2012

The effects of sharp-force thoracic trauma on the rate and pattern of decomposition in New England

BOSTON UNIVERSITY

SCHOOL OF MEDICINE

Thesis

THE EFFECTS OF SHARP-FORCE THORACIC TRAUMA ON THE RATE AND PATTERN OF DECOMPOSITION IN NEW ENGLAND

by

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B.A., University of Memphis, 2010

Submitted in partial fulfillment of the

requirements for the degree of

Master of Science

2012

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ACKNOWLEDGEMENTS

There have been several people that have assisted me throughout the course of this project that deserve more than just acknowledgements, but my undying thanks for the work they have done. First of all I must thank my readers, Jonathan Bethard, Dr. Jennifer Hammers, and Dr. Tara Moore, without whom, this paper could never have been written. I especially want to thank Dr. Hammers for your constant support truly has kept me going. I also wish to thank Dr. Donald Siwek and Agt. Gary Reineck for all of the help you have offered me during the actual experimentation portion. The program as a whole, for allowing me the ability to utilize Holliston facility as well as the equipment you have supplied me has been invaluable. To my fellow classmates, especially Michelle Berretta, and Alan Damiani, I want thank you for all of your help as well.

In particular, though, I want to thank Danielle Carroll, Evonne Turner-Byfield, and Mellie Riddle for you constant moral support. You, along with my parents, have been instrumental in keeping me both grounded and sane. You are the best friends a person could ever ask for and I love you all.

To one of my best friends, Amelia Boaks, I really want to thank you. You have been there with me for every step of this project. You were there in the planning, execution, and writing. Without and your support this would never have been completed. You have been there for me when it seemed like no one else was; I will never forget this. No matter where we go in life, I will always be there for you.

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ABSTRACT

One of the most difficult tasks that a forensic pathologist or anthropologist is asked to accomplish is the determination of the postmortem interval (PMI) (Megyesi *et al* 2005; Ubelaker 1996). When soft tissue is still present, this interval is largely based on the extent of decomposition. Many factors, however, may alter the rate at which decomposition occurs including the temperature, humidity, insect activity, carnivore and rodent activity, and the depositional environment (Mann *et al* 1990; Sledzik 1997). In a 1990 study Mann *et al* determined that trauma was also a factor in decomposition, rating it a 4 out of a scale of 5 in importance. The results of the Mann et al (1990) study have been widely accepted by the field and today trauma is considered a major variable affecting the rate of decomposition in textbooks and other edited volumes (Byers 2011; Komar and Buikstra 2008; Sledzik 1997).

In 2006, a study by J. A. Kelly, in South Africa, challenged the notion that trauma affects the rate of decomposition. In her dissertation, Kelly (2006) found that there was no significant difference in the rate of decomposition between traumatic groups and non-traumatic groups. In 2010, this research was further followed up by a team in

the United Kingdom, specifically analyzing the effects of penetrative trauma on decomposition (Cross and Simmons 2010). Like the South African study, the authors discovered that there were no significant differences between a traumatic group and a non-trauma control (Cross and Simmons 2010; Kelly 2006). However, serious questions can be raised about these studies including the method of euthanasia, and the number of experimental subjects used (Cross and Simmons 2010; Kelly 2006).

This present study utilized eight porcine carcasses to determine the effects of trauma on the rate and pattern of decomposition in the New England area. Three of the subjects were lacerated with a 15cm long incision penetrating in the thoracic cavity and three other subjects where lacerated with a 15cm long incision in the thoracic area but the incision did not penetrate into the cavity. A seventh set of remains was utilized as a control with an eighth used to verify the results. The subjects were placed on a surface depositional environment at the Boston University Research Facility in Holliston, MA from June to August 2011. Because factors such as temperature are so variable and can affect the temporal rate of decomposition, this study utilized the accumulated degree day (ADD) published in Megyesi *et al* (2005) as a measure of time.

In addition, qualitative and semi-quantitative analyses were conducted, relying predominantly on the total body score (TBS) developed by Megyesi *et al* (2005). This system assesses a score, based on a stage of decomposition, for three specific regions of the body: head and neck, trunk, and limbs (Megyesi *et al* 2005). The sum of these scores is the total body score for a particular time. The present study assessed the TBS of all

eight subjects and compared them on a temporal, ADD, and accumulated humidity day (AHD) bases.

Following the experiment, a repeated measures analysis of variance (ANOVA) was conducted to determine if there was a statistical difference between the three subject groups. The results of this analysis revealed that there was no significant difference between the penetrated group, non-penetrated group, and control group. Trauma had no significant value in the rate of decomposition. A difference, however, was seen in the pattern of decomposition, with decomposition beginning at the wound site in traumatic groups and the facial region for the non-trauma group.

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LIST OF ABBREVIATIONS

°C	Degree Celsius
ADD	Accumulated Degree Day
AHD	Accumulated Humidity Day
ANOVA	Analysis of variance
ARF	Anthropological Research Facility, The University of Tennessee-Knoxville
ATP	Adenosine triphosphate
C	Control
D	Disturbed
GPS	Global Positioning System
IACUC	Institutional Animal Care and Use Committee
MA	Massachusetts
N	Non-penetrative
NOAA	National Oceanic and Atmospheric Administration
P	Penetrative
PMI	Postmortem Interval
TBS	Total Body Score
U	Undisturbed
USHoRJC	U. S. House of Representatives Judiciary Committee

CHAPTER 1: INTRODUCTION

When working with skeletal or otherwise badly decomposed remains, the primary responsibilities of the forensic anthropologist are those of assessing the age, sex, and ancestry (also known as the biological profile), assessing traumatic injury and determining their timing and potential cause, and lastly, establishing a viable post-mortem interval (PMI). While no one of these areas is simple to determine, due to the number of variables acting on the decomposition process, PMI determination is one of the most difficult. Temperature, humidity, insect access and activity, animal activity, rainfall, sunlight exposure, soil pH, depositional environment, and trauma have all been listed as factors that affect decomposition. Recent questions have arisen, however, regarding the process of determining the PMI, especially in cases involving human remains with inflicted trauma (Cross and Simmons 2010; Kelly 2006; Mann *et al.* 1990).

Since the early 1990s, the presence and degree of trauma to the remains has been considered a major contributor to the decomposition process. In 1990, Mann *et al* described the factors effecting decomposition based upon the review of numerous studies measuring the effects of a variety of factors on decomposition (see Table 1). Each factor was then scored by its relative impact on the overall decompositional process. It was within this review that trauma, specifically penetrative trauma, was identified as a significant factor in the decomposition process. Despite scientific limitations in the study, such as the use of only two subjects, the results were nonetheless widely accepted by the field.

Table 1: ***List of factors potentially effecting decomposition (modified from Mann* et al *1990)***

Factor	Type	Effect of Decay Rate
Temperature	Environmental	5
Access by insects	Faunal	5
Burial and depth	Environmental	5
Carnivores and Rodents	Faunal	4
Trauma (penetrating/crushing)	Human Activity	4
Humidity/aridity	Environmental	4
Rainfall	Environmental	3
Body size and weight	Intrinsic	3
Embalming	Human Activity	3
Clothing	Human Activity	2
Surface Placed on/in	Environmental	1
Soil pH	Environmental	unknown

"Subjective criteria rating based on a five-point scale. 5 being the most influential."

In 1993, the Supreme Court of the United States heard the case of *Daubert vs. Merrell Dow Pharmaceuticals*, the result of which has been coined the "Daubert Guidelines." The guidelines govern the admissibility of scientific evidence and testimony in all federal and most state courts. With these guidelines in mind, many researchers in the field of forensic anthropology have developed studies to further investigate many existing procedures, methodologies and information used in case work. Included in these studies has been a re-examination of the factors that affect the decompositional process. Some of these factors have withstood greater scientific scrutiny, and indeed were enhanced by them. The assessment of the effect of trauma on decomposition, however, has become rather problematic as conflicting data has arisen.

The present study will endeavor to validate either the findings of Mann *et al* (1990), or similar studies that have borne differing results. To accomplish this, it is the purpose of this project to determine statistically if trauma is influential in the rate of decomposition. In addition, this study will determine if trauma alters the pattern of decomposition.

To answer these questions, this thesis has been designed in the following manner. In chapter two, the author discusses the relevant history of decompositional research as well as the research investigating the various factors that affect decomposition. The chapter is concluded with a section describing the research relating to trauma and decomposition, including those reports that have confirmed Mann et al (1990) and those who have reported contradictory results. In the third chapter of this thesis, the author describes the various methods and materials utilized in the current project. Included is a synopsis of the environment where the project was conducted as well as on the methodologies used to measure decompositional rates and patterns. Chapter four contains a description of the results of the project, particularly with regards to the group comparison and the statistical results; the individual results are in the appendix. The fifth chapter of this text includes the discussion section which readdresses the questions regarding the results and conclusions of Mann et al (1990) as well as other studies.

CHAPTER 2: BACKGROUND AND LITERATURE REVIEW

History of Decomposition Research

Decompositional studies are not a recent addition to the anthropological literature, but the methodologies have changed over the years. Prior to Jerry Payne's 1965 landmark carrion study on juvenile *Sus scrofa*, decompositional studies had mainly focused on taphonomic processes and arthropod succession in a more entomological (biological) context (Micozzi 1991; Payne 1965). Early studies of soft tissue decomposition were first published in 1965, when Payne completed an arthropod succession study as it related to the decompositional process (Payne 1965). An early milestone in the arena of human decompositional research occurred in 1972 when Dr. William Bass opened the Anthropological Research Facility (ARF) at the University of Tennessee – Knoxville Health Science Center (Bass 1997; Marks and Tersigni 2005; Micozzi 1991; Rodriguez and Bass 1983). After the opening of the ARF, a number of studies investigating the decompositional process were completed, including studies regarding those factors that affect decomposition (Bass 1997; Mann *et al.* 1990; Rodriguez and Bass 1985; Rodriguez and Bass 1983).

For the early part of the history of decompositional studies, the vast majority have been conducted at the ARF in Knoxville. However, within the last decade, a greater emphasis has been placed on investigating the regional aspects of decomposition as studies at the ARF reveled, through the understanding of how climactic factors, such as temperature, and humidity affect the decompositional process, that more regional studies

must be conducted. For example, the temperature and climates of central Texas, New England, the Pacific Northwest, the Central United Kingdom, etc. vary drastically from Tennessee (DeCota 2011; Joy *et al.* 2006; Mann *et al.* 1990; Parks 2011; Prieto *et al.* 2004; Sharanowski *et al.* 2008; Shean *et al.* 1993; U.S. Department of Commerce) and therefore the rate and process of decomposition would be different in each of these regions.

When examining and classifying decompositional studies, one can essentially place them into one of two methodological categories: experimental studies and surveys. Experimental studies, as the name implies, are those in which physical experiments, either qualitative or quantitative, are conducted on the remains of a mammalian subject, traditionally with humans or porcine models, such as the traumatic injury study conducted by Cross and Simmons (2010) (Bachmann and Simmons 2010; Cross and Simmons 2010; Rodriguez and Bass 1985; Simmons *et al.* 2010). A survey study is conducted by the author examining reported case data to develop a new methodology, as Megyesi (2005) utilized accumulated degree days to establish a post-mortem interval (Galloway 1997; Galloway *et al.* 1989; Janaway *et al.* 1995; Komar 2003; Megyesi *et al.* 2005).

Daubert, Kumho **and the Shift in Decompositional Research**

Since the early 1920's scientific evidence in the U.S. federal and state courts has followed the *Frye*, or general acceptance, test (293 F 1013 1923; Grivas and Komar 2008; Komar and Buikstra 2008). Under this test, scientific testimony can be accepted

as evidence if the premise and methodology in question have been accepted as standard practice within the field (293 F 1013 1923; Komar and Buikstra 2008). In 1993, however, a major shift occurred in the field, particularly in the manner and method that forensic research accepted new premises and claims. It was then that the Supreme Court of the United States issued the landmark *Daubert* ruling, which was subsequently followed up by the *Kumho* ruling in 1997. Both of these rulings required the forensic science fields to quantify and modify accepted standards in order to be accepted by the federal court system, as well as many state court jurisdictions that had adopted the *Daubert* rule over *Frye* (509 US 579 1993; 526 US 137 1999; Christensen 2004; Christensen and Crowder 2009; Grivas and Komar 2008).

The ruling in *Daubert v. Merrell Dow Pharmaceuticals, Inc* essentially created five guidelines for evidence to be accepted in court, commonly refered to as the "*Daubert* critera,". The guidelines required that (1) the methodology in question must be testable and developed through the scientific method, (2) subject to peer reivew, (3) establish standards by which the methodology can be tested, (4) publish a known or potential error rate, and (5) which has been widely accepted by the particular field (509 US 579 1993; Christensen 2004; Christensen and Crowder 2009; Grivas and Komar 2008).

The case was a class-action suit filed against Merrell Dow Pharmacuticals by the parents of Jason Daubert and Eric Schuller, both of whom had been born with "serious birth defects" (509 US 579 1993). The parents of the two minors argued that the drug Bendectin, which was developed by Merrell Dow, caused the defects. The parents

argued, via scientific experts, that the safety tests conducted by Merrell Dow Phatmaceuticals were inadequate. The respondants (Merrell Dow) countered with their own scientific experts, claiming that animal models for hazards testing were sufficient for the medical field; to which the court agreed with Merrell Dow (509 US 579 1993).

In 1999, the U.S. Supreme Court issued a caveat to the *Daubert* critera in the form of it's decision in *Kumho Tire Co. v. Carmichael* which allowed for observational testimony to be introduced based on experience and deemed the *Daubert* critera to be more "guidelines" than hard rules (526 US 137 1999; Grivas and Komar 2008). This case allowed for those sciences that cannot work in a strictly quantifiable methodology, such as forensic anthropology and pathology, that the experience of the observer hold equal weight to that of the methodology (Grivas and Komar 2008). For example, if a set of skeletal remains were anaylized, and contained a defect similar to a traumatic fracture, an anthropologist can testify as to the potential cause of the fracture, given their observations and their individual experince dealing with such defects.

The suit was brought by Patrick Carmichael against Kumho tire following a fatal vehicle crash caused by a tire blowout. The petitioners (Carmichael) produced an expert who utilized the technical information from the respondents (Kumho Tire Co.). While the technical information was not disputed, the methodology as well as the expertise of the analyst was (526 US 137 1999). It was determined by the court that there is no clear line between "scientific" and "technical" testimony and the testimony was accepted (526 US 137 1999; Grivas and Komar 2008). Furthermore, in response to conflicts within the

Federal Rules of Evidence, *Daubert* asserted that it was ultimately the role of the judge to be the "gatekeeper" of what testimony was allowed; *Kumho* reaffirmed this assertion (509 US 579 1993; 526 US 137 1999; U. S. House Committee on the Judiciary Committee 1994; Grivas and Komar 2008).

