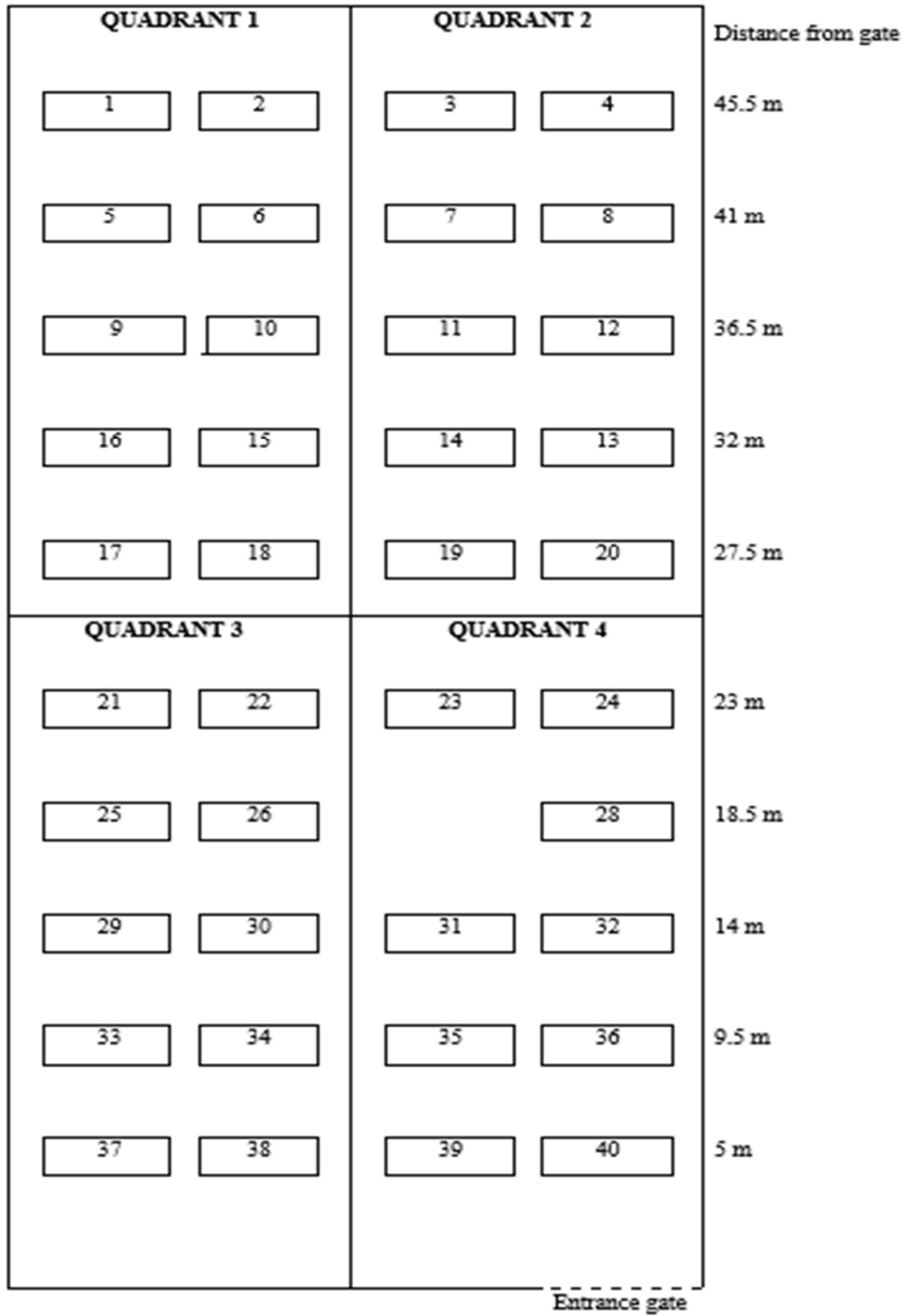


Figure 19: Cemetery layout.

3.6. Statistical analysis

The data was statistically analysed using the program IBM SPSS version 28 (Statistical Package for the Social Sciences). Inter- and intra-observer reliability was conducted on the scoring criteria used in this study. This was done by the primary researcher and a second observer using the scoring sheet provided. Four randomly chosen pigs were scored by each observer and the error rates were determined using a weighted Cohen's Kappa coefficient.

Initial tests were first run to determine if there was a linear relationship between the variables. This was done using scatter plots. Additionally, the data were tested for normality as well as if there were any outliers. These initial tests were to make sure that the assumptions of the subsequent tests were met. Spearman rank correlation coefficient tests were run given that the data were nonparametric. Three multiple correlation analyses were run for each region's score against each other, the taphonomic scores against the soil composition (pH and organic content), and the taphonomic scores against the burial environment (depth and the grave positioning).

To determine if there was a difference between the grave soil's layers with the controls, a student T test was done if the data was parametric and a Wilcoxon signed-rank test if the data was non-parametric. This was done to establish the effect that the carcass had on the grave soil. Additionally, an ANOVA (parametric) or a Kruskal Wallis (nonparametric) test was completed to assess any differences between the layers.

CHAPTER 4: RESULTS

4.1. Type, degree, and frequency of macroscopic taphonomic alterations

The sample consisted of 39 pigs that were originally buried between September 2014 and April 2015 (Marais-Werner, 2016). The excavations for this study started in March 2021 and ended in October 2021. The following taphonomic alterations could be observed on the sample: depositional soil staining, adipocere formation, bone weathering, acidic soil corrosion, plant activity and animal activity. The results outlined here will discuss the degree and frequency of each of the taphonomic alterations observed in the sample. For a breakdown of the individual scores for each grave refer to the appendix, Table 1.

Depositional staining, weathering and plant activity were the most common taphonomic alterations as they were observed on all the pigs (Table 2). Adipocere was also frequently observed with 36 (92.3%) pigs showing signs of adipocere formation. Animal activity was the least observed taphonomic alteration, being present in only four pigs (10.3%), whereas acidic soil corrosion was present in 29 pigs (74.4%).

Table 2: Overall presence of taphonomic alterations observed on the sample (n=39)		
	Absent	Present
Depositional staining	0	39 (100%)
Adipocere	3 (7.7%)	36 (92.3%)
Weathering	0	39 (100%)
Acidic soil corrosion	10 (25.6%)	29 (74.4%)
Plant activity	0	39 (100%)
Animal activity	35 (89.7%)	4 (10.3%)

4.1.1. Interrater reliability

Intra-observer error

A subsample of 4 pigs was scored by the primary investigator a month after data collection. Results suggested that the scores for depositional staining, adipocere formation, weathering, and plant activity were reliable (Table 3). The agreement for staining, adipocere formation and plant activity was substantial, while acidic soil corrosion was moderate using the definitions provided by Cohen. The agreement for weathering was almost perfect. The Kappa-value for animal activity suggested a 100% agreement as no animal activity was observed in this subsample.

Table 3: Intra-observer error using weighted Cohen's Kappa (n=4)		
Weighting	Kappa	P-Value
Staining	0.770	0.000

Adipocere	0.740	0.000
Weathering	0.821	0.000
Acidic	0.515	0.001
Plant	0.745	0.000
Animal	1.000	-

Interobserver

The comparison of the taphonomy scores between two observers had a moderate to a substantial agreement as the Kappa-values were above 0.5 (Table 4). These values are statistically significant as $p < 0.05$. The Kappa-value for animal activity had a perfect agreement between the observers, however this is to be expected seeing as though no animal activity was observed in the subsample. Staining, adipocere formation, acidic soil corrosion and plant activity were moderate, while weathering was excellent.

Table 4: Inter-observer error using weighted Cohen's Kappa (n=10)		
Weighting	Kappa	P-Value
Staining	0.595	0.000
Adipocere	0.694	0.000
Weathering	0.882	0.000
Acidic	0.500	0.001
Plant	0.524	0.000
Animal	1.000	-

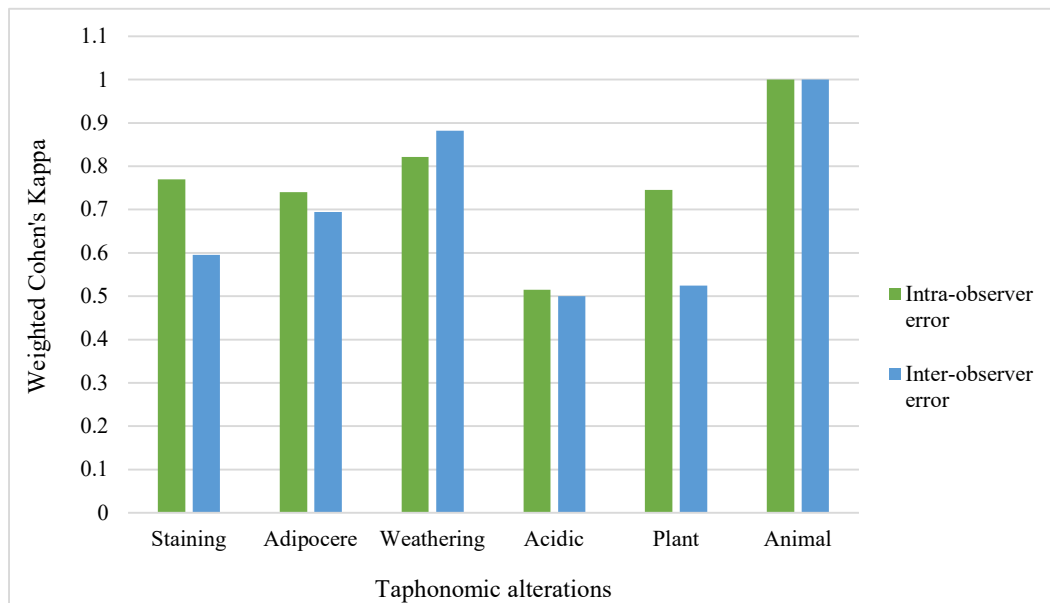


Figure 20: Histogram of the intra - and interobserver errors using a Weighted Cohen's Kappa

4.1.2. Taphonomic alterations

Skeletal completeness

The average percentage of skeletal elements recovered was 43.0%, with the maximum being 68.8% (grave 21) and a minimum of 11.4% (grave 33). The least recovered bones were the bones of the fore- and hindlimbs, specifically the metapodials, the thoracic, lumbar, and coccygeal vertebrae, as well as the fibula. Of the carpals, tarsals, meta-carpals, and -tarsals and phalanges only about 10 bones of 96 were recovered for each skeleton. The long bones and crania were the most often recovered. The details of the individual skeletal preservations are provided in the appendix, Table 2.

Depositional staining

Staining was separated into six categories based on the Munsell soil colour chart. These categories included black, dark brown, brown, yellowish-brown, yellowish-grey, and greyish brown. The most observed colours were dark brown (41.0%) and brown (46.2%) (Table 5).

Table 5: The frequencies of depositional soil staining observed per pig and for each of the skeletal regions

Score	Colour description	Sample (n=39)	Skeletal regions (n=232)
1	Black	0*	3 (1.3%)
2	Dark brown	16 (41.0%)	83 (35.8%)
3	Brown	18 (46.2%)	101 (43.4%)
4	Yellowish-brown	2 (5.1%)	11 (4.7%)
5	Yellowish grey	1 (2.6%)	4 (1.7%)
6	Greyish brown	2 (5.1%)	30 (12.9%)

*Black was not observed at the sample level as the most common colour observable throughout the skeleton was used to obtain the result per pig, which were more often associated with dark brown or brown.

Looking at the differences in the soil colour staining between different skeletal regions it was noted that the trunk presented with the darkest coloured soil staining compared to the other regions. The trunk was the only region presenting with black-coloured stains (n = 3), with the second most frequent soil colour staining observed being dark brown (n = 21) (Fig. 19). This seems to contrast with the head and neck as well as both the left and right limbs, which were primarily scored as brown (Fig. 19).

When comparing the left and right sides, the right side was generally darker than the left as the right limbs had more scores for dark brown staining (n forelimbs = 12; n hindlimbs = 14) (Fig. 19). The left side's limbs had a higher count for the light brown staining, including brown (n forelimbs = 20; n hindlimbs = 21), yellowish-brown (n forelimbs = 5; n hindlimbs = 0) and greyish brown (n forelimbs = 5; n hindlimbs = 7).