As a result of these two cases, the field of forensic anthropology has been forced to re-examine the standards used to establish post-mortem interval, including the methodologies utilized in decompositional studies (Christensen and Crowder 2009; Grivas and Komar 2008). These re-examination and verification/validation studies have, for the most part confirmed those standards, but have thrown others into some doubt.

Decompositional Processes

In order to understand the factors that affect decomposition, one must first understand the process of decomposition. The decompositional process is a two level process. Internal decomposition which occurs at a predominantly cellular level; and external decomposition, which impacts mostly the external soft tissue (skin) changes and skeletalization, or the amount of bone exposed (Bass 1997; Cabirol *et al.* 1998; Carter *et al.* 2007; Clark *et al.* 1997; Dekeirsschieter *et al.* 2009; Galloway 1997; Janaway 1995; Pinheiro 2006; Sledzik 1998; Vass 2001). Both processes have been organized into series, with scales created to assist in determining post-mortem interval (Bass 1997; Clark *et al.* 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005; Prieto *et al.* 2004; Rodriguez and Bass 1983). It should be noted that while a basic scale exists, none of the aforementioned scales are precise, and have either overlapping

sequences, or have differing names for identical sequences (see Table 2). The basic categories of decomposition, as defined in the literature, include the following: fresh, bloat, early decay, advanced decay, and skeletalization (Clark *et al.* 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Rodriguez and Bass 1983).

Table 2: ***Stages of Decomposition***

Rodriguez and Bass (1983)	**Galloway *et al* (1989)**	**Megyesi *et al* (2005)**
Fresh	Fresh	Fresh
Bloated	Early Decomposition	Early Decomposition
Decay (Active)	Advanced Decomposition	Advanced Decomposition
Dry	Skeletalization Extreme Decomposition	Skeletalization

Fresh

Beginning approximately four-minutes post-mortem, the cells begin going through a process of autolysis (Vass 2001). During this period, the lysosomes in the cells are released into the cytoplasm because of a decrease in the intercellular pH level caused by the deprivation of oxygen (O_2) and adenosine triphosphate (ATP) (Clark *et al.* 1997; Dautartas 2009; Gill-King 1997; Vass 2001). Subsequently the cell walls begin to deteriorate and the intercellular junctions begin to dissolve causing a breakdown in the structural tissues (Dautartas 2009; Gill-King 1997; Marks and Tersigni 2005; Vass 2001). Early in the process, livor mortis sets in due to the capillary beds and circulatory vessels breaking apart, causing a pooling of blood in the gravitationally dependent portions of the body (Burton 1974; Clark *et al.* 1997). The lividity of the body causes the skin to take on

a pale appearance, save in those areas where the blood has pooled which tends to be more dark pink in hue (Bass 1997; Burton 1974; Clark *et al.* 1997; Galloway *et al.* 1989).

Once the intrinsic autolysis process is multiplied to the wider tissue level, approximately forty-eight to seventy-two hours, intestinal and endogenous bacteria initiate putrefaction (Dautartas 2009; Dekeirsschieter *et al.* 2009; Janaway 1995; Vass 2001). During this stage, the digestive tract (intestines, stomach, and other accessory organs) begins to break down giving a green discoloration, often first seen in the abdomen (Clark *et al.* 1997; Galloway *et al.* 1989; Gill-King 1997). This breakdown of the intestinal tract leads to the second universally accepted stage of decomposition, the bloat stage (Clark *et al.* 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Sledzik 1998).

Bloat

During the process of putrefaction, anaerobic bacteria become increasingly active, engaging both the blood, causing a "marbling" appearance to the skin, and abdominal organs (Bass 1997; Clark *et al.* 1997; Komar and Buikstra 2008). This activity in the abdominal cavity, combined with the loss of intestinal and gastric wall integrity, causes a release of excess gasses, resulting in abdominal distention (Bass 1997; Clark *et al.* 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005; Rodriguez and Bass 1983; Sledzik 1998). This deterioration further adds to both the discoloration of the remains, spreading from the abdomen to the remainder of the body, as well as distention in the limbs and head (Bass 1997; Galloway *et al.* 1989; Megyesi *et al.* 2005; Rodriguez

and Bass 1983). In addition to the general discoloration the remains tend to have an intense odor as well (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Rodriguez and Bass 1983).

Early (or Active) Decay

Following the ultimate release of the bloat gases, the intrinsic decompositional processes will have slowed and the extrinsic processes will begin to accelerate (Bass 1997; Clark *et al.* 1997; Galloway *et al.* 1989; Rodriguez and Bass 1983). During the previous three phases, extrinsic activity including arthropod oviposition as well as other insect and macrofaunal activity generally takes place; this activity is greatly enhanced and now engages in a more active role in the decompositional process (Galloway 1997; Galloway *et al.* 1989; Komar 2003; Komar and Buikstra 2008). Larvae activity at this stage accelerates as the maggots grow from their first instars to their second and third instar stages and begin to form maggot masses, or a large collection of maggots numbering upwards into the thousands (Campobasso *et al.* 2001; Catts 1992; Catts and Goff 1992; Dadour 2011; Galloway *et al.* 1989; Introna *et al.* 1991; Komar and Buikstra 2008; Lord *et al.* 1994; Marks and Tersigni 2005; Sharanowski *et al.* 2008). During this stage the thoracic and abdominal cavity collapse (referred to as "post-bloat") and the skin turns from a greenish hue to one that is much darker including browns and even black (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005). In addition to the discoloration of the skin, during the early decay stage the putrefactive fluids begin to purge from the open orifices of the remains, leading to the

death of immediately surrounding vegetation and forming a "decomposition ring" around the remains (Bass 1997; Dadour 2011; Galloway *et al.* 1989; Megyesi *et al.* 2005).

Advanced Decay

The final stage of soft tissue decomposition is that of "advanced decay" (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005; Rodriguez and Bass 1983). It is during this stage that the maggot masses leave the body and burrow themselves into the ground and begin to pupate (Bass 1997; Catts 1992; Catts and Goff 1992; Komar and Buikstra 2008; Rodriguez and Bass 1983). Bone exposure beings to appear in all regions of the remains and the remaining soft tissues begin a desiccation process. This process can ultimately lead to the remains either skeletonizing completely or, should a majority of the remaining soft tissue desiccate rather than disintegrate, mummifying (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005). It should be included that, while the literature is divided, most studies that attempt to categorize the decompositional process into stages ultimately place mummification into the "advanced decomposition" stage (Bass 1997; Galloway *et al.* 1989; Megyesi *et al.* 2005).

Skeletalization

The final stage of decomposition is skeletalization. During this stage the majority of the soft tissue has deteriorated or been consumed, leaving only the remaining hard tissues behind (Bass 1997; Dautartas 2009; DeCota 2011; Galloway *et al.* 1989; Janaway

1995; Komar and Buikstra 2008; Marks and Tersigni 2005; Marks *et al.* 2009; Megyesi *et al.* 2005). Once attaining this terminal stage, the remains, theoretically can remain in this stage indefinitely (Komar and Buikstra 2008). Within the skeletalization stage, several sub-stages exist, depending on how long they have been in the state. In the first form, or initial skeletalization, the skeleton generally has a "greasy" appearance with limited soft or desiccated tissue remaining (Bass 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005). Given time, the remaining soft tissue deteriorates leaving only the hard tissue (bone) remaining for the second stage. Though the literature does not adequately come to a consensus on a definition of this stage, it can be termed as the "wet bone" stage because the remaining bone has a greasy texture to it generally, but may be in the process of drying (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Megyesi *et al.* 2005). The third sub-stage of skeletalization is that of the "dry bone" stage where the greasy texture of the bone has dried out leaving bone that is "wood-like" in appearance. Given a short amount on time in the sun (as early as a couple of days) these dry bones become bleached and take on a rather white, chalky hue (Bass 1997; Galloway 1997; Galloway *et al.* 1989; Komar and Buikstra 2008; Megyesi *et al.* 2005). Should the remains continue to be exposed to the elements, normal taphonomic processes will proceed with the skeletal elements deteriorating due to weathering (Bass 1997; Behrensmeyer 1978; Galloway *et al.* 1989).

Factors Effecting Decomposition

Decomposition is a natural process that the remains of all terrestrial organisms undergo after death, but is not as simple as passing from one phase to another in a linear fashion. Many factors influence the process of decomposition, predominantly the rate and pattern of decomposition. Within the decompositional process, ambient temperature, humidity, depositional environment, moisture/rainfall content, access of the remains to insect and carnivore/scavenger fauna, the amount of sun or shade, and body weight are factors that can alter the rate of decomposition. Higher ratios of any one factor can either accelerate or retard that rate. (Breitmeier *et al.* 2005; Cross and Simmons 2010; Dautartas 2009; DeCota 2011; Mann *et al.* 1990; Rodriguez and Bass 1985; Rodriguez and Bass 1983; Shean *et al.* 1993; Vass *et al.* 1992)

Temperature

Of all of the factors affecting the rate of decomposition, temperature has the most significant affect (Mann *et al.* 1990; Sorg *et al.* 1998). This one singular factor has a direct impact on subsequent factors such as insect activity and can even accelerate or decelerate the amount of bacterial activity required for putrefaction (Catts 1992; Gill-King 1997; Gill 2005; Introna *et al.* 1991; MacAulay *et al.* 2009; Mann *et al.* 1990; Micozzi 1986; Rodriguez and Bass 1983; Sharanowski *et al.* 2008). With an increase in the temperature, a direct correlation between the amount of bacterial and insect activity has been observed and this increase then accelerates the rate at which decomposition occurs. Conversely, a temperature drop below a specific threshold, approximately around

5-13°C, results in a slowing of insect ovipositor activity, resulting in fewer larvae, the primary instigator in the early and advanced stages of decomposition (Bachmann and Simmons 2010; Galloway 1997; Galloway *et al.* 1989; Komar 1999; Komar 1998; Komar 2003; Komar and Buikstra 2008; Mann *et al.* 1990; Simmons *et al.* 2010). Beyond simply a cool ambient temperature decelerating insect activity to the remains, freezing temperatures (below 0°C) cease the natural intrinsic decompositional process by freezing the cytoplasm found within the cells, thus preventing the process of autolysis and ultimately putrefaction (Micozzi 1986).

In Mann *et al.* (1990), the authors ranked twelve factors that can potentially affect the rate of decomposition; temperature was one of three factors that received their highest rating of 5 (access by insects and burial/deposition being the other two). This rating has, since the issuance of *Daubert* and *Kumho*, been confirmed by several other studies as the primary factor determining the rate of decomposition (Bachmann and Simmons 2010; Cross and Simmons 2010; Galloway 1997; Komar 1999; Komar 1998; Komar 2003; Komar and Buikstra 2008; Megyesi *et al.* 2005; Micozzi 1986; Simmons *et al.* 2010).

Humidity

Humidity has also been shown to be another significant factor that can affect the rate at which remains decompose (Galloway 1997; Galloway *et al.* 1989; Komar 1999; Komar 1998; Komar 2003; Mann *et al.* 1990; Payne 1965; Statheropoulos *et al.* 2005). Even though humidity has some effect on insect activity, the greatest area humidity

effects the decompositional process is in soft tissue desiccation (Mann *et al.* 1990; Payne 1965). As the humidity in the ambient air is increased, the soft tissue retains moisture allowing the putrefactive processes to continue. This process enhances microbial activity, creating an ideal environment for maggots, fostering soft tissue loss (Dekeirsschieter *et al.* 2009; Mann *et al.* 1990; Statheropoulos *et al.* 2005). Conversely, in climates that are more arid, the lack of ambient humidity draws out the moisture content of the soft tissues, thus decelerating the putrefaction process. This deceleration decreases significant maggot activity and therefore soft tissue desiccation, leading ultimately to mummified remains rather than skeletalization (Galloway 1997; Galloway *et al.* 1989; Komar 1999; Komar 1998; Komar 2003; Mann *et al.* 1990; Statheropoulos *et al.* 2005).

Moisture

An additional minor factor in the determination of the rate of decomposition can be found in the presence or absence of moisture within and surrounding the body (Aturaliya and Lukasewycz 1999; Carter *et al.* 2010). In the study on the effect of moisture on decomposition, the authors found that an absence of moisture, in both the surrounding area and within the organic cellular matrices, was the primary deterrent of desiccation. They found that the enzymes for the creation of putrefactive fluids required an aqueous medium (Aturaliya and Lukasewycz 1999). Similarly, in a further study on the presence of moisture content within soils and how that can affect the rate of decomposition, determined that moisture, particularly in soils, can accelerate the process of decomposition by increasing the enzyme activity, which in turn can extend the

putrefaction processes. However, as the authors note, too much moisture can, instead of increasing the amount of enzyme activity, decrease the amount of CO_2, thus retarding the enzyme activity and the decomposition rate (Carter *et al.* 2010).

Sun/Shade

A fourth major environmental factor that can affect the rate of decomposition is the amount of exposure of the remains to sunlight (Shean *et al.* 1993). This factor, when combined with the ambient temperature and humidity, is the catalyst in determining the rate of decomposition and whether remains will become mummified or skeletonized (Galloway *et al.* 1989; Janaway 1995; Komar 1999; Komar 1998; Komar 2003; Shean *et al.* 1993). In general, the amount of sun to which the remains are exposed will naturally increase the temperature surrounding the remains, while a more shaded environment will decrease the ambient temperature (Shean *et al.* 1993). In a more humid climate, increases in the amount of sunlight and temperature will accelerate the putrefaction process and maggot activity. However, an increase in temperature in a more arid climate will accelerate the desiccation process, which in turn accelerates the ultimate process of mummification (Galloway *et al.* 1989; Komar 1999; Komar 1998; Komar 2003).

Depositional Environment

A fourth major factor in determining the rate of decomposition is the depositional environment (Campobasso *et al.* 2001; Mann *et al.* 1990). The three major depositional environments are aquatic, subterranean, and terranean (surface), which can affect the

decompositional rate and pattern for a set of remains (Boyle *et al.* 1996; Breitmeier *et al.* 2005; Manhein 1996; Rodriguez 1996; Rodriguez and Bass 1985; Schotsmans *et al.* 2011; Sorg *et al.* 1996).

With aquatic depositions, wherein the remains are completely submerged, the rate of decomposition is significantly slowed but not entirely arrested (Boyle *et al.* 1996; Rodriguez 1996; Sorg *et al.* 1996). The colder temperature of the water slows the putrefaction process, and intrinsic decomposition. However, while the intrinsic process may be slowed, particularly at greater depths, the extrinsic processes are in full force. While aerial/terrestrial insects may not be present, aquatic fauna are present and feed upon the soft tissue of the remains (Boyle *et al.* 1996; Manhein 1996; Mann *et al.* 1990; Rodriguez 1996; Sorg *et al.* 1996).

Like aquatic depositions, in the subterranean environment the processes are not totally arrested, but are decelerated. Furthermore, in both aforementioned depositions, the depth of burial plays a major role in the rate that deceleration takes place (Breitmeier *et al.* 2005; Manhein 1996; Mann *et al.* 1990; Rodriguez 1996; Schotsmans *et al.* 2011; Sledzik 1998). In the observational study by Mann *et al.* (1990), it was found that remains buried at a depth of 0.3 m to 0.6 m decomposed at a slower rate than surface decompositions and achieved skeletalization in approximately a few months to a year. Remains buried at even greater depths, 0.9 m or greater, can take years or longer to achieve that same rate of skeletalization (Mann *et al.* 1990; Schotsmans *et al.* 2011; Sledzik 1998).

Conversely, terranean depositions render the most rapid rate of decomposition (Mann *et al.* 1990). Surface decomposition allows for both intrinsic processes of putrefaction and autolysis and extrinsic processes from faunal activity to take place at a normal to accelerated rate (Bass 1997; Mann *et al.* 1990; Payne 1965; Sledzik 1998). However, the most predominant way in which a surface deposition accelerates the rate of decomposition is by allowing arthropods the greatest, and near unfettered, access to the remains (Bass 1997; Campobasso *et al.* 2001; Mann *et al.* 1990; Rodriguez and Bass 1983).

Insect Activity and Access

Other than temperature, the greatest single factor that can determine the rate at which remains decompose is the extent of access insects have to the remains (Adlam and Simmons 2007; Bass 1997; Campobasso *et al.* 2001; De Jong *et al.* 2011; Introna *et al.* 1991; Kelly 2006; Lord *et al.* 1994; Mann *et al.* 1990; Micozzi 1991; Payne 1965; Rhine and Dawson 1998; Rodriguez and Bass 1983; Simmons *et al.* 2010; Sledzik 1998). Larvae from various insects, predominantly the Diptera, feed off the bacteria in the body, which are engaged in the putrefaction processes (Campobasso *et al.* 2001; Mann *et al.* 1990). Many of the previously mentioned factors work to accelerate or decelerate decomposition by directly influencing the insect activity found on the remains. As seen in the study conducted by Payne (1965) the remains placed in an "insect proof" box mummified with minimal soft tissue loss. With the absence of insect and larval activity, only the intrinsic processes of decomposition occurred which do not affect the external

aspects of the skin (Payne 1965). It is only with insect activity that external soft tissue decomposition can take place (Mann *et al.* 1990).

Weight

The effect of carcass weight on decomposition has been scantily studied, with the exception of entomological studies. However, the studies that have been completed have produced ambiguous results. One of the first studies to investigate the effect of carcass weights on decomposition was Mann *et al* (1990) which found that a heavier subject decomposed at a 50% faster rate than their lighter counterparts. In dissention, however, several studies have shown that smaller massed individuals will decompose at a faster rate, particularly those studies by Spicka *et al.* (2011), Simmons *et al.* (2010), and Nagano and Suzuki (2007). This discordance can be explained through the fact that Mann *et al* (1990) utilized an obese individual to count for the heavier subject, which would make the excess weight lipid based which, through putrefaction, decomposes at a faster rate than other, more solid organs. In addition, the excess adipose tissues insulate the body, decelerating the cooling process of the body (Cabirol *et al.* 1998; Mann *et al.* 1990; Notter *et al.* 2009). It is clear from these studies that weight does have an impact on the rate of decomposition; in particular, as have reported, smaller massed individuals decompose at a faster rate than larger massed individuals. Dissenting to both of these, however, is a study discussed by Brand (2008) who found that, based on body mass, weight has no discernible effect of the rate of decomposition.

Clothing

Another minor factor in the rate of decomposition is the presence or absence of clothing or other wrappings on the body (Dautartas 2009; Kelly 2006; Kelly *et al.* 2009; Mann *et al.* 1990). Similar to weight, however, some disagreement exists within the field as to the exact effect coverings have on the rate of decomposition. Mann *et al.* (1990) state, that "clothing serves to protect the body from sunlight, which maggots avoid, and aids in speeding up the decay process" (Mann *et al.* 1990: 107). Meanwhile, Kelly *et al.* (2009) found that there was both an observational difference as well as a statistical correlation between the presence of coverings and the rate of decomposition. However, while the rate of decomposition was initially increased, that pace had stalled during the "advanced decomposition" stage, as described by Megyesi *et al.* (2005). The authors attributed this stall to the retention of moisture by the various coverings, which maintained the bacteria involved in putrefaction (Kelly *et al.* 2009). A further study which examined this issue was conducted by Dautartas (2009) at the ARF in Knoxville. The results of this study showed that there was an observational difference in the rate of decomposition, utilizing the total body score (TBS) as devised by Megyesi *et al.* (2005), but found that no statistical difference existed between the covered and uncovered subjects (Dautartas 2009). This is in contrast to the Kelly *et al.* (2009) study who found that there was both an observational and statistical difference between covered and uncovered remains.

Trauma and Decomposition

The presence or absence of perimortem trauma has been argued to play a significant role in the rate of decomposition (Mann *et al.* 1990; Micozzi 1986; Micozzi 1991). As with other factors that potentially effect decomposition, this too has been reexamined in the post-*Daubert* era, but unlike most of the other factors, studies on the influence of trauma have come under stricter scrutiny (Cross and Simmons 2010; Kelly 2006).

One of the first articles that mention the effects of trauma on the rate of decomposition is by Micozzi (1986) and describes the effects of freezing, thawing, and mechanical injury on decomposition. This study concludes that the presence of trauma may alter the rate of decomposition. However, the only mention of trauma in the study is cervical dislocation in Wister rats; with no external trauma produced (Micozzi 1986). In addition, when further examination of the study is conducted, one will note that the subjects in this case were previously frozen (Micozzi 1986). In Micozzi (1997), the author articulates the effects on freezing on decomposition, namely that it will suspend the autolysis process until the remains are thawed, at which time the process will accelerate (Micozzi 1986; Micozzi 1991; Micozzi 1997). With this in mind, the results from the Micozzi (1986) study that trauma is influential in decomposition, have to be considered with caution, given that it would be nearly impossible to distinguish what effects were from the trauma and what effects were from the freezing.

The second major study in the area of trauma and decomposition is from Mann *et al.* (1990). During this observational study, the authors placed two human subjects of similar weight at the ARF at the same date and time. The only difference between the two subjects is that one has a penetrative perimortem gunshot wound in the thorax whereas the second had no discernible perimortem trauma (Mann *et al.* 1990). The result of this observational study was that the subject with a wound decomposed at a faster rate than the subject without. There was no other qualitative or quantitative data described in this study (Mann *et al.* 1990).

Following the publication of this study, the findings that decompositional rates are affected by the presence of perimortem trauma appear to have been widely accepted (Bass 1997; Byers 2011; Campobasso *et al.* 2001; Clark *et al.* 1997; Sledzik 1998; Vass *et al.* 1992). With articles such as Vass *et al.* (1992), Shean *et al.* (1993), and texts like *Postmortem Change in Human and Animal Remains: A Systematic Approach* by Micozzi (1991), one can see how the field accepted the findings upon the release of both Micozzi (1986) and Mann et al. (1990).

Recently, however, some challenges to the premise that trauma influences the rate of decomposition have arisen. In 2005, Breitmeier, *et al* published a study on the correlation between time that human remains spent in the ground and the findings at exhumation. The authors expected to find that those subjects buried with perimortem trauma would have attained a greater state of decomposition than those subjects without perimortem trauma. However, at exhumation, the authors noted that the presence of

trauma had no significant effect on the rate of decomposition (Breitmeier *et al.* 2005). This, however, could be due to the fact that the remains were in a subterranean deposition with an overall decelerated rate of decomposition.

J. A. Kelly (2006) , in Bloemfontein, Free State, South Africa, conducted a study examining the effects of both clothing and trauma on decomposition from an entomological point of view. Kelly's traumatic trials included 14 porcine carcasses that were euthanized using Pentobarbitone sodium 200 mg/ml. It should be noted, that little study has been done on the effects of chemically induced euthanasia on the rate of decomposition. Those carcasses were then inflicted with various dimensions of knife wounds, with several having their throats deeply lacerated and others having more superficial wounds to the fore and hind limbs, and deposed on the surface in a mostly sun environment (Kelly 2006). The author utilized both observational methodologies and statistical analysis of measures of weight loss, temperature fluctuations, and a form of the TBS, combining aspects of Megyesi *et al.* (2005) and Anderson and VanLaerhoven (1996), the latter of which is a decompositional scale based on an entomological context (Kelly 2006). The author conducted two trials in this study: one during summer months and one during autumn months, the results of which demonstrated that the presence of trauma did not influence the rate of decomposition either observationally or statistically (Kelly 2006: 122).

In 2007 a study conducted at the University of Central Lancashire in the United Kingdom by Cross and Simmons (2010) specifically examined the influence that

penetrative trauma had on the rate of decomposition as it pertains to the field of forensic anthropology and animal decomposition, rather than as an entomological study focusing on insect succession (Cross and Simmons 2010). In their study, the authors used several quantitative measurements, similar to those of Kelly (2006), to determine and compare rates of decomposition between sets of porcine remains, with and without inflicted additional trauma (Cross and Simmons 2010). In particular the authors examined temperature fluctuations, weight loss, and TBS, and based the progressions on ADD specifically devised by Megyesi *et al.* (2005) (Cross and Simmons 2010). The authors utilized 34 sets of remains (*S. scrofa*), euthanized by the captive bolt method with the wounds of the control group closed using pithing cane and plasticine (Cross and Simmons 2010). The subjects were divided into three groups: a trauma-disturbed group consisting of three remains, a non-trauma-disturbed group consisting of three remains, and an undisturbed group consisting of twenty-eight remains. With the undisturbed group, no distinction was made as to how they were divided (traumatic and non-traumatic) (Cross and Simmons 2010). With the traumatic group, 9mm gunshot wounds were inflicted approximately 4 to 6 hours postmortem. The subjects were then placed approximately 50cm apart, in a surface depositional environment that received an equal amount of sun and shade with wire mesh tacked down around each subject to discourage scavenging (Cross and Simmons 2010). Like the Kelly (2006) study, the results of Cross and Simmons (2010) found that there was no significant difference in the rate of decomposition between subjects with trauma and those subjects without trauma.

Several problems exist with this study, particularly in the lack of clarity in how traumatic and non-traumatic groups were divided, and the timing and placement of the wounds. By separating the remains into disturbed and undisturbed sub-groups, the authors attempted to control the effects of investigator disturbance on the maggot and decompositional activity. This was done in conjunction with a concurrent study that was being conducted at the location that investigated effects of investigator disturbance on decomposition (Adlam and Simmons 2007; Cross and Simmons 2010). However, while the separation of disturbed and undisturbed is an important factor to evaluate, the authors did not mention how many of the undisturbed groups were traumatic and how many were non-traumatic (Cross and Simmons 2010).

A second major issue with the study is in the area of the timing and placement of the trauma. The authors of the study state that wounds were placed in the limbs and chest, 4 to 6 hours postmortem, which exceeds the standards for the onset of livor mortis by at least 2 hours (Burton 1974; Clark *et al.* 1997; Cross and Simmons 2010). To correct for this, the authors replaced the lost blood with refrigerated porcine blood (Cross and Simmons 2010). The naturally occurring blood, as well as radiating heat from fresh remains, would have long been lost. Both of which are potential factors in early onset oviposition by insects (Dadour 2011). An issue as to the methodology in how trauma was inflicted can also be brought into question. The authors used a 9mm handgun, but do not state the distance between the muzzle and the remains, which can influence the amount of actual trauma that was inflicted (Cross and Simmons 2010; MacAulay *et al.* 2009). While an interesting study and the most comprehensive study of the impact of

trauma on decomposition to date, there remains a critical need for additional studies in this area. Accordingly, the present study was constructed to investigate the effects of trauma on decomposition, using a porcine model that more closely replicates a real case scenario with remains that have experienced perimortem trauma. It is hypothesized that using remains with incised injuries within one hour of the time of death, placed in a controlled environment and the acquisition of both qualitative and quantitative data will provide greater insight into the degree to which trauma alters that rate of decomposition.

CHAPTER 3: MATERIALS AND METHODS

The present study, conducted at the Boston University Research Facility in Holliston, Massachusetts (MA) from June 13 to July 12, 2011 and August 3 to August 15, 2011, examined whether a differential rate and pattern of decomposition could be ascertained between subject groups presenting with penetrative trauma, non-penetrative trauma, and a non-trauma control group. The project was terminated when all subjects reached a stage of complete skeletalization or mummification.

Sample

Due to the inability to use human remains for decompositional projects at the current time, porcine models were utilized as an alternative. It has been widely accepted by the field that porcine remains can be a viable substitute for human remains in decompositional studies (Catts and Goff, 1992; Cross and Simmons, 2010; Mann *et al.*, 1990). In this study, eight domestic bred porcine carcasses (*S. scrofa*) were used. Seven animals, six experimental subjects and one control, were placed in wire cages at the outdoor facility from June 13 to July 12, 2011, and a second verification control was placed from August 3 to August 15, 2011. The animals were obtained from the Tufts University Cummins School of Veterinary Medicine (Grafton, MA), and were euthanized by a captive bolt with the time of death recorded. Captive bolt euthanasia is an accepted method for euthanasia of swine by the American Veterinary Medical Association and was approved by the Institutional Animal Care and Use Committee (IACUC) at Tufts University School of Veterinary Medicine. The subjects were divided into three groups

depending on the type of postmortem trauma inflicted. One group of three received sharp force penetrative trauma inflicted in the thoracic region of the remains, consisting of a single incision, approximately 15cm in length, penetrating into the cavity (see Figure 1 and Figure 2). The second group of three received a single incision, approximately 15cm in length, incising both the dermis and part of the musculature anterior to the costal cage, but not penetrating into the thoracic cavity itself (see Figure 3 and Figure 4). The incisions on both the penetrative and non-penetrative trauma groups were uniform in both size and location and were inflicted using a generic #20 scalpel blade on a steel #4 scalpel handle. The only variation between the groups was the depth of the incisions. The trauma was inflicted on the side of the animal where lividity was present approximately 30 minutes after euthanasia in an effort to simulate natural bleeding from the wound, which was achieved. The remaining two subjects were the control group of the project and had no trauma inflicted. One was placed with the experimental groups while the second was placed 3 weeks following the termination of the first trial. Because the first control reached a terminal decomposition point at a faster rate, the second control, also known as the verification control or verification trial, was used to verify the results of the first.