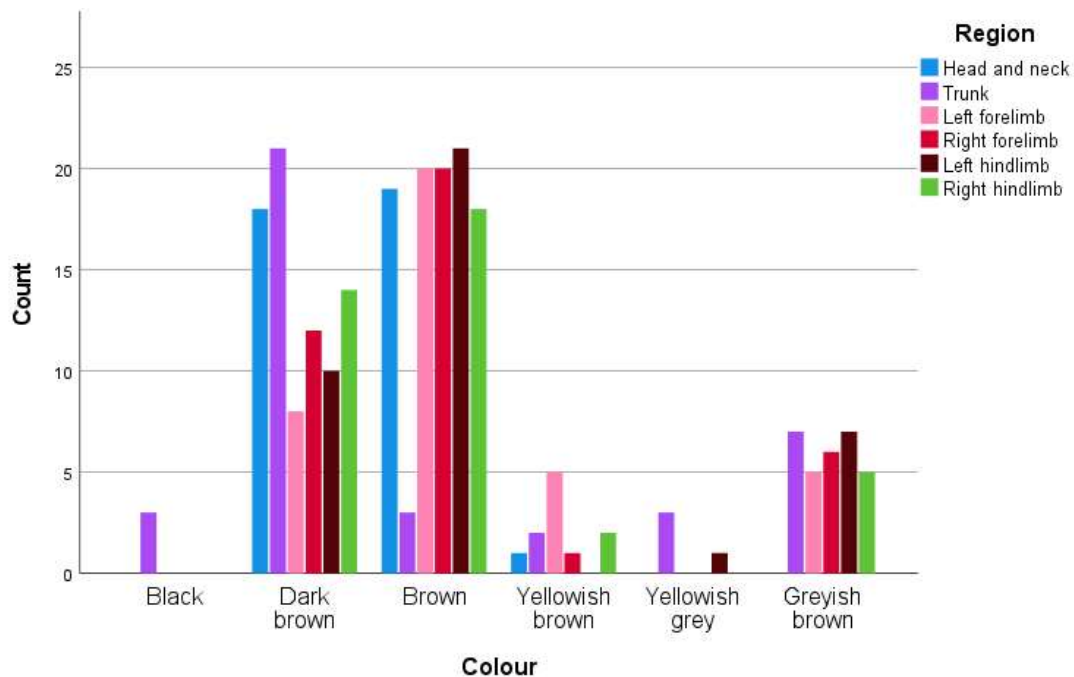


Figure 21: Bar graph depicting the different depositional stains per region.

Interesting staining was observed on four individuals. These were represented by light-red pinkish stains present on the crania of graves 13 and 17 as well as the right rib from grave 17 and the right femur from grave 26. These stains were not uniform and did not cover the bone surface.

Adipocere formation

Adipocere was present on 36 of the pigs. Eleven of these pigs (28.2%) had advanced adipocere formation (Table 6). In assessing adipocere per skeletal region a score of 2 (59.9%) was mostly obtained (Table 6) which indicates that less than half of the region was covered in adipocere.

Table 6: The frequencies of adipocere formation observed per pig and for skeletal regions

Score	Adipocere formation	Sample (n=39)	Skeletal regions (n=232)
1	Absent	3 (7.7%)	76 (32.8%)

2	Less than half of the skeletal region/skeleton affected	25 (64.1%)	139 (59.9%)
3	More than half of the skeletal region/skeleton affected	11 (28.2%)	17 (7.3%)

It was most prevalent in the regions of the trunk (n = 29 for score 2; n = 4 for score 3) followed by the head and neck region (n = 25 for score 2; n = 3 for score 3) (Fig. 20). It is important to note that adipocere also formed within the cranial vault and not only on the external surface of the bone (Fig. 4). Additionally, the hindlimbs more often presented with adipocere when compared to the forelimbs (n right = 22: n left = 22) (Fig. 20). Score 2, which represents adipocere on less than half the region, was observed on more of the right limbs (n forelimbs = 21, n hindlimbs = 22) than on the left. However, score 3, which is advanced adipocere formation, was scored on more of the left limbs than the right (n forelimbs = 3, n hindlimbs = 3) (Fig. 20).

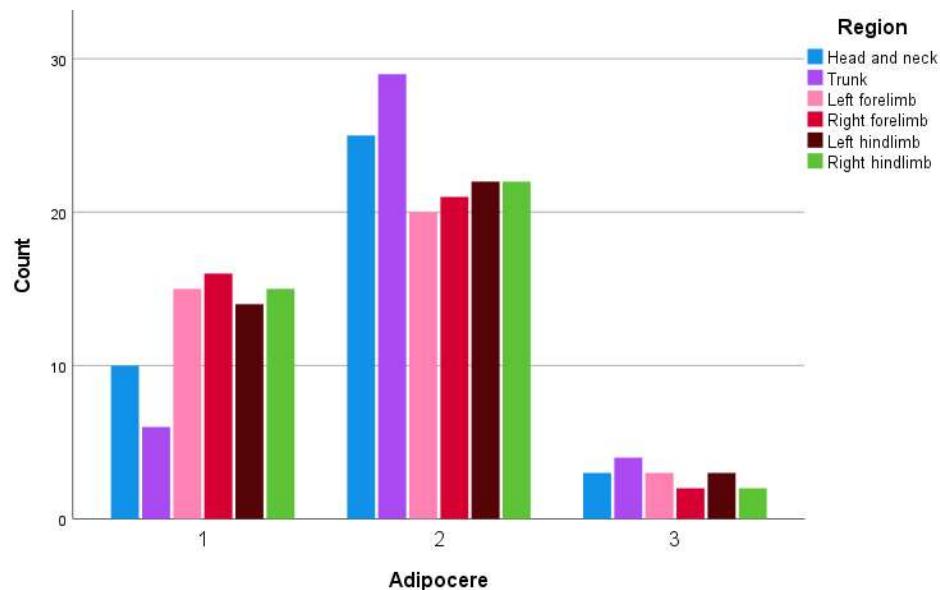


Figure 22: Bar graph depicting the adipocere formation per region.



Figure 23: Adipocere inside a cranial vault observed through the foramen magnum.

Weathering

Weathering was present throughout the sample and was commonly scored as a 3 (53.8% of the sample) which reflects weathering with partial exposure of the trabecular bone (Table 7). Stage 6, reflecting complete bone disintegration, was only observed in one of the skeletal regions (0.4%).

Table 7: The frequencies of weathering observed per pig and for each of the skeletal regions			
Score	Weathering	Sample (n=39)	Skeletal regions (n=232)
1	No weathering, the bone was intact with a smooth surface.	0	0
2	Rough bone surface as cortical bone begins to break down with delamination of the cortical bone.	4 (10.3%)	35 (15.1%)
3	Weathering penetrates the inner cavities with partial exposure of the trabecular bone and longitudinal cracking may be present	21 (53.8%)	97 (41.8%)

4	Extensive exposure of the trabecular bone which is beginning to break down	8 (20.5%)	64 (27.6%)
5	Bone is extremely fragile and fragmented	6 (15.4%)	35 (15.1%)
6	Complete disintegration/bone shadow	0*	1 (0.4%)

* Score 6 was not observed at the sample level as the most common score observable throughout the skeleton was used to obtain the result per pig.

Looking at the individual skeletal regions, the head and neck were most scored as a stage 5, representing bones that are fragile and fragmented (n = 20; Fig. 22). The area that showed the least amount of weathering (score 2) was the right forelimbs (n = 15; Fig.22), whereas the trunk was the only region that had a score of 6, representing complete disintegration. Figure 22 depicts that the left side was more weathered than the right side as the left side had a higher count for scores 3 and 4 (n forelimbs = 30; n hindlimbs = 33) compared to the right side.

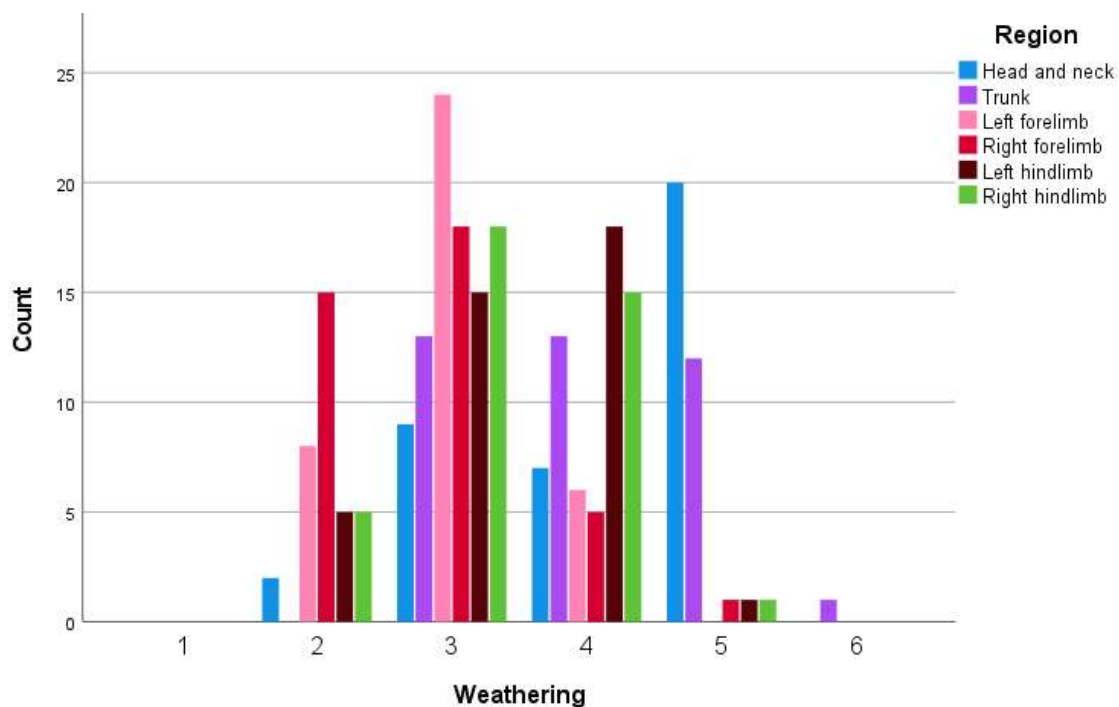


Figure 24: Bar graph depicting the weathering scores for each region.

Acidic soil corrosion

Acidic soil corrosion was present on 29 of the pigs (Table 8). Where present, acidic soil corrosion tended to affect less than half of the skeletal region assessed (18.5%).

Table 8: The frequencies of acidic soil corrosion observed per pig and for each of the skeletal regions

Score	Acidic soil corrosion	Sample (n=39)	Skeletal regions (n=232)
1	Absent	10 (25.6%)	163 (70.3%)
2	Less than half region/skeleton coverage	29 (74.4%)	43 (18.5%)
3	More than half region/skeleton coverage and windowing	0*	26 (11.2%)

*Acidic soil corrosion caused windowing of skull and corroded the astragali and thus were scored as a 3 and thus score 3 was present per region but was not representative of the whole skeleton.

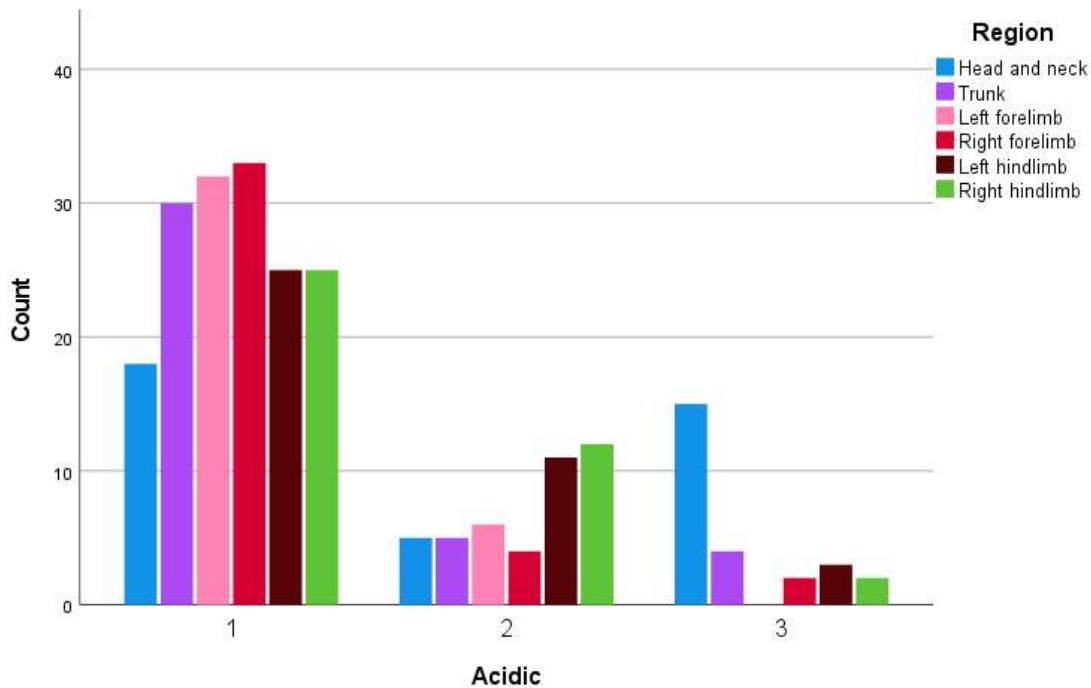


Figure 25: Bar graph depicting the acidic scores per region.

Acidic soil corrosion was commonly observed on the head and neck as well as the hindlimbs (Fig. 23). Corrosion on the head and neck was more extensive as 17 crania (43.6%) were scored

as a 3 (Fig. 23). They were characterised by the presence of windowing, especially on the lacrimal bones (Fig. 24). The hindlimbs mostly presented as score 2, which is represented by a scooped and corroded cortical layer that extends into the trabecular bone, of which the femur was most often affected ($n = 17$). The astragali of 12 graves also had a corroded appearance. Additionally, grave 40 had a rib that showed evidence of “windowing” (Fig. 25) as well as grave 36 had a circular indentation on the right scapula’s glenoid cavity



Figure 26: Common acidic soil corrosion appearance. Windowing on the lacrimal bones of the skull



Figure 27: "windowing" of the rib from grave 40 (a) Circular indentation on the right scapula (b).