Due to the manner of euthanasia, the wounds created in the skull by the captive bolt were closed and sealed using Krazy® (n-Butyl cyanoacrylate) adhesive, and PlastiDip International® liquid tape spray in order to prevent insect activity at this site. The adhesives did not repel insect activity, but did prevent oviposition at the wound site.

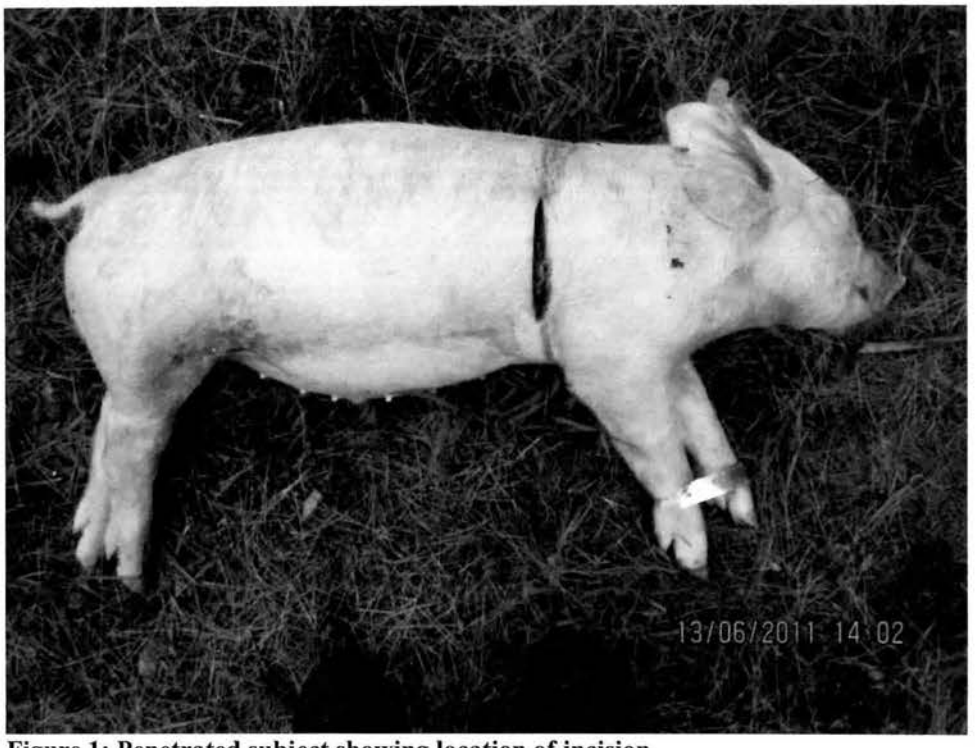


Figure 1: Penetrated subject showing location of incision

Figure 2: Penetrative incision

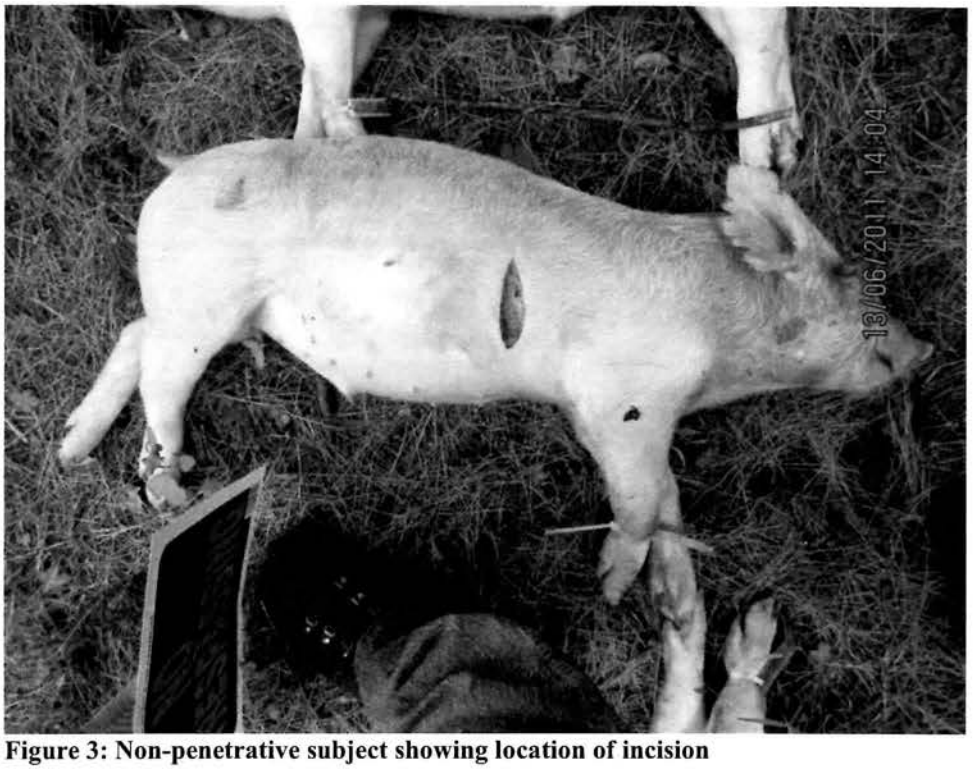


Figure 3: Non-penetrative subject showing location of incision

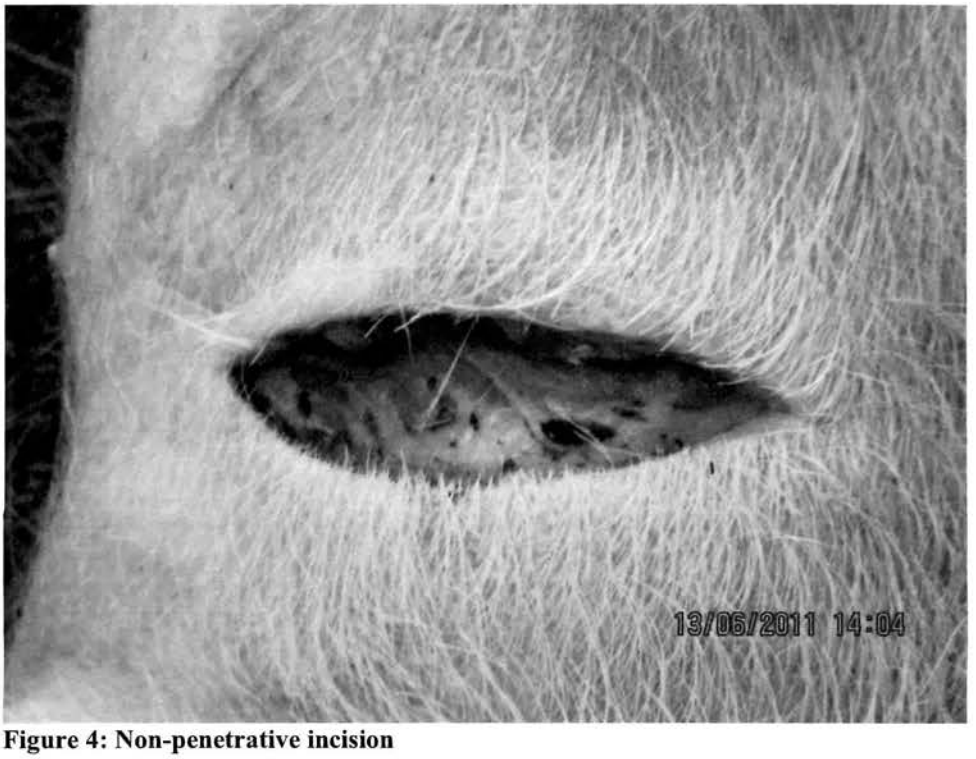


Figure 4: Non-penetrative incision

In a study by Adlam and Simmons (2007), it was found that repeated physical disturbances could potentially alter the decomposition process. Following the methodological protocol of Adlam and Simmons (2007), this study further divided each of the three groups into two sub-groups: a disturbed group which consisted of one subject, and an undisturbed group which consisted of the remaining two carcasses. For the control subjects, the subject placed during the experimental trial was considered a disturbed subject while the verification control was undisturbed. None of the animals had exsanguinated during the euthanasia process. Each of the disturbed subjects were weighed on day 1 immediately following euthanasia, along with their containment units, an iCrate® model number 1594, using a Feedback™ Expedition® model hanging scale with a load capacity of 49.895kg. The crates were weighed at 4.56kg and the subjects at 16.40 kg (control), 18.15 kg (penetrated), and 19.75 kg (non-penetrated).

Holliston Research Facility

The subjects were placed at the Boston University Research Facility in Holliston, Massachusetts, a 32 acre outdoor research facility approximately 38.62km from the center of Boston[1]. The facility is a predominantly wooded environment with swamp lands and a large empty field (see Appendix A). The city of Holliston averages annual temperatures ranging from approximately -10.55°C (low) in mid-late January to 29.44°C (high) in mid-July. The annual highs range from 2.77°C in mid-January to 29.44°C in mid-July with the average lows ranging from -10.55°C to 15.55°C. The average intra-

[1] ≈32km from Boston University School of Medicine.

day temperature swing throughout the year is approximately 13.26°C, with an average temperature swing during the experimental phase of 13.05°C (see Table 3) (city-data.com, 2011). The average humidity ranges from 54% to 82% annually with the morning ranging from 70% to 82% and the afternoon ranging from 24% to 61%. During the experimental phase the average humidity ranged from 56% to 80%: 75% to 80% in the morning and 56% to 58% in the afternoon (see Table 4) (city-data.com, 2011).

Table 3: ***Annual Average Temperature, Holliston, MA (calculated in °F) (copied from city-data.com, January, 2012)***

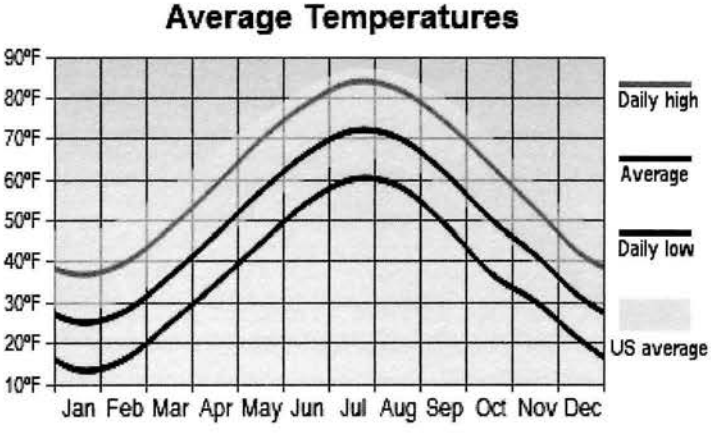
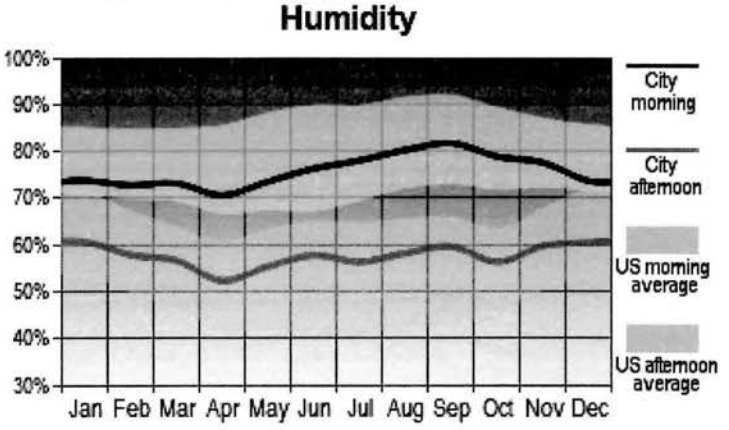


Table 4: ***Annual Average Humidity, Holliston, MA (copied from city-data.com, January, 2012)***



The city receives monthly rain totals ranging from 88.9mm in February to 119.38mm in November with winter snowfalls ranging from 50.8mm in November to a peak of 398.78mm in late-January and descending back down to an average of 12.7mm in early-May. Both the rainfall and snowfall are above the national average (city-data.com, 2011). In addition, the city receives below the national average of daily sunshine ranging from 48% in early-January, peaking at 60% in July and August with the lowest amount of daily sunshine being late-November at 46% (city-data.com, 2011).

During the experimental and verification stages of the experiment the ambient temperature at the research site ranged from 9.22°C to 32°C with an average daily temperature of 20.7°C and an intra-day swing of 10.42°C. During the experimental stage the average daily temperature was 20.47°C with a range from 9.22°C to 32°C and an intra-day temperature swing of 11.11°. For the verification control, the average daily temperature was 21.23°C with a range from 12.61°C to 28.23°C and an average intra-day temperature swing of 8.82°C.

The daily humidity during the experimental phase ranged from 73.08% to 99.71%, and during the verification phase from 80.6% to 98.75%. The average humidity for the experimental phase was 84.95% and during the verification phase was 80.60%; the overall average was 85.82%. The average humidity during the experimental phase at ADD 276 (the ADD at which the verification control reached a TBS of 35) was 87.40%. All of the weather data can be found in Appendix B and was collected using a Davis® Vantage Pro2™ Weather Data Station.

Deposition

The subjects were placed in wire cages on a flat, elevated field with limited shade, with a 4.6m distance between each subject. Due to the potential that the amount of sun can affect the overall rate and process of decomposition, care was taken to ensure that all depositional groups received an equal daily amount of sun and shade, by placing the remains more towards the center of the field rather than around the tree line (Shean *et al.*, 1993). The location of the cages was mapped using a Leica TCR803power total station with a datum point located at N 42.20709/W 071.41846, with an altitude of 76.2m above sea-level and an error of ±3.96m. Those points were then plotted on a combination plot and relief map using the Surfer® program (version 10.4, Golden Software, Inc. Golden, CO) (See Appendix C). The coordinates of the datum were taken using a Garmin™ Rino® 530HCx 2 way radio and global positioning system (GPS), utilizing 7 satellites.