Plant activity

All the remains were affected by plant activity (n = 39; 100%) (Table 9). Root infiltration causing partial macroscopic damage of the skeleton was present on the majority of the sample (n = 28; 71.8%) and skeletal regions (59.1%) (Table 9). Four pigs and 52 of the regions had extensive damage caused by plant activity.

Table 9: The frequencies of plant activity observed per pig and for each of the skeletal regions

Score	Plant activity	Sample (n=39)	Skeletal regions (n=232)
1	Absent	0	1 (0.4%)
2	Root etching/staining	7 (17.9%)	42 (18.1%)
3	Present with partial macroscopic damage	28 (71.8%)	137 (59.1%)
4	Present with extensive macroscopic damage	4 (10.3%)	52 (22.4%)

Extensive damage (score 4) was commonly observed on the head and neck region (n = 23; Fig. 26). Root infiltration into these crania was extensive leading to fracturing along the suture lines and damage to the cortical and trabecular bone, especially around the nasal bones (Fig. 27).

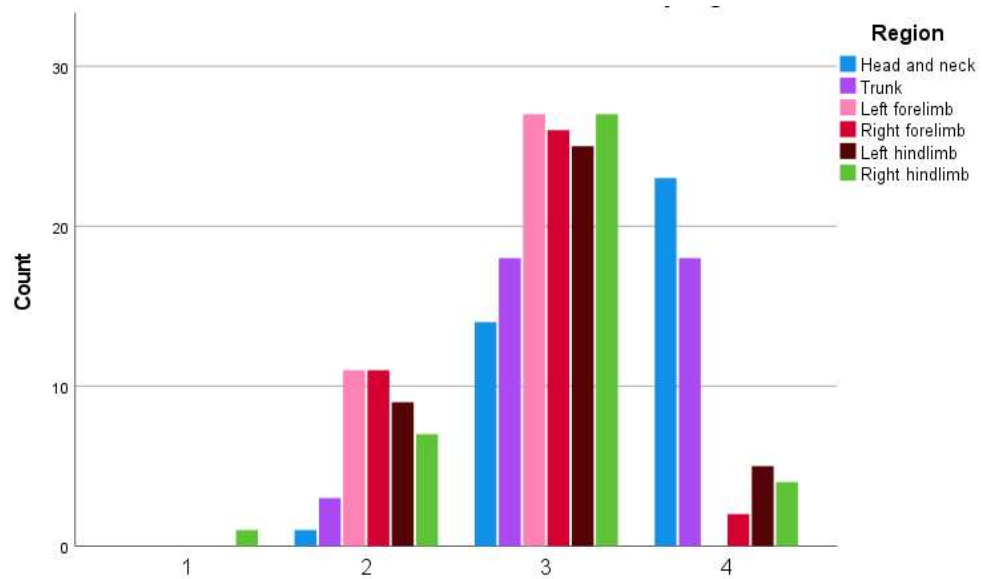


Figure 28: Bar graph depicting the plant activity scores per region.



Figure 29: The breakdown of the nasal bones and break along the suture lines.

Additionally, grave 39 had evidence of possible fungi formation on the left scapula. This was not scored as adipocere since it had a fuzz-like texture (Fig. 28). The growths were on top of the cortical bone with a very uniform and circular appearance.

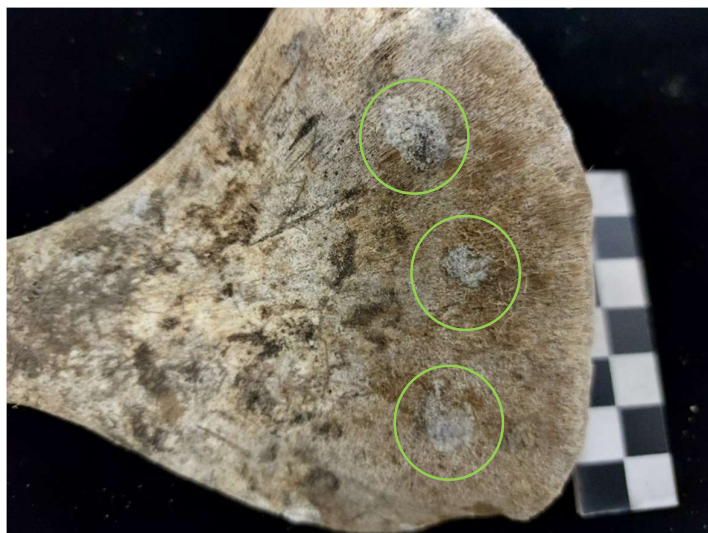


Figure 30: Fungi formation on the left scapula from grave 39. The green circles indicate the growth which are circular.

Animal activity

Animal activity was primarily absent in the sample (89.7%) (Table 10). There was some insect activity which was identified by the presence of pupa casings from graves 36 and 28 and the tunnel appearance of a tibia from grave 36 (Figs. 29 & 30). The tunnel appearance only affected some aspects of the bone, especially the tibial tuberosity. The tunnels ran throughout the bone and there were many entrance and exit holes and thus was excluded as being acidic soil corrosion.

Table 10: The frequencies of animal activity observed per pig and for each of the skeletal regions

Score	Animal activity	Sample (n=39)	Skeletal regions (n=232)
1	Absent	35 (89.7%)	228 (98.3%)
2	Present	4 (10.3%)	4 (1.7%)



Figure 31: Insect pupae in the skull and mandible.



Figure 32: Tunnelled appearance of a tibia.

Additional observations

Course thick hairs (from the rostral region) were attached to various skeletal elements from five graves (1, 15, 22, 25 and 26) (Fig. 31). The presence of hair is interesting given that these typically decompose along with the other soft tissues.



Figure 33: Example of a hair clump attached to the vertebrae.

4.1.3. Taphonomic correlations

Spearman's rank correlation coefficient analyses were conducted to ascertain whether any correlations between the different taphonomic variables existed. Eight significant correlations could be observed (Table 11). Four significant correlations were observed between the completeness of the remains and depositional staining, adipocere formation, weathering, and plant activity, respectively. Colour had a weak negative correlation with the skeletal completeness ($\rho = -0.151$; $p = 0.021$), which suggests that the more complete the skeleton is the darker the depositional staining. Adipocere had a weak positive correlation with skeletal completeness ($\rho = 0.142$; $p = 0.030$). This would indicate that in cases where more adipocere was present remains were also generally more complete. Both weathering and plant activity had negative correlations with skeletal completeness but the relationship with weathering was stronger ($\rho = -0.364$; $p < 0.001$) than the relationship with plant activity ($\rho = -0.150$; $p = 0.023$). These relationships suggest, as can be expected, that less severe cases of weathering and plant destruction were associated with increased skeletal completeness (Table 11).

Additionally, depositional staining and acidic soil corrosion presented with a weak negative correlation ($\rho = -0.161$; $p = 0.014$), indicating that decreased levels of soil corrosion were typically seen in remains that presented with lighter depositional staining (Table 11). A weak positive correlation was also noted between the presence of adipocere formation and plant activity ($\rho = 0.136$; $p = 0.038$). This correlation suggests that as the adipocere formation of a region increases so does the plant activity (Table 11). Weathering had positive correlations with acidic soil corrosion and plant activity. The relationship between acidic soil corrosion was weaker ($\rho = 0.141$; $p = 0.032$) than that of the plant activity ($\rho = 0.524$; $p < 0.001$). Thus, illustrating that as the weathering score of a region increased (i.e., the weathering became more severe), the acidic soil corrosion and the plant activity scores also increased.

Table 11: Region specific inter-taphonomic correlations (n=232)							
		Completeness	Depositional staining	Adipocere	Weathering	Acidic	Plant
Completeness	Rho						

	Sig.						
Depositional staining	Rh _o	-0.151					
	Sig.	0.021					
Adipocere	Rh _o	0.142	0.050				
	Sig.	0.030	0.445				
Weathering	Rh _o	-0.364	-0.043	0.033			
	Sig.	<0.001	0.518	0.615			
Acidic	Rh _o	0.074	-0.161	-0.024	0.141		
	Sig.	0.260	0.014	0.716	0.032		
Plant	Rh _o	-0.150	0.017	0.136	0.524	0.017	
	Sig.	0.023	0.799	0.038	<0.001	0.795	
Animal	Rh _o	0.040	0.000	0.060	0.106	0.018	-0.011
	Sig.	0.541	0.994	0.360	0.106	0.779	0.864

4.2. Soil composition

The analysis of the soil composition of each grave included the determination of both the pH and the percentage organic content. Soil samples were taken from three levels for control and grave samples: 10 cm from the surface, at the level of the carcass, and 10 cm below the carcass. The raw values for the pH and organic content can be found in the appendix, Table 3.

The pH of the control soils was mildly acidic, especially the surface layer, while the grave soil averages were more acidic than the controls (Table 12).

Table 12: The pH levels as measured for the control and grave soils at each level (n=39)		
	Control	Average pH of graves
10 cm from the surface	4.62	4.57
At the level of the carcass	5.66	4.30
10 cm below the carcass	5.77	4.44

Generally, low percentages of organic content were observed in both the control and the grave samples. A comparison of the percentage organic content per layer between the control and grave samples indicated that for all levels, the percentage organic content measured in the graves was higher than that of the control, especially the samples at the level of the carcass and 10 cm below the carcass (Table 13).

Table 13: The percentage organic content at each level as measured for the control and grave samples (n = 39)		
	Control	Average % organic content
10 cm from the surface	3.6952	5.6010
At the level of the carcass	3.4989	6.9923
10 cm below the carcass	3.6215	8.0282

To determine whether the presence of a cadaver affected the pH, a one-sample t-test was used to compare the means at each level to the control (Table 14). There was no significant difference between the means of the control and the soil 10 cm from the surface ($p = 0.380$). However, there was a significant difference between the controls and the grave soil at the level of the carcass and below ($p < 0.001$).

Table 14: One-sample t-tests of the pH at the various levels compared to the controls	
	Test Value = 4.62
	Significance

	One-Sided p	Two-Sided p
Surface	0.190	0.380
Carcass	<0.001	<0.001
Below	<0.001	<0.001
Bold indicates significant results.		

To assess the influence the carcass may have had on the percentage organic content, a one-sample Wilcoxon sign rank test was used to compare the grave soils to the controls at each level due to the nonparametric nature of the organic content. All p-values were less than 0.05 indicating that all the grave soil levels were significantly different from the controls. This suggests that the presence of the cadaver altered the pH of the soil and increased the organic content.

4.2.1. Comparison between different layers of soil

The pH of the different soil layers was compared using an ANOVA as the pH was normally distributed (Table 15). Overall, there was a significant difference between the three soil layers ($p = 0.042$). To further analyse which layers had significant differences a Tukey *post hoc* test was conducted which established that there was a significant difference between the surface and carcass level ($p = 0.032$).

Table 15: ANOVA and Tukey <i>post hoc</i> results of the differences between pH values as measured at the three levels		
		Sig.
Between Groups		0.042
(I) Level	(J) Level	
Surface	Carcass	0.032
	Below	0.405
Carcass	Surface	0.032

	Below	0.418
Below	Surface	0.405
	Carcass	0.418
Bold indicates significant results.		

The comparison between the three different levels' percentage organic content was analysed using a Kruskal Wallis test as the percentage organic content was nonparametric (Table 16). Similarly to pH, there was an overall significant difference between the three layers ($p < 0.001$). When looking at the individual levels the significant differences lay between the surface and below-carcass level layers ($p < 0.001$), and the surface and carcass layers ($p < 0.001$). There was no significant difference between the level of the carcass and the level below the carcass.

Table 16: Kruskal Wallis results of the difference between the percentage organic content as measured at the three levels	
	Sig.
Asymptotic Sig. (2-sided test)	<0.001
Sample 1-Sample 2	
Surface-Below	<0.001
Surface-Carcass	<0.001
Below-Carcass	0.647
Bold indicates significant results.	

4.2.3. Soil composition correlations with taphonomy

Correlations between pH and organic content as well as between soil composition (pH and organic content) and the taphonomic alterations were observed for both the pigs and the skeletal regions (Tables 17&18).

pH presented with a moderately negative correlation ($\rho = -0.399$; $p = 0.012$) when correlated to organic content and skeletal completeness ($\rho = -0.417$; $p = 0.008$). These relationships

indicate that as the pH of the soil increases (becomes more basic) the percentage of organic content and skeletal completeness decreases.

Five correlations were observed between the taphonomic alterations per skeletal region and pH and the organic content. pH had a weak positive correlation with both depositional staining ($\rho = 0.219$; $p = 0.001$) and weathering ($\rho = 0.177$; $p = 0.007$). This relationship would suggest that as pH increases (becomes more basic) the depositional staining gets lighter, and the weathering becomes more severe. In contrast, pH had a weak negative relationship with adipocere formation ($\rho = -0.173$; $p = 0.008$) which suggests that as pH becomes more basic, the formation of adipocere decreases (Table 18).

The percentage organic content had two significant correlations with the skeletal region taphonomic alterations. Adipocere had a weak positive correlation ($\rho = 0.138$; $p = 0.035$) with the percentage organic content. This would suggest that as the organic content within the soil increased so did the adipocere formation observed on the bone. Conversely, weathering had a weak negative correlation ($\rho = -0.172$; $p = 0.009$) with percentage organic content. This relationship indicates that as the percentage of organic content in the soil increases the skeletal regions would be less weathered.