Codification and Identification

For this project, an identification system was created with each subject receiving an individual three to four character alpha-numeric code. The first letter of the code was used to identify the subject-group (C-control, N-non-penetrative, or P-penetrative) and the second letter was used to identify the sub-group type (D-disturbed, or U-undisturbed). The last character of the code identified the various depositional groups (1, 2 or 3). The subjects in the disturbed sub-group were given the depositional group identifier of 1.

Table 5: ***Codification Chart***

Code	Definition	Code	Definition
CD	Control Disturbed	CU	Control Undisturbed
ND	Non-penetrated Disturbed	NU	Non-Penetrated Undisturbed
PD	Penetrated Disturbed	PU	Penetrated Undisturbed

Measurements

Accumulated Degree Days

All days during the experiment were recorded as ADD, in degrees Celsius, and Accumulated Humidity Day (AHD) and were determined using an on-site Davis® Vantage Pro2™ weather station. The minimum and maximum temperatures, as well as humidity, were recorded for 24-hour periods at 0000 daily from June 13 until July 12 and August 3 until August 15 of 2011. This data was then cross-checked with the National Oceanic and Atmospheric Administration (NOAA) data daily for accuracy. To calculate the accumulated degree day, the minimum and maximum temperature for each day was averaged together and added to an accumulated total of the previous averages that began

with the temperature average on June 13 and August 15 respectively (Megyesi *et al.*, 2005). The weather data was measured from the weather station on the property in degrees Fahrenheit, but was then converted into degrees Celsius using Microsoft® Excel (version 14.0, Microsoft® Corporation, Redmond, WA). A similar procedure was conducted to calculate the AHD with the average daily humidity being substituted for the average temperature. Since the second control was placed a month after the first set of experimental and control animals, the weather data for each set of animals were recorded and then compared to each other to determine if there were differences in weather data between the two time frames. For example, if the verification control achieved skeletalization at an ADD of 276 and an average humidity of 86%, the state of the verification control was then compared to the state of the first control at the same ADD and average humidity. In this experiment, both the experimental and the verification control achieved skeletalization at the same ADD and humidity, but the verification control achieved complete skeletalization 2 temporal days sooner.

Weight and Body Temperature

Weight and body temperature measurements were taken from the disturbed sub-group only. The weight of the disturbed subjects was measured daily using a Feedback™ Expedition® model hanging scale with a load capacity of 49.895kg purchased from Old Will Knot Scales™. The scale was attached to a collapsible tripod made from 2"x4" lumber. A cable apparatus was constructed to lift the cages to the scale, with the bottom of the cages achieving a total clearance of 12cm above the ground. This apparatus allowed the weight reading to be taken rapidly and with as little disturbance to the

subjects as possible. Prior to the beginning of the project, the containment units were also weighed. This measurement was then subtracted from the measurements taken from the subjects to determine their tare weights and ensure the most accurate weight possible.

For body temperature measurements, an Onset® HOBO™ Pendent Data Logger (Onset Computer Corporation, Inc., Bourne, MA) was utilized in the anal cavities of all of the subjects in the disturbed sub-group. In addition a 4th pendent was placed in the wound cavity of the Subject PD1 to determine the presence of a potential temperature fluctuation at the wound site itself and if this varied from the other abdominal temperature measurements. The thermometers were placed immediately following euthanasia and were retrieved after all subjects reached the skeletalization stage of decomposition. However, no actual data was collected due to the fact that the data loggers sustained a 50% failure rate. The temperature data for the ND1 and the wound temperature for PD1 could not be retrieved.

Total Body Score

The primary investigative variable in this project was the measurement of the TBS, a measurement scale established by Megyesi *et al.*(2005). The TBS scoring system is a variation of the decompositional scoring system devised by Galloway (1997) in that it converts a qualitative measuring system into a quantitative score. Each of the four phases of decomposition described in Galloway (1997) (fresh, early, advanced, and skeletalization) were broken down into specific sub-phases that were each assigned a

specific score. The system was applied to one of three body regions (head and neck, trunk, and limbs) (see Table 6, Table 7, and

Table 8). Once the different regional scores were obtained, they were added together to establish the total body score. The TBS score ranges from a minimum of 3 to a maximum of 35. This project examined and recorded the TBS for all subjects in all depositional groups and began on June 13th, the day the project began. In addition, this project considered that total skeletalization occurred when a subject reached a TBS of 35, or when the remains reached terminal mummification. All subjects achieved this TBS score by July 12th, 2011. A similar process was conducted for the verification control which started on August 3rd and continued until August 15th. Primary TBS measurements were taken by the experimenter, and cross-checked by qualified researchers to determine the presence of potential inter-observer error. In addition, photographs were taken of each subject daily and the TBS re-examined at the conclusion of the project in an effort to establish whether inter-observer and intra-observer error may have occurred in the measurements.

Table 6: ***Categories and stages of decomposition of the head and neck (copied from Megyesi* et al *2005)***

A. Fresh	
1pt	1. Fresh, no discoloration
B. Early decomposition	
2pts	1. Pink-white appearance with skin slippage and some hair loss.
3pts	2. Gray to green discoloration: some flesh still relatively fresh.

4pts	3. Discoloration and/or brownish shades particularly at edges, drying of nose, ears and lips.
5pts	4. Purging of decompositional fluids out of eyes, ears, nose, mouth, some bloating of neck and face may be present.
6pts	5. Brown to black discoloration of flesh.

C. Advanced decomposition

7pts	1. Caving in of the flesh and tissues of eyes and throat.
8pts	2. Moist decomposition with bone exposure less than one half that of the area being scored.
9pts	3. Mummification with bone exposure less than one half that of the area being scored.

D. Skeletalization

10pts	1. Bone exposure of more than half of the area being scored with greasy substances and decomposed tissue.
11pts	2. Bone exposure of more than half the area being scored with desiccated or mummified tissue.
12pts	3. Bones largely dry, but retaining some grease.
13pts	4. Dry bone.

Table 7: ***Categories and stages of decomposition of the trunk (copied from Megyesi et al 2005)***

A. Fresh

1pt	1. Fresh, no discoloration.

B. Early decomposition

2pts	1. Pink-white appearance with skin slippage and marbling present.
3pts	2. Gray to green discoloration: some flesh relatively fresh.
4pts	3. Bloating with green discoloration and purging of decompositional fluids.
5pts	4. Post bloating following release of the abdominal gases, with discoloration changing from green to black.

C. Advanced decomposition

6pts	1. Decomposition of tissue producing sagging of flesh; caving in of the abdominal cavity.

7pts	2. Moist decomposition with bone exposure less than one half that of the area being scored.
8pts	3. Mummification with bone exposure of less than one half that of the area being scored.

D. Skeletalization

9pts	1. Bones with decomposed tissue, sometimes with body fluids and grease still present.
10pts	2. Bones with desiccated or mummified tissue covering less than one half of the area being scored.
11pts	3. Bones largely dry, but retaining some grease.
12pts	4. Dry bone.

Table 8: ***Categories and stages of decomposition of the limbs (copied from Megyesi et al 2005)***

A. Fresh

1pt	1. Fresh, no discoloration

B. Early decomposition

2pts	1. Pink-white appearance with skin slippage of hands and/or feet.
3pts	2. Gray to green discoloration; marbling; some flesh still relatively fresh.
4pts	3. Discoloration and/or brownish shades particularly at edges, drying of fingers, toes, and other projecting extremities.
5pts	4. Brown to black discoloration, skin having a leathery appearance.

C. Advanced decomposition

6pts	1. Moist decomposition with bone exposure less than one half that of the area being scored.
7pts	2. Mummification with bone exposure of less than one half that of the area being scored.

D. Skeletalization

8pts	1. Bone exposure over one half the area being scored, some decomposed tissue and body fluids remaining.
9pts	2. Bones largely dry, but retaining some grease.

10pts 3. Dry bone.

Insects

The presence and locations of insect and maggot masses on the carcasses was documented and used to determine if there were any differences between subjects.

Data Analysis

Qualitative Data

The timing and location of insects, clutches, and maggot masses on the remains were documented and the results were used to determine if the insects colonized the various groups in different patterns. In addition, the description of visual decomposition was also recorded and compared across subject groups.

Quantitative Data

An analysis, was conducted to compare terminal decomposition rates and daily TBS on an ADD/AHD basis across the groups using a two-way repeated measures analysis of variance (ANOVA) test using SPSS® (version 20.0; Integrated Business Machinery®, Chicago, Ill) with ADD set as the "within-subjects factor," and the three groups set as the "between-subjects" factors to determine if a differential between the various groups on a specific ADD was present.

Weight-loss data was also compared using a repeated measures ANOVA test using SPSS® v.20.0 with the recorded weights set as the "within-subject factor" and the

subjects set as the "between-subjects factor" to determine a differential in the weight-loss means.

Body temperature measurements were not taken due to data loss in 50% of the data loggers.

CHAPTER 4: RESULTS

Rate Differential Results

For the qualitative rate analysis, data was examined using the TBS on temporal, ADD, and accumulated humidity day (AHD) scales. The results of this analysis revealed limited difference in the rate of decomposition between subjects with trauma and the non-trauma controls. Statistical analysis was conducted comparing TBS across the ADD/AHD days using a repeated measures ANOVA test. The results of this test found that there was no significant difference in the rates of decomposition between the groups.

Temporal Differential

All subjects were graded as either a TBS of 35 (complete skeletalization) or terminal mummification within a temporal time span of 7 days. It should be noted that PD1, PU3, and NU2 reached a point of terminal mummification rather than complete skeletalization. The mean temporal day for this point (hereafter referred to as the terminal point) was 16 temporal days, with a standard deviation of 2.51 days. From this, three subjects reached a terminal point within 24hrs of the mean, five within 48hrs, and seven of the eight subjects reached their terminal point within 72hrs of the mean. In this particular case, subject PD1 reached a terminal point 4 days after the mean, and 7 days from the terminal point of the CU2 (verification control), which was the subject to reach its terminal point the quickest (13 days due to its accelerated terminal point, every subject's terminal point was compared to that of CU2). Using that as a reference, two

subjects (PU2 and PU3) reached a terminal point within 24hrs of CU2, and a third within 48hrs (NU3), and a fourth within 72hrs (CD1).

ADD Differential

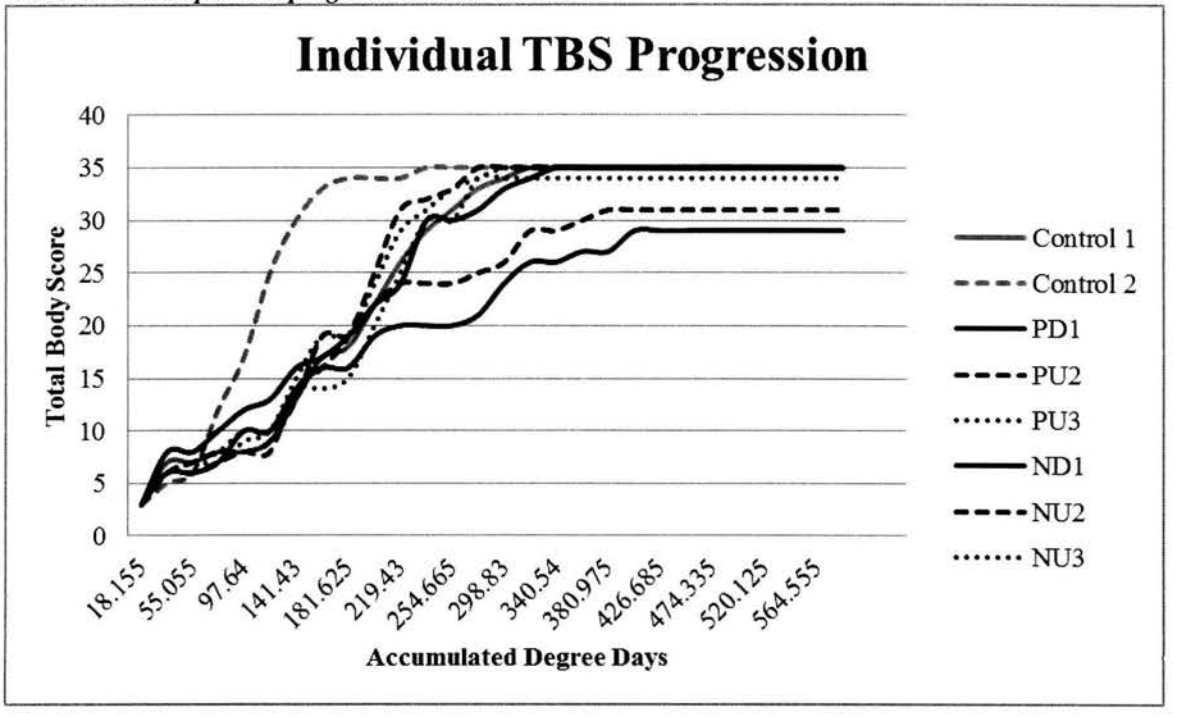
Due to variable weather data between the experimental trial and the verification control, a second set of comparisons was conducted using ADD. Within the subjects, there was a range from 254.31ADD (PU2 and PU3) to 380.92ADD (PD1). The mean terminal ADD point was 302.615 with a standard deviation of 47.54. The results of this study found that four subjects reached a terminal point within a range of 255.072 to 350.158 (the mean +/- the standard deviation), with an additional two subjects (PU2 and PU3) reaching a terminal point 0.76ADD below that range. The subjects attaining a terminal point with the lowest ADD were PU2 and PU3 at 254.31ADD, the latter of which reached a terminal mummification point rather than complete skeletalization. CU2 reached a terminal point at 276ADD and using it as a base, four subjects reached a terminal point within 24hrs (or 23ADD[2]). Two of the subjects (PU2 and PU3) reached a terminal point 24hrs prior, or -21.69ADD. Subject NU3 reached its terminal point at a similar ADD to CU2 (-0.03). The fourth subject (CD1) attained a terminal point 24hrs after CU2 with an ADD difference of +22.33. A fifth subject (ND1) reached its terminal point 48hrs (or 46ADD) after CU2, with an ADD difference at +45.69.

[2] This number was arrived at by taking the subjects that reached a terminal point 1 temporal day both above and below the temporal day with an equivalent ADD of 276 and rounded up to the nearest whole number. In this case, the ADD for day 15 was 275.97 so the ADD for days 14 and 16 were averaged together and rounded up to the next whole number to get the temporal day differences.

Table 9: ***Time of terminal decomposition (temporal, ADD, and AHD)***

Subject	Temporal	ADD	AHD
CD1	15	275.97	1307.167
CU2	13	276.00	1142.008
PD1	20	380.92	1723.313
PU2	14	254.31	1223.625
PU3	14	254.31	1223.625
ND1	17	321.69	1477.646
NU2	19	359.39	1638.875
NU3	15	275.97	1307.2
Mean	*15.875*	*299.82*	*1380.432*
S.D.	*2.368412*	*45.3479*	*197.0293*

Table 10: ***Decomposition progression based on ADD***

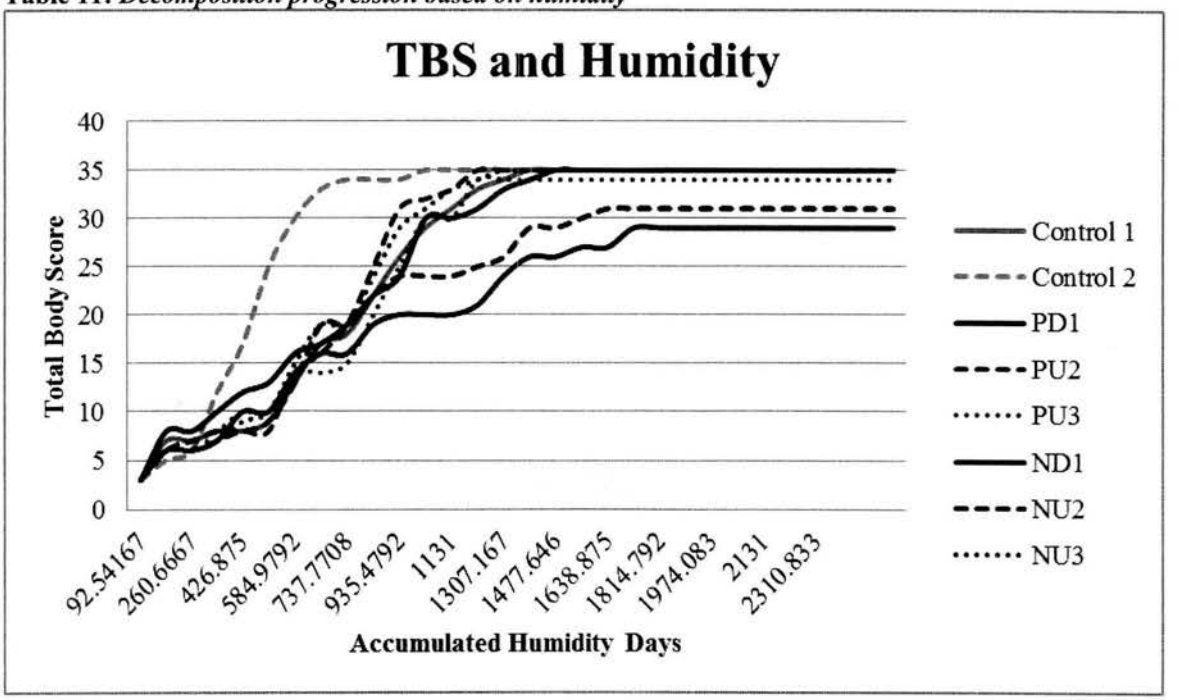


Humidity Differential

Humidity was an additional weather variable that was accounted for during this study. To achieve the accumulated humidity day (AHD), the total average humidity of a particular day (0000-2359) was added to a running total of the previous day's humidity, similar to that calculating the ADD. For this project, the terminal AHD ranged from 1142.008 (CU2) to 1723.313 (PD1) with a mean of 1391.143 and a standard deviation of 208.544. The results of this study found that five subjects (CD1, PU2, PU3, ND1, NU3) fell within a range of 1182.6 to 1599.687; the mean plus and minus the standard deviation. Additionally, two other subjects (CU2 and NU2) fell ±50HD from this range. Like ADD and temporal days, CU2 was used as a baseline with all subjects compared off of it. CU2 reached a terminal point at 1142.008AHD, with two subjects (PU2 and PU3) attaining a terminal point within 24hrs (93AHD[3]) of CU2 at +81.617AHD. NU3 attained a terminal point within 48hrs (+165.192AHD) and CD1 within 72hrs (+250.846AHD).

[3] This number was achieved using a similar method as the ADD. However, in this case, the terminal AHD did not have a corresponding AHD in the experimental trial. To arrive at this number, AHD of two consecutive days were found that the terminal AHD of CU2 would fall into; the difference of those two days was used. For this project CU2 had a terminal AHD of 1142.008; the AHD for temporal days 13 and 14 were 1131 and 1223.625 respectively. The difference of these two days was 92.625, rounded up to the nearest whole number gave a temporal equivalent of 93AHD.

Table 11: ***Decomposition progression based on humidity***



Quantitative Analysis

Statistical analysis was conducted utilizing the ADD data from all eight subjects using SPSS® v20.0 with significance levels set at *p-value* ≤ 0.05. A two-way repeated measures ANOVA with ADD (labeled as days) set as the "within-subjects factor," and the three groups (control, penetrative, and non-penetrative) set as the "between-subjects" factors. The ANOVA was used to determine if there was a statistically significant difference between the trauma groups and the non-trauma groups. The results of this analysis [$F(1,5) = 776.02$, *p-value* $= 0.361$], showing that there was no statistically significant difference between the subject groups.

Tests of Between-Subjects Effects

Measure:MEASURE_1
Transformed Variable:Average

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	140869.593	1	140869.593	776.024	.000
Group	456.774	2	228.387	1.258	.361
Error	907.637	5	181.527		

Figure 5: SPSS® v20.0 ANOVA results for the project

Additionally, statistical analysis was conducted utilizing the weight-loss data from CD1, PD1, and ND1 using SPSS® v20.0 with significance levels set at *p-value* ≤ 0.05. A two-way repeated measures ANOVA with weight (labeled as days) set as the "within-subject factor" and the subjects as the "between-subjects" factor. The ANOVA was used to determine if there was a statistically significant difference in the loss of weight between the three subjects. The results of this analysis [$F(1,1) = .56.593$, $p=0.382$] found that there was no significant difference in the loss of body weight between the subjects.

Tests of Between-Subjects Effects

Measure: Weightloss

Transformed Variable: Average

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	299.012	1	299.012	56.593	.084
Subject	11.300	1	11.300	2.139	.382
Error	5.284	1	5.284		

Figure 6: SPSS(R) v20.0 weight-loss results

Experimental Trial Results

Temporal Differential

All subjects in the experimental trial reached a terminal point within a range of 14 to 20 temporal days with five of seven subjects reaching their terminal point within 48hrs of each other. The mean temporal day was 16.43 days with a standard deviation of 2.37 days. The control of this subject reached its terminal point at 16 days, the closest to the mean. Two other subjects reached their terminal point within 24hrs of the mean and four others reaching it within 48hrs. Six of the seven subjects reached their terminal point within 72hrs of the mean, and five of the seven subjects reached their terminal point within the mean plus or minus the standard deviation. Two subjects, ND1 and NU3 reached their terminal point within 24hrs of the control (ND1 +1, NU3 -1) with two others (PU2 and PU3 within 48hrs [both at -48hrs]).

ADD Differential

Using the ADD, all subjects reached their terminal point within a range of 254.31 to 380.92ADD with a mean ADD of 306.42 and a standard deviation of 50.02. The control from this trial reached its terminal point the closest to the mean at 298.33 (-8.09) with two other subjects (ND1 and NU3) reaching their terminal point within 24hrs (23ADD) from the mean. Six of the seven subjects reached their terminal point within 72hrs (69ADD) of the mean. Three subjects (CD1, ND1, and NU3) reached their terminal points within a range of 256.40 to 356.44 (the mean plus or minus the standard

deviation) with PU2 and PU3 reaching their terminal points -2.09ADD and NU2 +2.95ADD of that range. Two subjects (ND1 and NU3) reached their terminal points within 24hrs of the control (+23.36 and -22.36 respectively) with an additional two subjects (PU2 and PU3) attaining it within 48hrs with a difference of -44.02ADD.

Humidity Differential

Examining for humidity, all subjected reached their terminal points in a range of 1223.625 to 1723.313AHD with a mean of 1426.734 and a standard deviation of 197.272. During the experimental trial, the control came the closest to the mean with an AHD of 1392.854 (-33.88). Two other subjects (ND1 and NU3) reached a terminal point within 24hrs (86AHD[4]) of the mean, with two others (PU2 and PU3) within 48hrs. Six of the seven subjects reached a terminal point within 72hrs (258AHD) of the mean, with only PD1 being the outlier. Three of the six subjects (CD1, ND1, and NU3) reached a terminal point within the range of 1229.462AHD to 1624.006AHD. PU2 and PU3 reached a terminal point -5.8369AHD below that range and NU2 reached a terminal point 14.869AHD above that range. Two subjects (ND1 and NU3) reached a terminal point within 24hrs of the control subject (+84.792 and -85.654 respectively) with two others (PU2 and PU3) within 48hrs; both of which reached a terminal point at an AHD difference of -169.229.

[4] The AHD for the subject closest to the mean was 1392.854 (the control); the difference in this AHD from the temporal day before was -85.654 and the one after was 84.792 yielding an average difference of 85.223 (or 86AHD).

Table 12: ***Time of terminal decomposition (Temporal, ADD, and AHD)***

Subject	Temporal	ADD	AHD
CD1	15	275.97	1307.167
PD1	20	380.92	1723.313
PU2	14	254.31	1223.625
PU3	14	254.31	1223.625
ND1	17	321.69	1477.646
NU2	19	359.39	1638.875
NU3	15	275.97	1307.2
Mean	*16.28571*	*303.223*	*1414.493*
S.D.	*2.249717*	*47.5139*	*187.3108*

Statistical Analysis

Similar statistical analysis was run using SPSS® version 20.0 on just the experimental group, with significance levels set at *p-value* ≤ 0.05. A two-way repeated measures analysis of variance (ANOVA) with ADD (labeled as days) set as the "within-subjects factor," and the three groups (control, penetrative, and non-penetrative) set as the "between-subjects" factors. The ANOVA test was used to determine if there was a statistically significant difference between the trauma groups and the non-trauma groups. The results of this analysis were [$F(1,2) = 533.29$, $p = 0.805$], demonstrating no statistically significant difference between the subject groups in the experimental trial. These results, taken together, suggest that the presence or absence of trauma does not have an effect on the rate of decomposition.

Tests of Between-Subjects Effects

Measure:MEASURE_1
Transformed Variable:Average

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	93751.488	1	93751.488	533.292	.000
Group	80.789	2	40.395	.230	.805

Figure 7: SPSS® v20.0 ANOVA results for the experimental trial

Group Pattern Results

After examining the qualitative data, a pattern of decomposition did emerge. Both of the control subjects, decomposition appeared to progress from the head and work inferiorly, down the body. CU2 followed this pattern until its termination point, and CD1 was following this pattern until extrusion took place. In the penetrative subjects, however, the initial decompositional changes and insect activity began around the wound and extrusion sites. This, however, was not the case for the non-penetrative subjects where the decompositional changes remained more at the head than the wound site, even though some changes and insect activity occurred at the wound site. In subjects with penetrative wounds, more blowfly activity was observed at the wound sites where in the non-penetrative subjects, limited insect activity was observed initially. All of the subjects' abdominal viscera extruded from the abdominal cavity within a 48 hour span of each other, after which a great deal of insect activity was seen at those sites.

For individual decomposition rates and patterns, please see Appendix D.

CHAPTER 5: DISCUSSION

Project Discussion

After analyzing both the quantitative and qualitative data generated by this project, the presence of trauma has been found to have no significant influence on the rate of decomposition in the current project. The subjects that incurred either a cavity-penetrating wound or deep laceration decomposed at the same rate as the observed in the non-trauma subjects. This result is in contrast to the Mann *et al* (1990) study, which declared that trauma was one of the more significant variables in the rate of decomposition. Instead, the current results were more consistent with the findings from both Kelly (2006) and Cross and Simmons (2010). While it has been widely accepted that trauma is a significant variable in the rate of decomposition, the findings from this study, combined with the results of Kelly (2006) and Cross and Simmons (2010), suggest that this premise may be inaccurate.

However, while it was shown in the present study that there is no difference in the overall rates of decomposition, it was demonstrated that there is a difference in the pattern of decomposition. During this project, the decompositional pattern of the 2nd control began in the facial region, and progressed caudally along the length of the body. This pattern was also observed in CD1 during the early phases of the project, but the pattern shifted upon the extrusion of the abdominal organs. With regards to the penetrated subjects, the decompositional pattern began at the wound site, as well as the extrusion sites, and moved outward, with small degree of activity occurring late in the

decomposition process. The decompositional pattern observed in the non-penetrated subjects, however, was a blend of the two patterns described above with decomposition occurring at both the wound site and the facial region, but not to the same degree as with either the penetrated or control subjects. This is not to say that the decomposition was retarded, but rather that it began on a wider scale on the remains of the non-penetrated subjects.

The prevailing reason for the pattern difference in the decompositional process is that of insect access to the remains. Diptera prefer moist cooler areas for oviposition, which is why they prefer to use the natural orifices of the face (Campobasso *et al.* 2001; Mann *et al.* 1990). By introducing trauma into the equation, the Diptera have an additional, larger orifice to inhabit; one that would allow for a greater number of insects to lay eggs in. The wound became the primary oviposition site, allowing for greater access to a moister environment which is more favorable for the larvae, as well as greater access to bacterial sites which the larvae feed off of. With the non-penetrating subjects, the wound site allowed for an additional favorable area for oviposition, but was not large enough to accommodate the entire clutch. This smaller area forced the Diptera to utilize the facial orifices for oviposition. In CD1, it wasn't until the exposure of the abdominal organs that the Diptera shifted from the face to the abdomen.

This explanation of the differential in pattern, however, does not explain why there was no difference in the actual rate of decomposition. Campobasso (2001) discussed how larvae on the remains were digesting the bacteria rather than the actual

flesh. The adding of trauma did have an impact on the Diptera and arthropod activity, but would not, and did not, have an impact of the intrinsic microbial activity found within the tissues. There was no change in the bacterial activity which is the primary agent in the rate of decomposition. The only factors which could potentially affect this agent would be extrinsic climactic factors such as temperature and humidity. A large maggot mass could alter the rate of intrinsic decomposition by altering the heat of a localized area, however if that mass is too large; as was seen with NU2, then the intrinsic decomposition process of the bacteria could ultimately cease due to excessive heat.