Table 17: Sample correlations with soil composition (n=39)										
		pH	% Organic content	Completeness	Depositional staining	Adipocere	Weathering	Acidic	Plant	Animal
pH		1.000	-0.399	-0.41	0.222	-0.130	0.160	-0.269	0.101	0.165
	Sig.		0.012	0.008	0.175	0.432	0.330	0.098	0.542	0.315
% Organic content		-.399*	1.000	0.113	0.182	0.174	-0.259	0.162	-0.133	-0.045
	Sig.	0.012		0.495	0.267	0.290	0.112	0.325	0.419	0.785
Bold indicates significant results.										

Table 18: Region-specific taphonomic correlations with soil composition (n=232)									
	pH	% Organic content	Completeness	Depositional staining	Adipocere	Weathering	Acidic	Plant	Animal
pH	1.000	-0.399	-0.417	0.219	-0.173	0.177	-0.093	0.085	0.082
		0.000	0.000	0.001	0.008	0.007	0.160	0.195	0.213
% Organic content	-0.399**	1.000	0.113	-0.004	0.138	-0.172	-0.029	-0.070	0.027
	0.000		0.086	0.947	0.035	0.009	0.655	0.291	0.687
Bold indicates significant results.									

4.3. Burial environment

4.3.1. Excavations and general observations

The grave surfaces were covered by vegetation such as natural grasses and pioneer plants including *Tagetes minuta* (Khakibos or African Marigold), *Bidens pilosa* (Blackjack) and *Themeda triandra* (red grass and red oat grass) (Fig. 33).

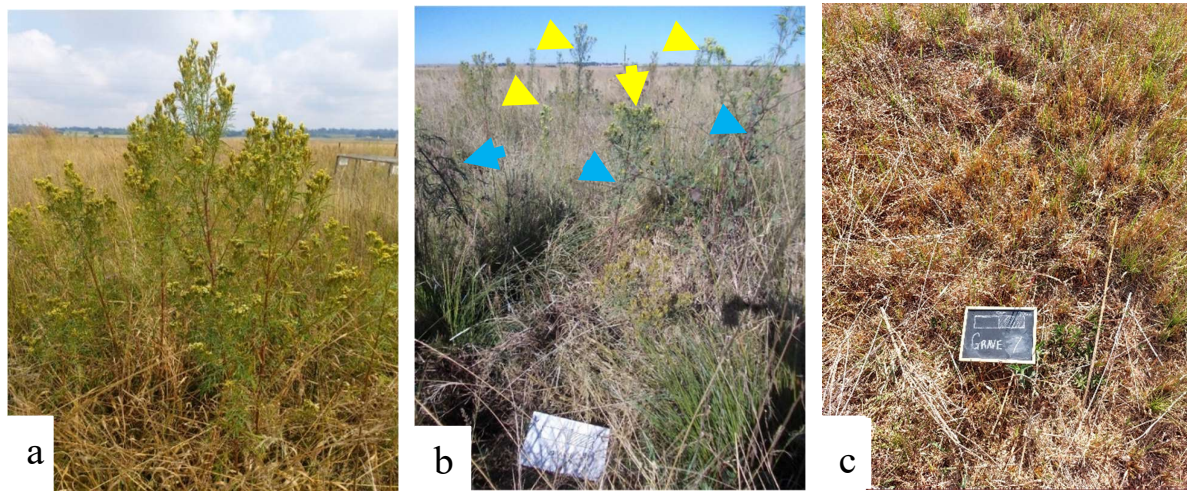


Figure 34: Examples of the vegetation. A) *Tagetes minuta* growing above a grave. B) The yellow arrow indicates *Tagetes minuta* while the blue arrows indicate *Bidens pilosa*. C) *Themeda triandra*.

The area where the remains were buried was at a slight slope with the top of the slope being closer to the entrance gate. This slope is a naturally occurring feature of the landscape.

After the vegetation was removed, the surface layer of soil could be observed (Fig. 34). The soil colour and texture were brownish red, and the soil was dry and rocky. Each grave was then systematically excavated in 10 cm layers to enable observation of grave features, including soil colour and texture changes. In general, it was noted that the soils above and around the carcass were brown to dark brown in colour, whereas soil directly beneath the carcass was red to orange and was often very hard and rocky, consistent with sterile soil (Fig. 35). Similarly, the control soils were brownish-red or orange in colour. An interesting observation was the presence of black-coloured soil around the head, neck, and trunk regions of the skeletons (Fig. 36).



Figure 35: A cleaned grave surface.

Another interesting observation was air pockets around the limbs and trunk, and at different depths in the soil layers covering the carcass (Fig. 37). These air pockets were observed in 13 of the 39 graves (33.3%). No obvious pattern in the positioning of these 13 graves could be observed, which suggests that the presence of air pockets is not associated with grave placement. Potential scavenger holes were observed on the surfaces of graves 9, 14 and 16 (Fig. 38). These were characterised by shallow holes or indentations on the grave surface. For the most part, holes extended to approximately 10 cm from the surface with only one carcass seemingly having been affected by scavenger tunnelling. For a breakdown of the individual features observed for each grave refer to the appendix, Table 3.



Figure 36: Sterile soil indicated by the green box. The surrounding brown soil is the grave soil.



Figure 37: Examples of black soil around the trunk and skull as indicated by the red boxes.



Figure 38: Holes and air pockets found within the graves. a) Air pockets between the ribs (grave 25) b) An air pocket approximately 20cm from the surface of grave 12.



Figure 39: An example of a scavenger hole on the surface of grave 16.

Grave 33 was not backfilled correctly and 42 cm of soil was missing. Blackjacks covered the entire grave. The skull as well as most of the left side of the body were missing which could be an indication of scavenger activity.

The cemetery is situated in a north-westerly direction. Most of the pig carcasses ($n = 38$; 97.4%) were placed on their sides. Of these 38, 27 (69.2%) were on their right sides and 11 (28.2%) were on their left. The carcass in grave 3 was positioned on its back (Fig. 39).



Figure 40: Carcass of grave 3 positioned on its back.

The raw values of the depths of the graves can be seen in the appendix, Table 4. The average depth of the graves was 71.83 cm. Grave position refers to the division of the “cemetery” into four quadrants depending on their location from the entrance gate. The “cemetery” where the pigs were buried was on a slight slope which caused the accumulation of water in the back corner in a north-westerly direction (designated as quadrant 2).

4.3.2. Correlations of burial environment with taphonomic alterations

There were three significant relationships between the taphonomic alterations per pig and the burial environment factors: depth and grave position. Adipocere had a fairly positive relationship ($\rho = 0.349$; $p = 0.029$) with depth suggesting that as the depth of the grave increased so did the adipocere formation on the skeleton. In contrast, the weathering of the individuals had a moderate negative correlation with depth ($\rho = -0.338$; $p = 0.035$). This relationship indicates that as the depth of the grave increased the weathering of the skeleton

was less severe. Additionally, plant activity had a significant negative relationship with the position of the grave ($\rho = -0.344$; $p = 0.032$) suggesting that the closer the graves were to the entrance gate and at the top of the slight slope, the less plant activity was observed on the skeleton (Table 19).

When comparing the relationships between depth, grave position, and the skeletal regions' taphonomic alterations, there were four significant results. Similarly to the individual results, adipocere and depth of the grave had a weak positive relationship ($\rho = 0.175$; $p = 0.008$) indicating that as the depth of the grave increased, adipocere formation increased. Grave positioning had moderate negative correlations with depositional staining ($\rho = -0.217$; $p = 0.001$), adipocere formation ($\rho = -0.286$; $p < 0.001$) and plant activity ($\rho = -0.333$; $p < 0.001$). These relationships suggest that the graves closer to the entrance gate and at the top of the slope were stained darker, had less adipocere on the skeletal regions, and had less plant activity (Table 20).

Table 19: Sample taphonomic correlations with the burial environment (n=39)										
		Depth	Grave position	Completeness	Depositional staining	Adipocere	Weathering	Acidic	Plant	Animal
Depth		1.000	-0.229	0.015	0.174	0.349	-0.338	0.136	-0.059	-0.087
	Sig.		0.161	0.930	0.288	0.029	0.035	0.409	0.723	0.601
Grave position		-0.229	1.000	-0.045	-0.122	-0.261	-0.013	-0.180	-0.344	0.237
	Sig.	0.161		0.785	0.461	0.109	0.937	0.272	0.032	0.147
Bold indicates significant results.										

Table 20: Region-specific taphonomic correlations with the burial environment (n=232)										
		Depth	Grave position	Completeness	Depositional staining	Adipocere	Weathering	Acidic	Plant	Animal
Depth		1.000	-0.229	0.015	0.063	0.175	-0.124	-0.118	-0.050	-0.016
	Sig.		0.000	0.825	0.343	0.008	0.060	0.072	0.445	0.809
Grave position		-0.229	1.000	-0.045	-0.217	-0.286	-0.078	-0.012	-0.333	0.123
	Sig.	0.000		0.493	0.001	0.000	0.236	0.860	0.000	0.061
Bold indicates significant results										

CHAPTER 5: DISCUSSION

The purpose of this study was to study the interrelated relationships between dry bone taphonomic alteration, soil compositions and the burial environment on a sample of buried skeletonized domestic pigs (*Sus scrofa*) in the South African Highveld. Results from this study identified six different taphonomic alterations namely depositional staining, adipocere formation, weathering, acidic soil corrosion, plant activity, and animal activity. Differences in skeletal completeness were also noted throughout the sample. Slight changes in soil compositions between and within graves (as measured at different levels) could be observed. Findings from this study identified several correlations between skeletal completeness, taphonomic alterations, soil composition and the burial environment.

5.1. Taphonomic alterations

The intra – and interobserver error rates suggest the scoring methods used in this study were generally reliable. Weathering scores had a similar agreement for both the intra- and interobservers as they both had near perfect agreement (Kappa = 0.8). These results may be owed to the extensive research done on weathering stages (Behrensmeyer, 1975, 1978, 1980; Ross and Cunningham, 2011) albeit on surface remains, and the fact that the scorers are already familiar with observing these patterns as the Behrensmeyer's scoring system is typically taught and used in forensic anthropology. The agreement between the interobservers was, however, lower for the depositional staining, acidic soil corrosion and plant activity. The low scores for depositional staining are to be expected as colour perception is subjective (Bloch *et al.*, 2021). In this study the use of the Munsell Soil Colour chart was utilized in an attempt to minimize the subjectivity of scoring colours, however it seems that even with the use of this system the scoring of colour changes remains problematic. The lower scores observed for the acidic soil corrosion and plant activity may be because these alterations have similar destructive characteristics (e.g., etching) and may have been scored as either acidic soil corrosion or plant activity. Root etching is typically caused by weak acids excreted by plant roots (Forbes, 2008). In cases, where remains are already severely weathered it may therefore become increasingly difficult to distinguish between acidic destruction caused by acidic soils versus plant root excretions. There were positive correlations with weathering, acidic soil corrosion ($\rho = 0.141$; $p = 0.032$) and plant activity ($\rho = 0.524$; $p < 0.001$), which may also allude to this phenomenon.

5.1.1. Skeletal completeness

The average skeletal completeness was low (43.02%). This can be attributed to selective preservation as most of the smaller bones that make up the limbs were not recovered. The fibula, which is a very small bone in juvenile pigs, and the bones of the feet such as the carpals, tarsals, metacarpals and metatarsals, and the phalanges were most often absent. These are small and thus could easily be missed during the excavation process (Bello *et al.*, 2006; Djurić *et al.*, 2011; Manifold, 2012). Additionally, small bones are not as well preserved as larger bones since small bones have a larger surface area to volume ratio which increases the rates of reaction between the bone and the surrounding environment. This will cause the bones to degrade faster and thus cannot be recovered (Von Endt & Ortner, 1984; Bello *et al.*, 2006; Djurić *et al.*, 2011; Manifold, 2012)

It was, however, noticed that skeletal completeness was increased by the presence of adipocere ($\rho = 0.142$; $p = 0.030$). Adipocere is a modification of the decomposition process and often occurs in burial environments (Fiedler & Graw, 2003). It is formed from the body's fats which through the process of hydrolysis and hydrogenation are broken down into fatty acids. These fatty acids then solidify and create a soft wax or crust-like substance that covers soft tissues and skeletal elements (Fiedler & Graw, 2003; Forbes *et al.*, 2005). The presence of adipocere has been found to decrease the rate of decomposition leading to the preservation of remains as adipocere is extremely resistant to degradation (Fiedler & Graw, 2003; Forbes *et al.*, 2005; Fiedler *et al.*, 2009). This would, therefore, account for the increased skeletal completeness in cases where adipocere was observed.

5.1.2. Soil staining

The stains that were observed in this sample were black, dark brown, brown, yellowish-brown, yellowish-grey and greyish brown. The most prominent colours observed on the pig skeletons were dark brown (41.0%) and brown (46.2%). Documentation of soil colours during the excavation of the graves indicated that soils at the level of the carcass mainly presented as brown or dark brown (7.5YR, 10YR, 2.5YR and 5Y). This finding corresponds with what we know about the leaching of soil tannins into porous bone. Bone interacts with soil through the soil solution which contains soil tannins, minerals, and micro-organisms. The water will flow through the bone's pores where various chemical reactions will take place including the adsorption of the soil tannins onto the bone surface (Hedges & Millard, 1995). The colour of soil, and thus the subsequent bone colour, is indicative of the soil's composition with regards to the minerals present and the percentage of the organic content

(Conklin Jr, 2014). Dark soils are higher in organic content which decomposes to form a black product known as humus. Dark soils are often also anoxic and have reducing conditions. This contrasts with bright red or brown soils that contain iron and are well-drained and oxidizing (Conklin Jr, 2014; Dupras & Schultz, 2014).