In summation, what was observed from this study was that the rate of decomposition is an intrinsic affair executed by microbial activity while the pattern of visible decomposition is an extrinsic affair executed by insects and other fauna. The presence of trauma has an impact of that visible decomposition but has no significant impact on the intrinsic decomposition.

Limitations

As with all scientific studies, there are limitations that must be addressed in future studies. Primarily, one must consider if the use of porcine models to replicate humans is as accurate as has been stated in the literature. A variable shared by Kelly (2006), and Cross and Simmons (2010), and this project, but not by Mann *et al* (1990), is the use of porcine models. Mann *et al* (1990) utilized human subjects, while the others utilized porcine. While in the literature it has been demonstrated that porcine remains have been used frequently as the closest model to human decomposition, little quantitative research

has been conducted to determine what, if any difference exists between the two. The only study found which has shown a similarity was an entomological study which compared insect succession rather than rate and pattern differentials. There are several differences anatomically between humans and *S. scrofa*. Included in these differences is the biochemical composition of the muscle fibers, particularly with regards to the ratio of actin to myosin. In addition, one has to consider the lipid concentration differential between the two species. No conclusive study has been conducted as yet to determine if these differences do, in fact, play a role in the rate of decomposition.

In addition, one must also consider the size of the subjects as well. The subjects in this project were relatively small juveniles, weighing approximately 18.05kg (±1.7kg), while the subjects in both Kelly (2006) and Cross and Simmons (2010) were larger (approximately 35kg), adult specimens. While it has been shown in the literature that subject size can affect overall decompositional rates, studies in the literature has explained that this is most likely due to the fact that smaller remains have less to decompose (Spicka *et al.* 2011; Simmons *et al.* 2010; Nagano and Suzuki 2007). Given the small size of the subjects in the current project, while it is possible to state that the presence of trauma is not a significant variable in decomposition, one cannot compare the rates to larger, human remains to develop a PMI estimator formula.

One must also consider the methods of euthanasia and its effect on decomposition. For this study, captive bolt was used, inflicting a universal trauma on all subjects which was closed using Krazy® (n-Butyl cyanoacrylate) adhesive, and PlastiDip

International® liquid tape spray. These adhesives did seal the wound, preventing oviposition, but did not act as an insect repellant, for Diptera was seen landing at the wound site. Likewise, for Cross and Simmons (2010), captive bolt was utilized, with pithing cane and plasticine used to seal the wound. However, in an ideal experiment, the non-traumatic control subjects would be just that—non-traumatic. Kelly (2006) utilized a different method, Pentobarbitone sodium 200 mg/ml, to euthanize the subjects, yet no definitive research has been conducted showing the effects of chemical euthanasia on decomposition. Mann *et al* (1990) used one subject that had expired because of the present trauma, and another (the control) which expired of other, non-traumatic, reasons that were not listed. There in that study, no additional trauma was introduced into the experiment which provided for a true control, nor was any chemical used which could potentially affect the decompositional process.

Further Research

This project confirmed known variables of decomposition as well as presented future lines of research. Two variables that this project confirmed as having major influences on decomposition were that of temperature and humidity. However, this project has tentatively shown that neither work in a vacuum, nor can one be considered a greater variable than the other. Instead, it has demonstrated that is it the ratio between temperature and humidity that is the overriding variable in the rate of decomposition. Temperature has, as discussed previously, long been considered a major factor in decomposition, while humidity has been considered a lesser variable (Catts 1992; Gill-King 1997; Gill 2005; Introna *et al.* 1991; MacAulay *et al.* 2009; Mann *et al.* 1990;

Micozzi 1986; Rodriguez and Bass 1983; Sharanowski *et al.* 2008). This project, however, has shown some evidence that humidity might play a larger role than originally thought.

When examining the decompositional rate differences between the experimental trial and the verification trial, the ADD was relatively the same. Indeed, two subjects in the experimental trial reached a terminal point faster than the verification control which decomposed at a faster temporal rate. The variable with the largest difference between the two groups was the humidity, and particularly the humidity-to-temperature ratio. The average daily temperature for the verification control subject was similar to that observed in the experimental trial, yet the average humidity was lower. Due to these results, further research and experimentation is needed to study the effects of both humidity, and the humidity-to-temperature ratio on decompositional rates.

Along the same lines as the humidity-to-temperature ratio, this project also demonstrated that further research is needed in examining the effect of intra-day temperature shifts. During the experimental trial, the average daily temperature shift was 11.03°C, with a shift of up to 17°C on certain days. Meanwhile, during the verification trial, the average shift was 8.82°C with maximum of 13.50°C. While little is published on the effects of intra-day temperature shifts on the rate or pattern of decomposition, the occurrence of a larger shift in the experimental trial could explain the occurrence of the early extrusion of all of the subjects; a phenomenon observed in every subject placed at the Holliston Research Facility in June, yet not seen in the subject placed in August.

CHAPTER 6: CONCLUSION

From June to August of 2012, eight porcine remains were used to test whether or not trauma was a significant variable in either the rate or pattern of decomposition in the New England area. In June of 2012, three subjects were lacerated with a 15cm wound which penetrated into the thoracic cavity while three others were lacerated with a similar wound, though not as deep. A seventh subject was used as a control and all were placed at the Holliston Research Facility in Holliston, MA to decompose until all reached either skeletalization or terminal mummification. As a verification of the one control, an eighth subject was placed at the same facility in August of 2012. The TBS from each subject was measured daily and compared on a temporal, ADD, and AHD basis.

The results of this study found that trauma was not a significant variable in the rate of decomposition, contradicting the Mann *et al* (1990) study, a paper which has been heavily cited in the literature as well as in the field, and confirming the research conducted by Kelly (2006) and Cross and Simmons (2010). The results of this project also found that trauma did play a significant role in the pattern of decomposition with the traumatic subjects decomposing from the wounds-outward, while the control subjects generally decomposed from the facial region-caudally.

Further research is needed, however, to examine the relationship between decomposition and trauma. This project has shown that temperature, humidity, and the ratio between the two can greatly affect the rate of decomposition suggesting that more

regional research is needed. Furthermore, this study utilized porcine models, and while the literature suggests that porcine can replicate human decomposition, confirmation with human remains should be conducted. Even using porcine models, other research should be conducted with regards to the season of the year. This study was conducted in the summer months; addition studies should be conducted to determine if there is a rate differential between trauma and non-trauma subject in the Spring, Autumn, or Winter.

Decomposition is a highly variable formula, and while this project has challenged the assertion that trauma is a significant variable, more research is needed to study the relationship of trauma to decomposition, as well as all other variables that affect the decompositional rate. Understanding the factors that affect decomposition will allow for a more accurate understanding of the PMI, shortening the range, reducing the potential pool of decedents, and assisting law enforcement officials investigate potential homicides and lines of inquiry.

APPENDICIES

APPENDIX A: HOLLISTON, MA RESEARCH FACILITY

Figure 8: Holliston, MA (copied from Google Maps)

Figure 9: Holliston Research Facility (copied from Google Maps)

Figure 10: Decomposition field (copied from Google Maps)

APPENDIX B: PROJECT WEATHER DATA

Experimental Trial Data

Table 13: ***Experimental Trial Weather Data***

Date	Min	Max	Avg	ADD	Swing	AHD
6/13/2011	11.72	20.33	16.03	16.03	8.61	92.54167
6/14/2011	12.00	16.28	14.14	30.17	4.28	183
6/15/2011	10.22	24.11	17.17	47.33	13.89	260.6667
6/16/2011	10.83	29.83	20.33	67.67	19.00	335.5833
6/17/2011	14.44	21.56	18.00	85.67	7.11	426.875
6/18/2011	16.44	27.22	21.83	107.50	10.78	511.8958
6/19/2011	14.78	24.61	19.69	127.19	9.83	584.9792
6/20/2011	9.22	26.22	17.72	144.92	17.00	661.3333
6/21/2011	11.94	28.56	20.25	165.17	16.61	737.7708
6/22/2011	17.28	21.44	19.36	184.53	4.17	835.7708
6/23/2011	15.17	18.22	16.69	201.22	3.06	935.4792
6/24/2011	13.89	16.61	15.25	216.47	2.72	1035.104
6/25/2011	12.39	21.33	16.86	233.33	8.94	1131
6/26/2011	16.22	25.72	20.97	254.31	9.50	1223.625
6/27/2011	14.56	28.78	21.67	275.97	14.22	1307.167
6/28/2011	16.17	28.56	22.36	298.33	12.39	1392.854
6/29/2011	18.78	27.94	23.36	321.69	9.17	1477.646
6/30/2011	12.61	26.17	19.39	341.08	13.56	1556.813
7/1/2011	11.17	25.44	18.31	359.39	14.28	1638.875
7/2/2011	14.61	28.44	21.53	380.92	13.83	1723.313
7/3/2011	16.44	26.83	21.64	402.56	10.39	1814.792
7/4/2011	18.44	30.22	24.33	426.89	11.78	1898.271
7/5/2011	16.56	31.28	23.92	450.81	14.72	1974.083
7/6/2011	15.78	31.00	23.39	474.19	15.22	2051.604
7/7/2011	17.67	29.67	23.67	497.86	12.00	2131
7/8/2011	17.78	24.22	21.00	518.86	6.44	2273.13
7/9/2011	18.94	28.11	23.53	542.39	9.17	2310.833
7/10/2011	12.22	28.33	20.28	562.67	16.11	2386.625
Experimental Trial Daily Swing:					11.03	

Verification Trial Data

Table 14: ***Verification Trial Weather Data***

Date	Min	Max	Avg	ADD	Swing	AHD
8/3/2012	14.78	25.78	20.28	20.28	11.00	81.3617
8/4/2012	15.33	26.67	21.00	41.28	11.33	163.028
8/5/2012	17.28	25.72	21.50	62.78	8.44	243.112
8/6/2012	19.39	28.72	24.06	86.83	9.33	327.424
8/7/2012	20.17	25.39	22.78	109.61	5.22	424.383
8/8/2012	20.44	28.06	24.25	133.86	7.61	517.028
8/9/2012	18.06	25.56	21.81	155.67	7.50	609.653
8/10/2012	17.56	26.00	21.78	177.44	8.44	702.028
8/11/2012	15.33	25.94	20.64	198.08	10.61	782.966
8/12/2012	12.61	26.11	19.36	217.44	13.50	863.57
8/13/2012	13.83	26.56	20.19	237.64	12.72	947.487
8/14/2012	18.11	22.28	20.19	257.83	4.17	1043.26
8/15/2012	15.78	20.56	18.17	276.00	4.78	1142.01
				Verification Trial Swing:	8.82	

APPENDIX C: LOCATIONS OF SUBJECTS

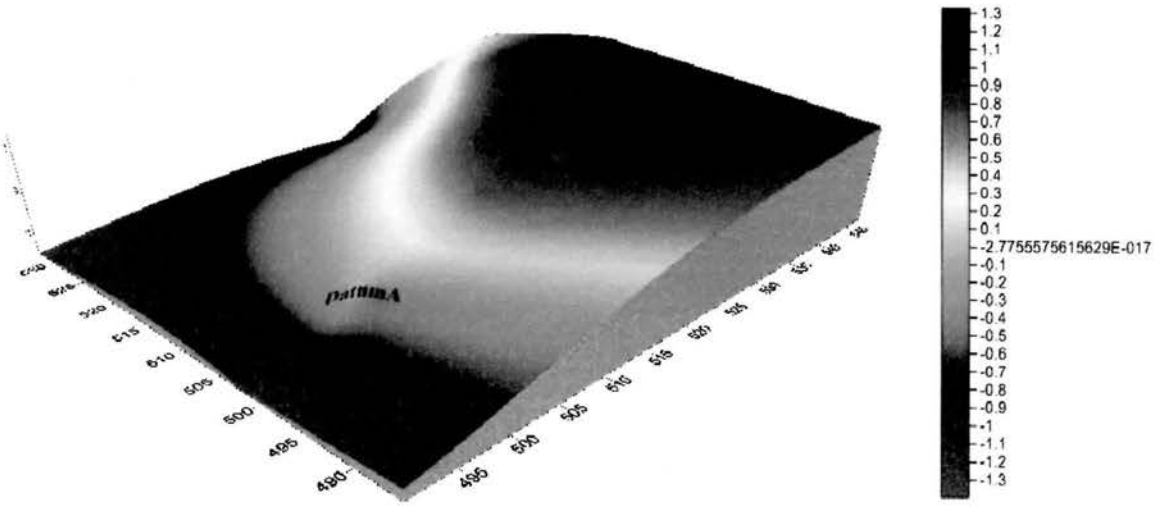


Figure 11: Map of subject layout using Surfer® v10.4

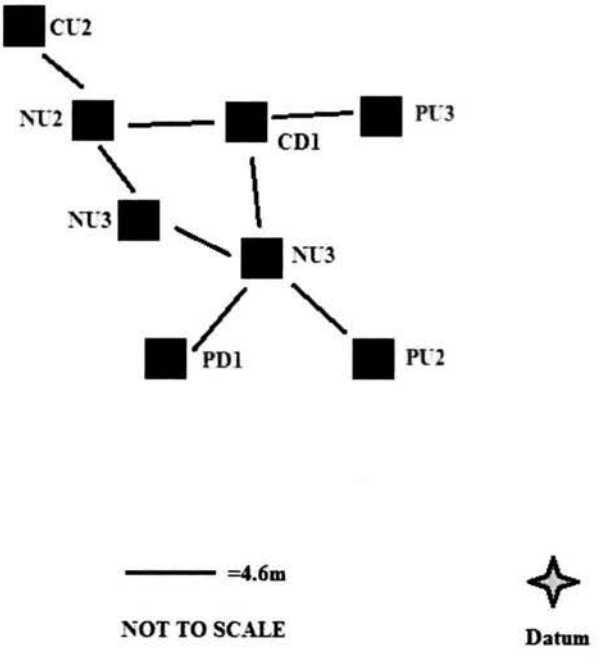


Figure 12: Map of subject location (drawn using Microsoft® Windows 7™ Paint (version 6.1, Microsoft® Corporation, Redmond, WA)

APPENDIX D: INDIVIDUAL DECOMPOSITION PATTERNS

CD1

Figure 13: CD1 Day 1 (16.03ADD: TBS 3)

The initial control was placed out at the Holliston research facility on June 13th, 2011 and was listed as TBS of 3 because all aspects of the body were fresh. By the second day, 24hrs later (30.17ADD), the subject reached a TBS of 7 with the head, body, and limbs pink to white in color with a slight amount of bloat in the abdomen. In addition to the bloat, a faint discoloration of a greenish hue was found on the distal aspect of the abdomen and the beginnings of an egg mass was found on the snout, mouth, and eyes. On the 17th (day 5: 85.67ADD) the green discoloration had extended to the entirety of the abdomen with the beginnings of extrusion taking place, just anterior to the left hind leg. The egg mass had also grown into maggot mass of 1st instar maggots with the

beginnings of bone exposure to the snout. By the following day (day 6: 107.5ADD) the extrusion process had accelerated with noticeable small intestines being exposed and more bone exposure had taken place in the facial region.