Differences in the distribution of soil staining could be observed throughout the skeleton. Only the trunk region presented with black (n=3) as well as dark brown (n= 21) staining. During soft-tissue decomposition, the trunk region would produce more decomposition fluids compared to the neck/head and limb regions (Galloway *et al.*, 1989; Megyesi *et al.*, 2005). The reason for this is that the trunk contains most of the internal organs that will liquefy during the process of putrefaction and decay. Subsequently, the decomposition of soft tissues leads to an influx of carbon and nutrients into the surrounding soils creating a cadaver decomposition island (CDI) that is high in organic content and can stain the soil a darker colour (Gill-King, 1997; Carter *et al.*, 2007; Forbes, 2008). The CDI can be confirmed by the presence of black soil observed at the level of the carcass from 20 of the graves. The black soil was present after six years of interment which could suggest that these soils do not drain well, and that groundwater movement was limited. Marais-Werner's (2016) study suggested that the soil in this area may be clay. Clay soils are made up of small particles which decreases water and gas movement (Owens & Rutledge, 2005).

The black soil surrounding the skull and abdomen could be a by-product of organic content decomposition which forms a black substance known as humus assisted by micro-organisms (Conklin Jr, 2014). The abdomen contains the digestive tract and thus this area, along with any undigested food, would have more organic matter than the rest of the body creating substantial amounts of humus. The pig sample all died of natural causes which is often from an *E.coli* or salmonella infection (Marais-Werner, 2016). Both these infections affect the small intestine and cause excessive diarrhoea and dehydration which may also cause internal haemorrhaging and thus haemolysis staining (Symes *et al.*, 2014; Novotny *et al.*, 2021). Another possible cause for the black soil could be the diet of the pigs. The diet dictates the types of microbes that are present in the gut and thus these microbes may have had an influence during the decomposition process and caused this black staining. However, there is currently no research that suggests that specific microbes influence post-decomposition staining.

Overall, the right side of the skeleton more often presented with dark brown and brown soil staining, compared to the left side. Twenty-six of the right limbs (including both the fore- and hindlimbs) were scored as dark brown. Eighteen trunks also presented with darker staining on the right side. This feature may be associated with the burial position. Twenty-seven of the carcasses/pigs were buried on their right side. As the majority of the right-sided trunks and limbs would have been in direct contact with the CDI it can be assumed that these regions would therefore also be stained a darker soil colour. The left side of the body had a higher count for the lighter colours such as brown ($n = 41$), yellowish-brown ($n = 5$) and greyish brown ($n = 1$). The left sides of the body were not in contact with the bottom of the grave where the CDI had been created and thus was not in contact with the darker soil.

Depositional soil staining had a negative relationship with the percent completeness ($\rho = -0.151$; $p = 0.021$) as well as acidic soil corrosion ($\rho = -0.161$; $p = 0.014$). These relationships suggest that the darker the skeleton the more complete it is, but the more acidic soil corrosion would be present. The darker staining is due to the increased organic content of the CDI which is also an environment that is conducive to adipocere formation that will protect the skeleton and increase the percent completeness. A CDI contains the bacteria and moisture necessary for adipocere formation (Fiedler and Graw, 2003; Forbes *et al.*, 2005; Carter *et al.*, 2007). These results contrast one another as acidic soil corrosion usually decreases the skeletal representation (Gordan & Buikstra, 1981). This could be because of the low agreement of scoring between the inter and intra-observer errors for acidic soil corrosion. The score for acidic soil corrosion may not be accurate or repeatable as it has similar characteristics to other taphonomic alterations such as weathering or plant activity, as previously mentioned.

Three of the skeletons also presented with pink-red stains that were on the crania, a rib and the shaft of a femur. These stains could have been caused by haemolysis which is when the red blood cells rupture and stain the bone (Huculak & Rogers, 2009; Dupras & Schultz, 2014). An alternative explanation for these stains could be that the soil may have also contained manganese which is a common metal in soils (Dupras & Schultz, 2014). Manganese is highly reactive and can form compounds that have distinct colours such as manganese carbonate or permanganate in the soil which are both purple or pink (Dupras & Schultz, 2014). However, due to the location of the staining which was on both the external and internal surface of the cranium, one of the superior ribs as well as the shaft of the femur would suggest that the stains were more probably caused by haemolysis. The rupturing of

the blood vessels may have occurred postmortem when the carcasses were transported and deposited into the graves.

5.1.3. Adipocere formation

In this sample, adipocere was present in 92.3% of the graves. It was most prevalent in the regions of the head and neck as well as the trunk. Adipocere was also observed more on the hindlimbs (n = 22) and the right sides of the body (n = 43).

Adipocere formation requires bacteria for hydrolysis of the neutral fats and thus will form in any environment that promotes bacterial survival (Forbes *et al.*, 2005). The most conducive environment is one that is warm, anaerobic with moisture. Aquatic and waterlogged environments provide an ideal environment for adipocere formation (Ubelaker, 1997b; Fiedler & Graw, 2003; Higgs & Pokines, 2014). Five of the graves during the previous study (Marais-Werner, 2016) were waterlogged. During the excavation phase of the current study, there were no waterlogged graves, however, excavations were undertaken during the dry winter months and would, therefore, not necessarily have been waterlogged at the time of excavation. The presence of the decomposing body may have provided sufficient moisture for the formation of adipocere (Forbes, 2008; Moses, 2012). The head and neck had increased adipocere formation to the extent where adipocere had formed on the internal surface of some of the crania. This could be due to the high-fat content of the brain due to the myelin sheaths that surround the neurons (Gill-King, 1997). The trunk region has an especially high-fat content that increases the likelihood that adipocere would form in this region. The hindlimbs are in closer contact with the abdomen. The gut also contains the bacteria that initiate decomposition and thus would be present for adipocere formation (Gill-King, 1997; Forbes, 2008; Hamilton & Green, 2017). However, the grave environment itself may also have contributed to the formation of adipocere. As mentioned previously, the grave soils may not have been well-draining as was indicated by the lack of dark soil stains beyond the level of the carcass. Thus, the remains would have been surrounded by the decomposition fluids for an extended period leading to the formation of adipocere as there would be sufficient moisture and bacteria from the body. An alternative explanation relates to graves acting as water catchment areas as the aerated grave soil (due to backfilling) allows for water movement whereas the sterile soil (representing a clay layer) would prevent the water from seeping down (Steyn *et al.*, 2000; Dupras *et al.*, 2006). The water retention of the graves can promote pioneer vegetation species for example in this

study *Themeda triandra* grew only above waterlogged graves and thus can be used for identifying a grave (Dupras *et al.*, 2006; Cheetham & Hanson, 2016).

There was also a positive relationship with adipocere formation and increased plant activity ($\rho = 0.136$; $p = 0.014$). This suggests that graves that presented with adipocere also showed increased plant activity. This would further serve as evidence for the increased moisture retention in these graves, as well as the increased presence of organic matter (brought on by decreased degradation as a result the adipocere), which will stimulate plant growth (Steyn *et al.*, 2000).

5.1.4. Weathering and plant activity

Adipocere formation was positively correlated with an increase in the skeletal completeness whereas weathering ($\rho = -0.364$; $p < 0.001$) and plant activity ($\rho = -0.150$; $p = 0.023$) were correlated with a decrease in skeletal preservation. These two taphonomic alterations commonly form part of the diagenesis process as they are typically associated with the destruction of bone (Behrensmeyer, 1978; Pokines & Baker, 2014; Buekenhout *et al.*, 2018).

Most of the pigs ($n = 21$; 53.8%) had moderate weathering of the skeletal regions, however, the head and neck, and the trunk were especially fragile and fragmented. The trunk region was the only region that presented with complete disintegration. This was especially true for the skeletons that did not present with adipocere in this region, that would otherwise have slowed down the degradation process. The bones of the trunk are inherently more fragile compared to the rest of the body. Flat bones, such as the ribs, and irregular bones, such as the vertebrae and carpals and tarsals, have thin layers of cortical bone relative to the trabecular bone, which make it more prone to weathering. These bones also typically have an increased surface area to volume ratio with which the soil solution can react (Hedges & Millard, 1995). This will increase the rate of diagenesis and breakdown of the organic and mineral content of the bone which explains why decreased skeletal preservation was observed in these skeletal elements. The bones of the trunk are also exposed more to the organic acids that are produced during decomposition when compared to the other regions (Gill-King, 1997; Carter & Tibbett, 2008). In addition, the left side of the body was more weathered than the right. Again, the right side had more adipocere which would protect it from the weathering process.

Plant activity, such as root infiltration with bone damage, was mostly seen in the same regions as weathering, namely the head, neck and the trunk. The roots of plants grow into

and around the bone as bone is a good source of nutrients and water (Pokines & Baker, 2014). Roots infiltration can be very destructive and cause extensive fracturing of bones (Pokines & Baker, 2014). Additionally, plant roots excrete organic acids to enable the uptake of minerals from the bone. In the advanced stages, organic acid secretion can cause damage to the bone that resembles features associated with acidic soil corrosion, such as a roughened cortical surface (Pokines & Baker, 2014). In this study, 29 pigs presented with advanced stages of organic secretion which was especially prevalent on the head/neck and hindlimbs.

There was some fungi growth on the scapula from grave 39 which could indicate that this grave may have had some oxygen as most fungi are aerobic (Dent *et al.*, 2004). This is further supported by the lack of adipocere in this grave which requires an anaerobic environment.

5.1.5. Acidic soil corrosion

Acidic soil corrosion was common on the head and neck ($n = 20$) as well as on the hindlimbs ($n = 14$). The lacrimal bones of the skull were more often affected which could be due to their thin structure making them more susceptible to damage ($n = 17$). The hindlimbs presented with a scooped appearance on the cortical surface, especially on the bones of the femur and astragalus. The scooped appearance exposed the trabecular bone, especially on the astragali. The proximal femur may have been more commonly damaged as it is the most proximal point of the hindlimb and is close to the abdominal region. The abdomen produces substantial organic acids during soft tissue decomposition which may have contributed to the acidic erosion (Gill-King, 1997; Hamilton & Green, 2017). The hindlimbs are close to the abdomen and thus the large volume decomposition fluids and organic acids would have also influenced the bones of the hindlimbs.

Acidic soil corrosion had a positive relationship with weathering ($\rho = 0.141$, $p = 0.032$) which indicates that as the acidic soil corrosion increased so did weathering. Weathering and acidic soil corrosion have similar destructive characteristics and, therefore, this correlation would make sense.

5.1.6. Animal activity

There was very minimal animal activity observed in the sample ($n = 4$), and, therefore, also no correlations with the other taphonomic alterations. As the remains were buried, the access of insects such as flies and scavengers was reduced significantly (Pokines & Baker, 2014). There was evidence of insect exoskeletons in the grave 36. It was noted during the

excavation of grave 36, in the previous study, that there was mass colonization of the carcass during the *in situ* observations (Marias-Werner, 2016). Grave 28 had evidence of termite tunnels although no termites were observed, and none of the bones presented with termite damage. Termites are often drawn to buried remains as the bones provide them with essentials such as nitrogen and phosphorus (Pokines, 2014). Additionally, the cranium as well as most of the left side of the body from grave 33 were missing. This grave was especially shallow which may have attracted scavengers.

5.2. Soil composition

The control soils collected suggest that the pH of the soil at the Miertjie le Roux Agricultural experimental farm is mildly acidic. In addition, there is very little organic content which was supported by the red copper colour of the soil (Conklin Jr, 2014; Dupras & Schultz, 2014).

In this study, the pH of the control soils at the level of the grave was 5.66 whereas the average pH of the grave soil at the level of the carcass was 4.30. This indicates that the presence of the carcass decreased the pH of the soil to become more acidic. Additionally, the carcass increased the percent organic content as the controls had a low percentage of 3.4989, whereas the grave soil average increased to 6.9923. Both the changes in pH and the percent organic content compared to the control were significant. This is to be expected as a carcass is a substantial source of carbon and nutrients that will infiltrate the soil during soft tissue decomposition creating a CDI (Carter *et al.*, 2007; Aitkenhead-Peterson *et al.*, 2012). Most studies have found an increase in the soil pH due to the release of base cations (positively charged ions) and ammonium during the early phases of decomposition, followed by a subsequent decrease during the later stages as these base cations get oxidized to produce anions (negatively charged ions) (Vass *et al.*, 1992; Carter *et al.*, 2007; Aitkenhead-Peterson *et al.*, 2012; Anderson *et al.*, 2013).

It was also observed during this study that there was a difference in the pH and the organic content between the three levels. The pH was significantly different between the surface and carcass levels. Similarly, the percentage organic content was significantly different between the surface level and both the level of the carcass and 10 cm below the carcass. These differences are due to the presence of the decomposing carcass (Vass *et al.*, 1992; Aitkenhead-Peterson *et al.*, 2012; Anderson *et al.*, 2013).

There was a negative relationship between the pH and the organic content ($\rho = -0.399$; $p = 0.012$) and the depositional staining ($\rho = 0.219$; $p = 0.001$). This would mean that lower

percentages of organic content in the soil are associated with a higher or basic pH and a general lighter soil staining on the bones. The organic content in these graves could primarily be derived from the decomposing carcass as the control has a lower percent of organic content. The pH of the organic content from carcass is acidic as the graves are in the extended postmortem interval, as mentioned previously (Carter *et al.*, 2007; Vass, 2011; Aitkenhead-Peterson *et al.*, 2012; Anderson *et al.*, 2013), and thus less organic content would make the graves more basic. As mentioned previously, organic matter decomposes to form a black product (humus) that darkens the soil. Therefore, the lighter the depositional staining can be assumed in the presence of a lower organic content (Conklin Jr, 2014; Dupras & Schultz, 2014).

The soil pH determines the rate of diagenesis as it affects the solubility of the mineral hydroxyapatite and the protein collagen (Nielsen-Marsh *et al.*, 2000; Hedges, 2002; Trueman & Martill, 2002; Kendall *et al.*, 2018). The mineral component of bone is soluble at a low pH (acidic) whereas collagen is more susceptible to breakdown at the extremes of pH which include both acidic and alkali environments (Collins *et al.*, 2002; Hedges, 2002; Kendall *et al.*, 2018). Collagen is also broken down enzymatically by collagenases that are produced by bacteria. These collagenases are too large to enter the bones' compact structure thus the hydroxyapatite is first removed (Collins *et al.*, 2002). Thus, in an acidic pH environment bone will degrade quicker as the hydroxyapatite is dissolved (Hedges & Millard, 1995; Nielsen-Marsh *et al.*, 2000; Collins *et al.*, 2002; Hedges, 2002; Kendall *et al.*, 2018). The results of this study contrast to those in the literature as it was found that an increased pH (alkaline environment) was more damaging to the skeleton as there was a relationship with increased weathering ($\rho = 0.177$; $p = 0.007$) and a decrease in skeletal completeness ($\rho = -0.417$; $p = 0.008$). This could be caused by the very small pH ranges in this study as they were all acidic. This could skew the results as there is no real comparison to a basic pH. Additionally, these remains were those of juveniles which have a higher percentage of collagen to hydroxyapatite (Bello *et al.*, 2006; Manifold, 2012). Bello *et al.* (2006) suggested that adult skeletons were more susceptible to damage in acid environments as they had increased mineralization.

Additionally, at a basic pH the formation of adipocere was reduced ($\rho = -0.417$; $p = 0.008$). The environment in this study was acidic but adipocere was able to form. In the study, Forbes *et al.* (2005) conducted adipocere formation was enhanced by moist, warm, anaerobic, and mildly alkaline conditions, however; adipocere was also present in the acidic environment

with a pH of 2.4. Extreme alkaline environments inhibited adipocere formation (Forbes *et al.*, 2005). This may also serve as an explanation as to why increased soil pH showed a correlation with decreased skeletal preservation, as the formation of adipocere would have protected the degradation of skeletal elements to some extent.

Adipocere had a positive correlation with the organic content ($\rho = 0.138$; $p = 0.035$) within the soil. This may be explained by adipocere resulting in a decreased breakdown of organic material, therefore resulting in an increase in the percentage organic content (Gill-King, 1997; Fiedler & Graw, 2003; Fiedler *et al.*, 2004; Forbes, Dent & Stuart, 2005) in soil samples from graves that also presented with adipocere formation on the skeletal remains.

The percentage of organic content had a negative relationship with weathering ($\rho = -0.172$; $p = 0.009$) suggesting that graves with a lower organic content presented with skeletons with increased weathering. The presence of adipocere would have increased the organic preservation and delayed weathering of the bone. Alternatively, the conditions in the grave that enabled adipocere formation, also resulted in the retention of more organic matter.

5.3. Burial environment

Tagetes minuta, *Bidens pilosa* and *Themeda triandra* are pioneer plants that grow in disturbed areas (Cornelius & Wycliffe, 2016). These were only found to grow on top of the graves. This would make sense as the digging of a grave would result in the disturbance of the natural soil layers, creating an environment ideal for pioneer plant growth. Both *Tagetes minuta* and *Bidens pilosa* have been indicated as common surface indicators indicating the presence of a grave (Dupras *et al.*, 2006; Iscan & Steyn, 2013; Holland & Connell, 2016). *Themeda triandra* is a grass that grows in most soils, but it prefers clay soils with high organic content. It only grew above four graves (3, 4, 7, 8) and between graves 9 and 10. Graves 8, 9 and 10 were indicated to have been waterlogged in the study done by Marais-Werner (2016) thus explaining the presence of *Themeda triandra* as there was substantial water for plant growth. The graves retained significant moisture. In addition, skeletons from graves 3, 7, 8, and 9 had adipocere formation which could be due to the waterlogged environment.

There were air pockets found within the grave as well as around the skeletal elements, especially surrounding the ribs and some of the limbs. These air pockets within the graves are probably due to the decomposition of plant material that was mixed within the soil during the backfilling from the original study (A Meyer 2021, per communication). Whereas the

air pockets around the limbs and ribs could be from the decomposition of soft tissues and these spaces were not filled in by loose soil from around the body as the soil may become compacted during the soft tissue decomposition and once the soft tissues decompose the compact soil would not be able to fill the spaces.

As the depth of the graves increased so too did the adipocere formation ($\rho = 0.349$; $p = 0.029$) but the weathering of the remains decreased with increasing depth ($\rho = -0.338$; $p = 0.035$). The deeper a grave is the more likely it will become anaerobic and with sufficient commensal bacteria and moisture from the body, adipocere is more likely to form (Forbes *et al.*, 2005). Adipocere would then protect the remains from weathering (Fiedler *et al.*, 2004). The deeper the grave the cooler the temperature and thus there would be minimal temperature fluctuations resulting in less breakdown of bone collagen and the subsequent weathering of bone (Von Endt & Ortner, (1984; Carter & Tibbett, 2008a).

The landscape of the cemetery was on a slight slope. Quadrant 2 was positioned at the bottom of the slope where water has been seen to accumulate during and after extensive rainstorms (A. Meyer 2021, per communication). Quadrant 4 was at the top of the slope and represented the quadrant closest to the entrance gate. The remains closer to the entrance gate and at the top of the slope were stained darker ($\rho = -0.217$; $p = 0.001$), had less adipocere ($\rho = -0.286$; $p = 0.000$) and less plant activity ($\rho = -0.333$; $p = 0.000$). This is contrast to the results as consistently adipocere seemed to have some association with darker staining. These results could be explained by the use of the quadrant system, which included graves that presented with adipocere formation in quadrant 4, therefore skewing these soil staining results, as five graves in this quadrant had darker soil stains. However, considering that adipocere formation had no significant correlation with soil staining this discrepancy could be due to the inherent colour of adipocere which is an off white/grey and thus would have been scored as grey during data collection.

Due to the natural slope, water would flow down the slope and accumulate in quadrants 1 and 2 and, thus, adipocere in these quadrants was more common due to the excess in moisture (Fiedler *et al.*, 2004). Water is needed for plant survival and as there was less water in quadrant 4, there would be less plant activity compared to areas with more moisture. *Themeda triandra*, a species known for preferring moist clay soils was found in quadrant 2, where most of the water accumulated.

5.4. Limitations and future recommendations

There are several limitations to this study that need to be considered. These include limitations from the sample model, as pigs were utilized as analogues for human decomposition studies. Due to ethical considerations in South Africa, all decomposition studies need to be conducted on proxies such as pigs, however, it is not an ideal situation as it has been found that pigs decompose differently from humans (Matuszewski *et al.*, 2019). Additionally, the sample was comprised of juvenile pigs. This means that all the epiphyses of the long bones had not yet fused and it is known that juvenile remains decompose faster than adult remains (Bello *et al.*, 2006; Djurić *et al.*, 2011; Manifold, 2012).

There were limitations to the graves and the location as they were disturbed and exposed to the surface environment during the previous study which allowed insects and oxygen back into the grave. The backfilling was conducted twice thus further mixing up the soils which would not often occur in forensic cases. This “cemetery” has been used in multiple surface decomposition studies which have been found to significantly alter the soil chemistry (Carter and Tibbett, 2008b). Carter and Tibbett (2008) found that cadavers buried in pre-existing grave soil had an increased rate of decomposition, as well as the pH, had lowered.

Additionally, due to time constraints, only one repeat was done for the soil pH and the organic content testing. Additionally, the soil texture and moisture were not included in this study. A future study could possibly re-analyses the soil samples as they are still available.

There are no established scoring methods for adipocere formation, acidic soil corrosion, plant activity and animal activity. These scores may not be reliable especially acidic soil corrosion which had a low intra- and interobserver error.

Future studies should include the development of refined scoring criteria especially for the appearance of acidic soil corrosion on bone. Additional studies should be done with larger sample sizes thus increasing the reliability of the results which will also help to establish the scoring criteria. A longitudinal study with different scoring phases at different time intervals that could then be used to estimate the postmortem interval of skeletonised remains. Future studies should also include testing differences between different environments, specifically as related to soil texture (clay vs sandy) and the macroscopic appearance on bone.

CHAPTER 6: CONCLUSION

This study aimed to establish the relationships between the taphonomic alterations, soil composition and burial environment in 39 pigs buried in the South African Highveld region. Apart from this study, there are no studies within a South African context that have analysed macroscopic dry bone taphonomy in buried skeletons. This study, therefore, provides new information that may assist in better understanding the postmortem alterations occurring in burial environments, specifically in the context of the South African Highveld.

The following are the final conclusions made from this study:

- Six taphonomic alterations were observed on the skeletal remains including depositional soil staining, adipocere formation, bone weathering, acidic soil corrosion, plant activity and animal activity.
- Overall, the skeletal completeness was low (43.02%) due to the absence of many small bones such as the tarsals, carpals, metacarpals, and metatarsals as well as the phalanges and the fibulae. The low skeletal completeness was attributed to selective preservation as the bony elements mostly missing represent the smaller and more fragile skeletal elements that are often lost during excavation and subjected to more degradation. The latter is due these skeletal elements typically presenting with larger surface areas to volume ratios. A positive correlation between skeletal completeness and the presence of adipocere was observed. Adipocere is a decomposition by-product that is extremely resistant to degradation and therefore acted as a buffer between the skeletal remains and the taphonomic agents, decreasing bone breakdown.
- There were six different coloured soil stains observed in this sample with dark brown and brown being the most observed. The soils surrounding most of the skeletal remains were also of a dark brown and brown colour, which would account for the soil staining observed on the pig skeletons. Diagenetic changes would have enabled the leaching of soil tannins and minerals into the porous bone, subsequently staining it the same colour as the soil.
- The trunk was the only skeletal region to present with black staining. These stains were contributed to the decomposition fluids that are produced in the abdominal region during the stages of putrefaction. Black soil was also observed around the skeletons indicating the presence of a CDI. The presence of a clearly visible CDI

six years after burial was an interesting find and likely suggests that the soils were not well-draining.

- The right side of the skeleton had the darkest stains compared to the left. This phenomenon was attributed to most of the carcasses having been positioned on their right sides during the initial burial process. The right sides were therefore in direct contact with the decomposition fluids associated with the CDI, resulting in the darker staining. This is a significant find as it may serve as a method to reconstruct burial positions in remains that have been inadvertently disturbed.
- Depositional staining presented with a negative correlation with the skeletal completeness. This indicated that skeletons stained darker generally had an increase skeletal completeness, suggesting that darker soils indicate environments that are more conducive to the preservation of bone.
- Another interesting finding was that almost all the pig skeletons presented with adipocere (36 out of 39 skeletons). Adipocere was especially prevalent on the right side. The lack of waterlogged graves observed during this study's excavations may suggest that the moisture produced during decomposition created an anaerobic environment conducive to the formation of adipocere. The right sides were affected more by adipocere formation as most of the carcasses (27) were initially buried on their right sides. Skeletal elements that came into direct contact with the bottom of the grave where moisture accumulated would therefore have been more likely to develop adipocere. Adipocere and plant activity had a positive correlation which indicates that adipocere formation in a grave was associated with increased plant activity. This supports the notion that skeletons presenting with adipocere were in graves that retained water. It can also be argued that these graves were better in retaining water during episodes of rain resulting in the development of an anaerobic environment.
- The trunk was the most damaged region in terms of weathering and plant activity especially if adipocere was not present. The bones of the trunk are thin (ribs) and present with increased porosity (vertebrae) and are therefore more susceptible to damage from the organic acids produced by decomposition. Weathering and plant activity showed a positive correlation. Whereas weathering and plant activity had negative correlations with skeletal completeness. It has been shown that plant

activity (root etching and root infiltration) can cause extensive damage, and therefore this relationship would make sense.

- The scoring of acidic soil corrosion was somewhat problematic as indicated by the relatively low intra-and inter-observer repeatability. This may be attributed to the difficulty in distinguishing between acidic corrosion caused by plant root acid secretion versus acidic soil corrosion caused by actual acidic soils. Acidic soil corrosion had a positive relationship with weathering. Even where acidic plant root corrosion was inaccurately scored as acidic soil corrosion, both mechanisms are known to result in taphonomic alterations that are destructive which would account for the positive correlation with skeletal completeness.
- The presence of the cadaver decreased the soil pH making the environment more acidic as well as increasing the percent organic content. There was also a significant difference between the pH and organic content between the surface level and the level of the carcass. These differences were attributed to the presence of the carcass, which is known to alter the chemical structure of the surrounding environment as it progresses through the different stages of decomposition.
- There was a negative relationship between pH and the percent organic content as there was less organic content at a basic/higher pH. This would suggest that soils with a slightly more acidic pH were more conducive to the preservation of organic material. It was also found that an increased pH (alkaline environment) was more damaging to the skeleton as there was a relationship with increased weathering. These findings stand in contrast to some of the existing literature that suggests higher pH values (more alkaline) would promote skeletal preservation. Possible explanations for these contrasting results very small pH ranges (all were acidic) which may have skewed the results as there is no real comparison to a basic pH; or alternatively that the use of a juvenile sample may have influenced the rate of bone breakdown. Juvenile remains are known to a higher percentage of collagen to hydroxyapatite, the latter of which are more typically susceptible to damage in acid environments.
- A basic pH was associated with increased adipocere and thus increased skeletal completeness due to the preservation effect of adipocere. There was a negative correlation between the organic content and weathering suggesting that weathering

increased in graves with low organic content. This is again associated with reduced adipocere formation that would have protected the remains from weathering.

- Two pioneer species of plants grew above each grave: *Tagetes minuta* and *Bidens Pilosa*. A third species *Themeda triandra* grew above the graves at the bottom of the slope that are known to become waterlogged during the rainy seasons. The correlations of the burial environment with the taphonomic alterations included a deeper grave leading to an increase in adipocere formation. This can be attributed to deeper graves more often creating anaerobic environments that is conducive to adipocere formation. A deeper grave was also correlated with less weathering due to the protective effect of the adipocere.

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APPENDIX

Table 1: Taphonomic alterations scoring for each pig								
Staining								
ID	Head and neck	Trunk	Left ribs	Right ribs	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	2	6	6	2	3	6	3	3
2	2	3	6	6		3	6	2
3	3	6	3	3	3	6	3	3
4	3	2	3	2	6	3	6	3
5	3	3	4	4	4	6	6	2
6	3	6	2	6	6	6	3	6
7	4	2	4	1	4	2	3	3
8	2	4	4	4	4	4	3	6
9	3	5	5	5	3	3	6	6
10	3	6	6	6	3	3	6	6
11	3	2	3	2	3	3	3	2
12	2	6	2	6	2	2	2	2
13	2	2	3	2	6	2	3	4
14	2	2	2	2	4	3	2	3
15	2	2	2	2	2	3	3	3
16	2	2	3	2	6	6	3	2
17	2	2	1	2	2	3	2	3
18	2	2	2	3	3	3	3	3
19	3	6	5	3	3	3	2	3
20	2	5	6	6	3	6	5	3
21	3	2	3	2	3	2	3	2
22	2	2	2	2	3	3	2	3
23	3	2	2	1	3	3	3	2
24	2	2	1	2	2	2	3	2
25	3	2	2	2	3	3	2	3
26	2	1	2	3	3	3	2	2
28	2	1	2	2	2	2	6	2
29	3	2	2	2	2	2	2	2
30	2	2	2	2	2	2	2	2
31	2	2	2	2	3	2	3	3
32	2	2	2	1	3	2	3	3
33	3	6	3	3	6	2	3	3
34	3	1	2	2	2	2	2	2
35	3	5	5	6	3	3	3	2
36	3	2	2	2	3	3	6	3
37	3	2	3	3	3	3	3	6
38	3	2	2	3	3	3	3	4
39	3	4	4	3	3	3	3	3

40	3	3	3	3	4	3	3	3
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Adipocere						
ID	Head and neck	Trunk	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	1	2	1	1	1	1
2	2	2		1	2	2
3	3	2	2	2	2	2
4	1	2	2	2	2	2
5	3	2	2	2	2	2
6	2	2	2	2	2	3
7	2	2	1	2	2	2
8	2	3	3	3	2	2
9	2	3	3	2	2	2
10	2	2	1	2	1	2
11	2	2	2	2	2	2
12	2	2	2	2	2	2
13	2	2	2	2	2	2
14	2	2	2	2	2	1
15	2	2	1	2	1	1
16	2	2	2	2	3	2
17	2	2	2	1	2	2
18	2	2	2	2	2	2
19	2	3	2	2	2	2
20	2	2	2	1	3	2
21	2	2	1	2	2	2
22	2	1	1	1	2	2
23	1	2	2	2	1	1
24	1	2	2	1	2	2
25	2	2	1	1	1	1
26	1	2	1	2	1	1
28	2	2	2	2	3	3
29	2	2	1	1	1	1
30	1	2	1	2	2	2
31	1	2	2	1	2	2
32	3	2	3	3	1	2
33		1	1	1	2	1
34	2	1	2	1	2	1
35	2	3	2	2	1	1
36	2	2	1	1	1	1
37	1	1	1	1	1	1
38	2	2	2	1	1	1
39	1	1	1	1	1	1
40	1	1	1	1	1	1

Weathering						
ID	Head and neck	Trunk	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	5	3	3	3	2	3
2	5	5		4	3	4
3	5	4	3	3	4	3
4	5	3	3	2	4	4
5	5	4	3	3	4	3
6	5	3	4	3	4	4
7	3	4	3	3	2	3
8	3	3	2	3	3	3
9	4	3	2	2	2	3
10	4	5	3	4	4	4
11	5	5	3	2	3	3
12	3	3	3	2	4	3
13	5	3	4	4	4	4
14	5	5	3	3	4	4
15	3	4	3	3	4	3
16	4	4	3	3	3	3
17	5	5	3	3	4	4
18	3	4	3	3	4	3
19	5	4	3	3	4	4
20	5	6	3	3	3	4
21	3	3	2	2	3	3
22	5	5	4	3	4	3
23	5	5	3	3	3	2
24	5	5	4	3	3	4
25	4	5	3	3	3	3
26	5	5	3	4	3	3
28	5	3	3	2	3	4
29	3	3	2	2	4	4
30	5	3	2	2	2	2
31	2	4	3	2	3	3
32	5	4	2	2	4	2
33		5	4	4	4	3
34	2	3	2	2	3	2
35	3	4	3	2	3	2
36	5	5	4	5	5	5
37	3	4	3	3	4	3
38	4	3	2	2	4	4
39	4	4	3	2	3	4
40	4	4	3	2	2	4

Acidic soil corrosion						
ID	Head and neck	Trunk	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	1	1	1	1	1	1

2	1	1		3	1	2
3	1	1	1	1	1	1
4	1	1	1	1	1	2
5	1	1	1	1	1	2
6	1	1	1	2	2	1
7		1	1	1	1	2
8	2	1	1	1	1	1
9	3	1	1	1	2	1
10	3	2	2	1	2	2
11	1	1	1	1	1	1
12	3	1	1	1	2	2
13	3	2	2	2	3	2
14	1	2	2	1	2	1
15	3	1	1	1	2	2
16	3	3	1	1	1	1
17	3	1	1	1	1	1
18	3	1	1	1	1	1
19	3	1	1	1	1	1
20	1	1	1	1	1	1
21	3	1	1	1	3	1
22	1	1	1	1	1	1
23	1	1	1	1	1	1
24	3	1	1	1	1	1
25	1	1	1	1	1	1
26	3	1	1	3	3	3
28	1	1	1	1	1	1
29	2	1	1	1	1	1
30	2	3	2	1	2	2
31	3	2	1	1	2	3
32	2	3	1	2	1	1
33		1	2	1	1	1
34	1	1	1	2	2	1
35	1	1	1	1	2	2
36	3	2	2	1	1	2
37	1	1	1	1	1	1
38	1	1	1	1	2	1
39	1	1	1	1	1	1
40	2	3	1	1	1	2

Plant activity						
ID	Head and neck	Trunk	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	4	3	3	3	3	4
2	4	4		4	3	4
3	4	4	3	3	4	3
4	4	3	3	3	4	3
5	4	4	3	3	3	3

6	4	4	3	3	3	3
7	3	3	3	3	3	3
8	4	4	3	3	3	3
9	3	3	2	3	2	3
10	3	3	3	3	3	4
11	4	4	3	3	3	3
12	4	4	3	3	4	3
13	4	3	3	3	4	3
14	4	2	2	2	3	3
15	4	4	3	3	3	3
16	4	4	3	3	2	3
17	4	3	3	3	3	3
18	3	4	3	3	4	3
19	4	4	3	3	3	3
20	4	3	3	3	3	4
21	4	3	3	3	3	3
22	4	4	3	3	3	3
23	4	4	3	3	3	3
24	3	3	3	3	3	3
25	3	4	2	3	3	3
26	3	3	3	2	2	3
28	3	3	2	2	3	2
29	3	4	2	3	3	3
30	4	3	2	2	3	2
31	2	4	2	2	2	2
32		2	2	2	2	2
33		4	3	2	3	1
34	3	2	3	2	2	2
35	3	3	2	2	2	2
36	4	4	3	4	3	3
37	3	3	3	3	3	3
38	3	3	2	3	2	3
39	4	3	3	2	2	2
40	4	3	2	2	3	3

Animal activity						
ID	Head and neck	Trunk	Left forelimb	Right forelimb	Left hindlimb	Right hindlimb
1	1	1	1	1	1	1
2	1	1		1	1	1
3	1	1	1	1	1	1
4	1	1	1	1	1	1
5	1	1	1	1	1	1
6	1	1	1	1	1	1
7	1	1	1	1	1	1
8	1	1	1	1	1	1
9	1	1	1	1	1	1

10	1	1	1	1	1	1
11	1	1	1	1	1	1
12	1	1	1	1	1	1
13	1	1	1	1	1	1
14	1	1	1	1	1	1
15	1	1	1	1	1	1
16	1	1	2	1	1	1
17	1	1	1	1	1	1
18	1	1	1	1	1	1
19	1	1	1	1	1	1
20	1	1	1	1	1	1
21	1	1	1	1	1	1
22	1	1	1	1	1	1
23	1	1	1	1	1	1
24	1	1	1	1	1	1
25	1	1	1	1	1	1
26	1	1	1	1	1	1
28	2	1	1	1	1	1
29	1	1	1	1	1	1
30	1	1	1	1	1	1
31	1	1	1	1	1	1
32	1	1	1	1	1	1
33	2	1	1	1	1	1
34	1	1	1	1	1	1
35	1	1	1	1	1	1
36	2	1	1	1	1	1
37	1	1	1	1	1	1
38	1	1	1	1	1	1
39	1	1	1	1	1	1
40	1	1	1	1	1	1

Table 2: Skeletal inventory								
Axial skeleton								
ID	Cranium	Mandible	Cervical verts	T and L verts	Sacrum	Coccygeal	Sternum	Ribs
1	0.5	0.5	6	16	4	0	0	27
2	0.5	0.5	1	1	0	0	0	10
3	0.5	1	7	7	0	0	3	22
4	0.5	0.5	6	17	0	0	3	26
5	0.5	0.5	7	7	2	0	0	24
6	1	1	7	10	2	3	1	27
7	1	1	7	21	0	1	0	30
8	1	1	7	21	2	1	0	30
9	1	1	7	18	4	3	0	28
10	1	1	3	5	2	0	2	24
11	0.5	1	6	15	2	0	0	15
12	1	1	7	21	2	1	1	26
13	0.5	1	7	10	0	0	1	24
14	0.5	1	2	12	4	1	0	16
15	1	1	7	18	3	1	0	22
16	0.5	1	7	17	0	1	2	20
17	0.5	1	7	14	1	0	3	24
18	1	1	7	21	2	0	5	27
19	1	1	4	7	2	0	0	21
20	0.5	0.5	3.5	4	2	0	0	22
21	1	1	7	21	2	1	2	30
22	0.5	0.5	2	3	0	0	2	17
23	0.5	0.5	4	11	1	4	1	24
24	0.5	1	6	6	0	0	1	22
25	0.5	1	5	14	2	0	1	26
26	0.5	1	5	9	4	2	3	20
28	0.5	1	7	20	4	3	1	30
29	1	1	7	18	2	1	1	29
30	0.5	1	6	21	4	1	0	30
31	0.5	0.5	4	13	2	1	0	27
32	0.5	0.5	2	6	2	4	0	17
33	0	0	0.5	2	0	0	0	8
34	1	1	7	20	3	5	1	28
35	1	1	7	22	2	5	1	32
36	0.5	0.5	1	10	1	0	0	18
37	1	1	3	20	2	5	0	27
38	0.5	1	7	19	2	1	0	29
39	0.5	0.5	7	15	2	0	1	27
40	0.5	0.5	2	16	2	2	2	29
Av.	1	1	5	13	2	1	1	24

Appendicular skeleton: forelimbs								
	Scapulae	Pelvis	Humeri	Ulnae	Radii	Carpals	Metacarpals	Phalanges
1	2	5	2	2	2	3	6	2
2	1	4	1	1	1	2	2	1
3	2	5	2	2	1	1	3	0
4	2	6	2	1	1	0	0	0
5	1	6	2	0	1	0	0	0
6	2	6	2	2	2	4	2	5
7	2	6	2	2	2	4	7	3
8	2	6	2	2	2	6	6	9
9	2	6	2	2	2	5	7	4
10	2	6	2	2	2	3	6	5
11	2	6	2	2	2	0	0	0
12	2	6	2	1	1	3	5	4
13	2	6	2	2	2	3	8	6
14	2	6	2	2	2	2	4	0
15	2	6	2	2	2	6	8	6
16	2	6	2	2	2	3	8	10
17	1	6	2	2	2	5	5	2
18	2	6	2	2	2	1	3	0
19	2	6	2	2	2	6	8	7
20	2	6	2	1	1	0	0	0
21	2	6	2	2	2	5	8	10
22	2	5	2	1.5	1.5	0	0	2
23	2	6	2	2	2	4	6	5
24	2	4	2	2	2	4	5	2
25	2	6	2	2	2	1	5	3
26	2	6	3	3	3	7	6	0
28	2	6	2	2	2	3	8	3
29	2	6	2	2	2	3	6	2
30	2	6	2	2	2	0	3	1
31	2	6	2	2	2	3	3	2
32	2	4	2	2	2	8	3	0
33	1	2	1	1	1.5	3	1	0
34	2	6	2	2	2	3	3	2
35	2	6	2	2	2	9	3	3
36	1.5	5	2	1.5	1.5	0	1	0
37	2	6	2	2	2	4	7	4
38	2	6	2	2	1	0	2	0
39	2	6	2	2	1	6	0	2
40	2	6	1	1	1	0	1	0
Av.	2	6	2	2	2	3	4	3

Appendicular skeleton: hindlimbs						
	Femora	Tibiae	Fibulae	Tarsals	Metatarsals	Phalanges
1	2	1	0	0	0	0
2	2	2	1	4	1	0
3	2	1.5	0	0	0	0
4	1	2	0	3	1	0
5	2	2	1	8	7	10
6	2	2	1	6	4	2
7	2	1	1	4	0	0
8	2	2	1	6	6	3
9	2	2	2	8	8	8
10	2	2	2	4	6	4
11	2	2	1	6	3	0
12	2	2	2	6	7	10
13	2	2	1	3	1	0
14	2	2	2	6	3	0
15	2	2	1	7	5	5
16	2	2	2	9	8	2
17	2	2	1	7	4	0
18	2	2	1	2	0	0
19	2	2	2	6	2	2
20	2	2	1	3	0	0
21	2	2	2	12	8	11
22	2	2	2	2	0	2
23	2	2	2	5	3	2
24	2	2	1	9	5	4
25	2	1	1	5	2	2
26	2	2	1	8	4	2
28	2	2	2	6	8	7
29	2	2	2	6	6	1
30	2	2	2	8	2	0
31	2	2	2	6	3	3
32	2	2	1	5	3	3
33	2	0	0	0	0	0
34	2	2	2	6	7	2
35	2	2	1	6	6	4
36	2	1.5	0	2	0	1
37	2	2	2	5	1	1
38	1	2	1	2	0	0
39	2	2	1	4	0	0
40	2	2	1	0	4	4
Av.	2	2	1	5	3	2

	Percent complete (%)
1	40.1
2	17.8
3	29.7
4	35.6
5	40.1
6	46.5
7	48.0
8	58.4
9	60.4
10	42.6
11	33.4
12	55.9
13	41.3
14	35.4
15	54.0
16	53.7
17	45.3
18	44.1
19	43.1
20	26.0
21	68.8
22	24.3
23	45.0
24	40.8
25	42.3
26	46.3
28	60.1
29	51.5
30	48.3
31	43.6
32	35.1
33	11.4
34	54.0
35	59.9
36	24.8
37	50.0
38	39.9
39	41.1
40	39.1
Av.	42.5

Table 3: Soil composition at three different levels of 39 graves and the control							
	pH				Organic content		
	10 cm	At level	Below level		10 cm	At level	Below level
Control	4.62	5.66	5.77		3.6952	3.4989	3.6215
1	4.89	4.55	4.35		4.0642	5.4907	5.3470
2	4.63	4.59	4.55		5.0273	4.7191	3.8680
3	4.94	4.57	4.89		4.6237	4.5849	4.7458
4	5.09	4.80	4.76		3.6286	5.3470	4.1427
5	5.27	4.70	4.98		3.9828	4.4593	5.3531
6	4.79	4.03	3.98		5.5085	5.5432	4.6189
7	4.38	3.81	4.46		4.0174	6.8235	4.5556
8	4.56	4.03	3.87		4.0625	4.7782	4.8894
9	4.61	3.88	4.00		5.1600	4.9162	5.4084
10	4.22	4.37	5.04		4.3379	4.3636	4.8673
11	3.96	4.43	4.38		4.6409	5.4023	5.2692
12	3.95	3.65	4.14		5.4466	8.7990	5.4407
13	4.27	4.41	4.95		7.7009	4.9020	4.9830
14	4.63	4.26	3.91		4.9462	4.5657	4.6893
15	4.05	4.51	4.77		3.9387	4.7235	4.8780
16	4.41	4.18	3.42		4.9550	5.0000	5.7842
17	4.54	3.13	4.48		4.2530	5.2879	5.0971
18	4.79	4.54	3.69		5.1118	6.4334	6.2796
19	4.47	4.33	4.67		2.5229	6.0255	5.0351
20	3.98	4.37	4.67		6.1179	4.9605	4.5606
21	4.40	3.66	5.34		3.6994	5.4264	5.5363
22	4.44	4.26	4.14		3.6442	4.8423	5.6054
23	4.51	4.48	4.07		4.0130	4.3728	4.4237
24	4.24	4.10	5.34		4.0816	4.1804	4.9412
25	4.52	3.63	3.49		4.1371	6.1521	5.5814
26	4.56	3.91	3.77		4.0043	4.8780	4.9354
28	4.51	4.71	4.61		4.7619	4.2874	4.1469
29	4.28	3.82	3.63		3.5834	4.5977	7.6190
30	4.41	3.83	3.99		4.3084	4.8327	4.4365
31	4.74	4.19	4.33		3.3722	4.8303	4.3275
32	4.32	4.23	4.96		4.6380	7.0773	5.2260
33	4.73	4.84	4.66		3.8889	5.0057	3.7904
34	4.76	4.33	4.38		14.8724	6.0345	6.4554
35	4.95	4.55	4.43		5.5490	6.8796	5.2695
36	4.82	5.92	4.49		4.0586	4.6729	4.9080
37	5.13	4.74	4.28		4.3931	4.4444	5.0375
38	4.42	3.42	4.95		4.2093	19.7756	3.9517
39	4.95	4.78	5.24		4.0659	4.8387	5.0542
40	5.20	5.37	5.01		4.2141	4.1866	5.1621

Table 4: Observations noted during the excavation					
ID	Depth (cm)	Date of excavation	Air pockets	Black soil	Additional info
1	77	19/09/2021	-	-	-
2	76	19/09/2021	-	Thorax	-
3	88	19/09/2021	-	-	-
4	68	19/09/2021	-	-	-
5	74	26/09/2021	-	-	-
6	72	24/09/2021	-	Thorax	-
7	90	24/09/2021	-	-	-
8	78	24/09/2021	Skull	-	-
9	80	26/09/2021	-	-	Possible scavenger hole
10	65	24/09/2021	-	-	-
11	72	05/09/2021	Ribs	Abdomen	Bone disintegration
12	62	05/09/2021	Hindlimbs	-	Hard and dry soil
13	57	24/06/2021	-	Abdomen	-
14	68	24/06/2021	Multiple throughout the grave	-	Possible scavenger hole
15	81	31/07/2021	Limbs	Abdomen	-
16	65	24/06/2021	Multiple throughout grave	Abdomen	Possible scavenger hole
17	69	31/07/2021	-	Skull and thorax	Left ribs disintegration
18	82	22/08/2021	Skull	Abdomen	-
19	82	22/08/2021	-	Abdomen and skull	Left ribs disintegration
20	62	22/08/2021	-	-	-
21	54	24/06/2021	-	Abdomen and hindlimbs	-
22	60	24/06/2021	-	-	-
23	73	24/06/2021	Ribs	Thorax and abdomen	-

24	71	24/06/2021	-	Abdomen	-
25	76	16/06/2021	Ribs	Skull and abdomen	-
26	65	12/06/2021	Skull, thorax and limbs	-	-
28	65	12/06/2021	Upper ribs	Skull and abdomen	-
29	73	16/06/2021	-	Skull and abdomen	Rocky
30	66	02/05/2021	-	Abdomen and hindlimbs	-
31	76	02/05/2021	-	-	Soil very wet
32	90	17/04/2021	-	Skull, forelimb, thorax	-
33	80	17/04/2021	-	-	Covered in vegetation and black jacks, not back filled correctly
34	80	17/04/2021	-	-	Wet soil
35	75	05/04/2021	Limbs	-	Wet soil, lots of rocks
36	65	22/03/2021	-	Skull, thorax and abdomen	Many large black roots throughout the grave
37	67	02/04/2021	Thoracic vertebrae	-	Many small rocks embedded around the skeleton, large black roots
38	70	02/04/2021	-		Ribs and phalanges far away, rocky
39	56	02/04/2021	-	Abdominal	-
40	56	13/03/2021	-	Skull, forelimb and thorax	-

ETHICS FORM THE UNIVERSITY OF WITWATERSRAND

ANIMALS RESEARCH ETHICS COMMITTEE (AREC)



STRICTLY CONFIDENTIAL

CLEARANCE CERTIFICATE NUMBER: 2020/06/07/O

APPLICANT: Ms C Landsman

School: School of Anatomical Sciences; Department: N/A; Location: Miertjie le Roux, Experimental Farm, Pretoria

PROJECT TITLE: Skeletal taphonomy of *Sus scrofa* within a burial environment in the South African highveld

;

Category: ; Species and Numbers involved: 39X 40-80kg male/female *Sus scrofa*

Approval is hereby given for the use of animals for the research project named above and described in the application reviewed by a quorate meeting of the AREC held on 30 Jun 2020. This approval remains valid until 23 Aug 2022 and is conditional to the following (if blank there are no special conditions):

Condition 1	Condition 2	Condition 3	Condition 4

All material changes to the approved research must be reported to the AREC before they are implemented. Failure to do so will invalidate this clearance certificate.

An annual progress report must be provided to the AREC.

The use of these animals is subject to AREC guidelines on the use and care of laboratory animals, is limited to the procedures described in the application and is subject to additional conditions listed below:

I, the Chair of the AREC (or my designated representative) am satisfied that the proposed research is ethical as judged by local law, international standards and University policy.

Signed: _____  _____ Date: 25/08/2020
(Chairperson of the AREC)

I am satisfied that the persons listed in this application are competent to perform the procedures described in the application, in the context of Section 23 (1) (c) of the veterinary and Para-veterinary Professions Act (19 of 1982).

Signed: _____  _____ Date: 25 August 2020

CC: Student supervisor: «Title1» «Initials1» «Supervisor_surname»

Director Central Animals Service: Dr Kim Jardine

ANIMALS RESEARCH ETHICS COMMITTEE (AREC)



(Registered Veterinarian)

CC: Student supervisor: «Title1» «Initials1» «Supervisor_surname»
Director Central Animals Service: Dr Kim Jardine

ETHICS FROM UNIVERSITY OF PRETORIA



Faculty of Veterinary Science
Animal Ethics Committee

2 November 2020

Approval Certificate New Application

AEC Reference No.: 543/2020
Title: Skeletal taphonomy of *Sus scrofa* within a burial environment in the South African Highveld
Researcher: Miss C Landsman
Student's Supervisor: Dr J Myburgh

Dear Miss C Landsman,

The **New Application** as supported by documents received between 2020-08-31 and 2020-10-30 for your research, was approved by the Animal Ethics Committee on its quorate meeting of 2020-10-30.

Please note the following about your ethics approval:

1. The use of species is approved:

Species	Number
Pigs <i>Sus scrofa</i> (buried already) Mierjke le Roux Experimental Farm	39
Samples	
Skeletal remains	39

2. Ethics Approval is valid for 1 year and needs to be renewed annually by 2021-11-02.
3. Please remember to use your protocol number (543/2020) on any documents or correspondence with the AEC regarding your research.
4. Please note that the AEC may ask further questions, seek additional information, require further modification, monitor the conduct of your research, or suspend or withdraw ethics approval.
5. **All incidents** must be reported by the PI by email to Ms Marleze Rheeder (AEC Coordinator) within 3 days, and must be subsequently submitted electronically on the application system within 14 days.
6. As part of your approval, the committee requires that you record a **short video footage** of major animal procedures approved in your study. **The committee may request them for monitoring purposes at any later point.**

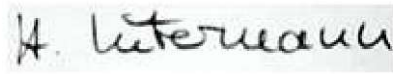
Ethics approval is subject to the following:

- The ethics approval is conditional on the research being conducted as stipulated by the details of all documents submitted to the Committee. In the event that a further need arises to change who the investigators are, the methods or any other aspect, such changes must be submitted as an Amendment for approval by the Committee.

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Fakulteit Veeartseniekunde
Lefapha la Diseanse tsa Bongakadiruiwa

We wish you the best with your research.
Yours sincerely

A handwritten signature in black ink, appearing to read 'H. Lutermann', on a light-colored rectangular background.

Dr. Heike Lutermann
DEPUTY CHAIRMAN: UP-Animal Ethics Committee

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and Metabolomic Profiles", *Frontiers in Microbiology*, 2021

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