Figure 14: CD1 Day 4 (67.67ADD: TBS 8)

Figure 15: CD1 Day 7 (127.19ADD: TBS13)

On June 19th (day 7: 127.19ADD) the subject had reached a TBS of 13 with the head being scored as a 5 and the trunk and limbs scored as 4 each. The abdominal organs had completely extruded with the whole of the body completely distended. There was a reddish-brown patch of skin located on the neck with grey and black spots located on the head and limbs. The right forelimb was greyish black in coloration and the bones of the snout were completely exposed. Skin slippage was taking place on the right hind-limb and the medial aspects of the left forelimb, around the shoulder region. By the following day, the slippage had become much more noticeable, encompassing approximately 50% of their respective limbs. However, during this time, the maggots seem to have lost interest in the facial region and had migrated to the point of evisceration. The exposed organs, which appeared previously to be filled lumens have broken apart and lost their

shape. The skin around the folds, extrusion site, and snout appeared to be brown and leathery in appearance.

Figure 16: CD1 Day 8 (144.92ADD: TBS 17)

On June 21st (day 9: 165.17ADD) the subject reached a TBS of 18 with the head receiving a score of 6, the trunk a 7, and the limbs a 5. The skin of the head had turned grey to black with brown leathery spots and greater bone exposure on the snout had taken place. The skin appeared to be peeling away from the bone at this stage as well. All four limbs had aspects that were leathery in appearance with skin slippage present; the right forelimb was the only limb that was blacker in coloration than brown. The greatest change could be seen in the trunk with the internal abdomen fully exposed as well as aspects of the thorax. The proximal portions of most (3-13) of the left ribs are exposed displaying a loss of costal cartilage. The maggot mass at this time had stretched from the

snout to the rectum, but were mainly in the folds, exposed intestines, or within the cavities.

By June 23rd (day 11: 201.22ADD) the subject reached a TBS of 26. A large maggot mass of 3rd instar maggots covered the remains. Bone exposure throughout the remains was approximately 50%. The head was scored at 10 with bone exposure less than 25% and the remaining skin brown and leather-like in appearance. No real bone exposure could be seen on the limbs, save that the left femur had been removed from the limb and was still attached to the os coxa which was buried under the maggot mass. The lack of exposure, however, could have been due to the extensive maggot presence which covered the limbs; due to this a conservative score of 7 was given. The region with the greatest amount of bone exposure was the trunk (scored as a 9) with over 50% of the skeletal elements fully exposed; however, large amounts of skin remained attached.

On June 24th (day 12: 216:47ADD) bone was exposed on all but the hind limbs. The skull was completely exposed with no skin present, and little skin is present on the rest of the remains. The bones were still greasy in appearance with segments of skin present underneath. By June 28th (day 16: 298.33ADD) all of the remaining soft tissue and been removed and all bones had dried out; the remains were scored with a TBS of 35.

Figure 17: CD1 Day 11 (201.22ADD: TBS 26)

Figure 18: CD1 Day 16 (298.33ADD: TBS 35)

CU2

Figure 19: CD2 Day 1 (20.28ADD: TBS 3)

The verification control (CU2) was placed at the Holliston facility on August 3rd, 2011 in the same field location and conditions as the initial control and was scored with a TBS of 3 given that all aspects of the remains were fresh. No real activity took place with the remains until August 5th (day 3: 62.78ADD) when the whole of the head was coated with an egg mass and 1st instar maggots. The rest of the remains of that day were still in a fresh condition, though the skin was of a white hue with splotches of pink, and scored with a TBS of 6. By the following day, the cranium was fully exposed and bloat was seen through the rest of the remains. Little maggot activity, however, was seen distally beyond the forelimbs. On August 7th (day 5: ADD 109.61) the proximal ⅓ of the subject had bone exposure with a large maggot mass encompassing that section. The

mass extended along the underbelly of the subject and appeared to be under the skin of the remaining torso. The skin of the trunk was green with a grey tint and the hind limbs were pink and still looked fresh. The forelimbs, however, were completely skeletonized. In order to assess the limb score, each individual limb was scored separately and with the average of those scores used to assess the TBS.

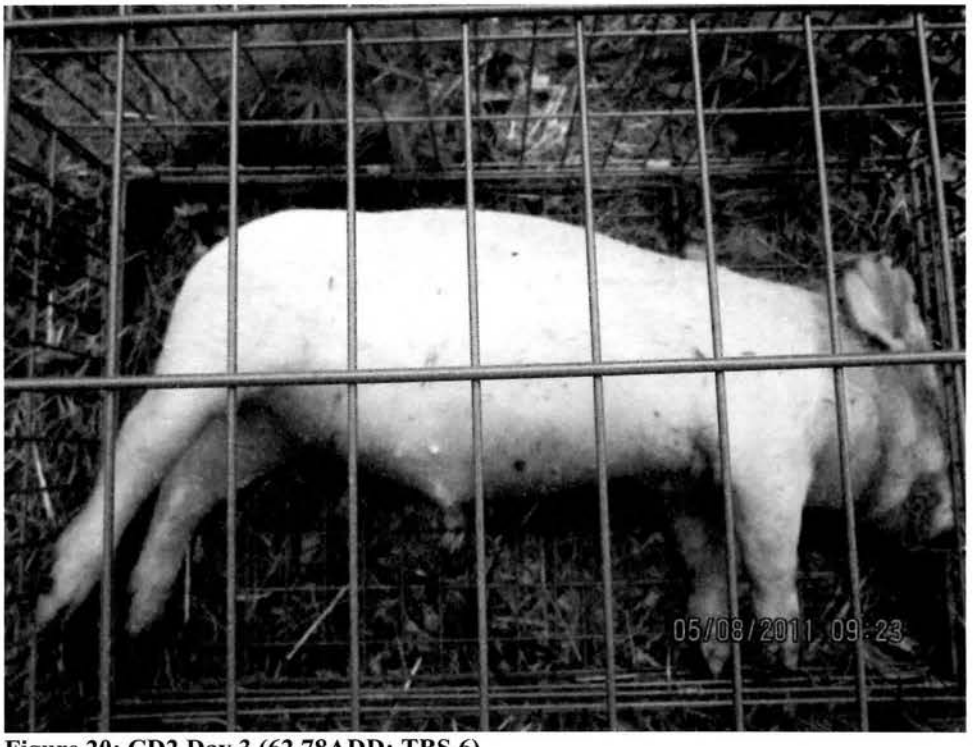


Figure 20: CD2 Day 3 (62.78ADD: TBS 6)

On August 8th (day 6: 133.86ADD), the superior ⅓ of the subject had been completely skeletonized with only a small patch of flesh remaining on the right mandible. The maggot mass had moved inferiorly encompassing the rest of the remains save the distal ⅓ of either hind limb. The skin of the abdomen appeared to be a yellowish-green with a grey tint, and the distal ⅓ of the hind limbs still appeared fresh. By the following day, the remains appeared to be completely skeletonized with greasy bone and some

remaining mummified skin, with the exception of the hind limbs which where encased in mummified brown leathery skin.

Figure 21: CD2 Day 5 (109.61ADD: TBS 17)

On August 9th (day 7: 155.67ADD) the subject was scored with a TBS of 30. The subject virtually remained in this state until August 15th (day 13: 276ADD) when the hind limbs achieved full skeletalization with dry bone. The skeletal elements of the rest of the remains dried out by August 11th (day 9: ADD 198.08).

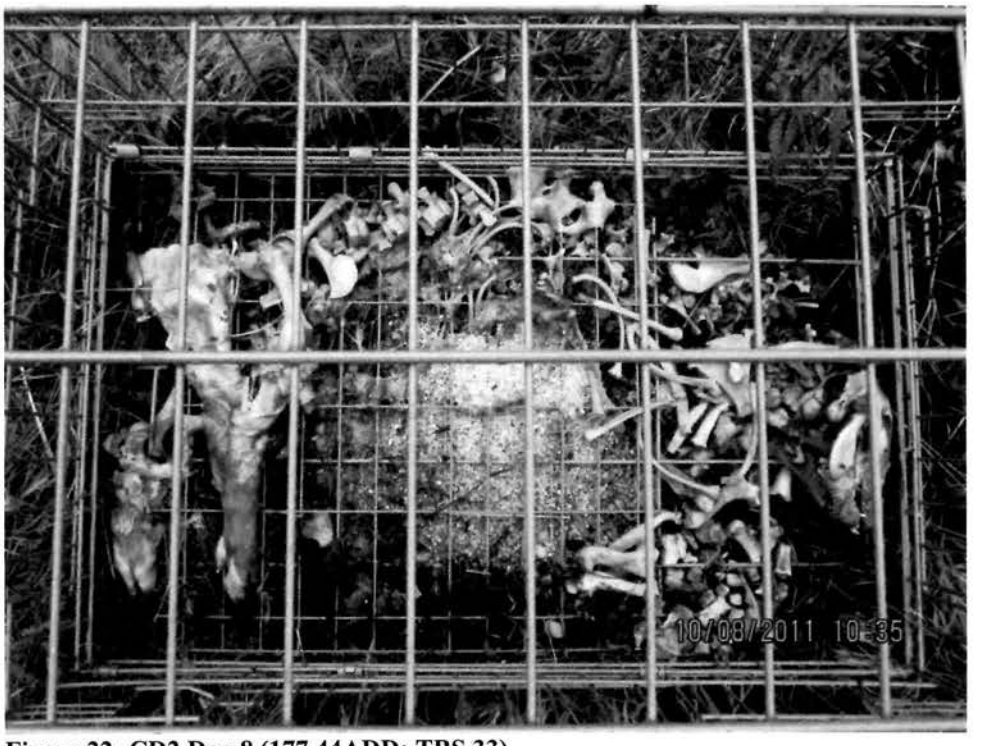


Figure 22: CD2 Day 8 (177.44ADD: TBS 33)

PD1

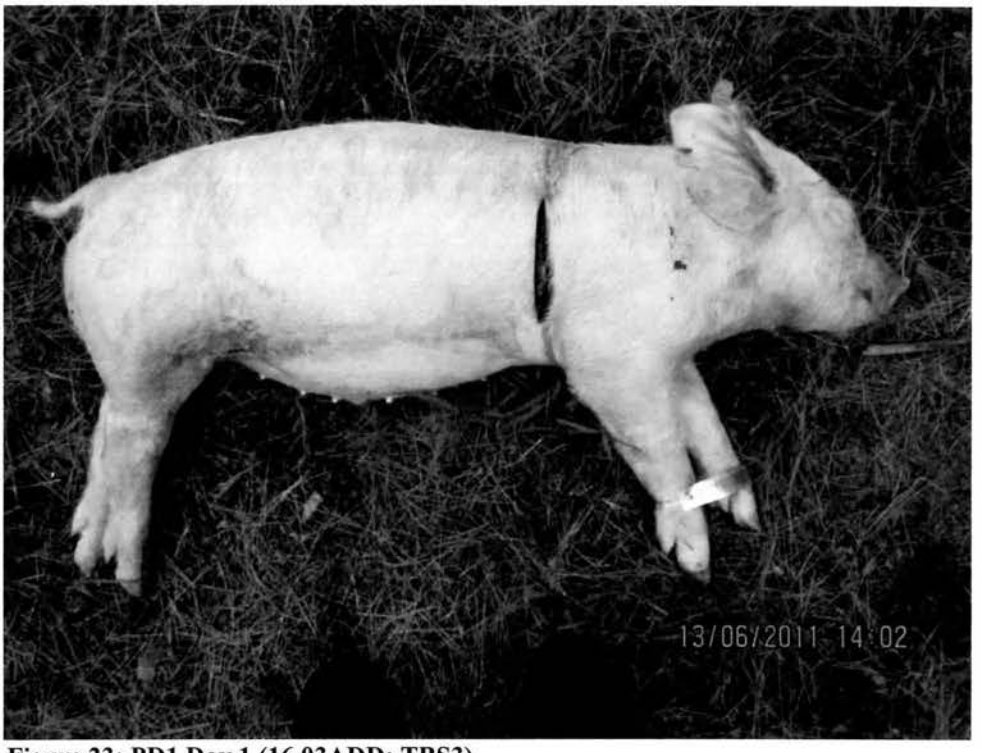


Figure 23: PD1 Day 1 (16.03ADD: TBS3)

Subject PD1 was placed out at the Holliston facility on June 13th, 2011 and was assessed with a score of 3. Little by way of decomposition and insect activity was seen on the subject until June 16th (day 4: 67.67ADD) with the presence of blow-flies at the eyes, snout, and wound site. A large egg mass was present on the superior ⅓ of the remains. The remains were bloated with the abdomen being pink with greenish tints mixed in. The head was white in color with the neck region being a dark red. For the 16th, the subject was assessed as a TBS of 7 with the head and limbs assessed as 2s and the trunk as a 3. By the following day the abdominal organs had extruded, and there was a large egg and 1st instar mass located at the wound site and on the facial region. Though the abdominal organs had become expelled, the subject was still in a full bloat stage. The

limbs were green in color with the left forelimb having a large grey to black spot on the proximal ½.

Figure 24: PD1 Day 4 (67.67ADD: TBS 7)

Figure 25: PD1 Day 5 (85.67ADD: TBS 10)

On June 19th (day 7: 127.19ADD) the subject had attained a TBS of 14 with the head receiving a score of 6 and the limbs and trunk receiving scores of 4. The skin around the wound site and of the head and neck region was brown and leathery in appearance. The abdominal organs had become exposed to its greatest extent with the skin around the organs grey to black in appearance. The skin on the left forelimb had begun to slip, with some additional slippage around the head. A very small maggot mass was located in the mouth and nose with some located at the wound site, but overall insect activity was low compared to the other subjects at the same time. By the following day, the extent of the brown leathery skin had expanded and included a large patch on the right rump. The abdominal organs had collapsed and were deteriorating in their own right. The area around the wound site became light tan in color with a more putrefactive appearance. The flesh around the snout and eyes appeared to be peeling away from the bone and the only real presence of maggots appeared to be on the wound site itself and in the mouth and nasal orifices. On July 20th (day 8: 144.92ADD) the subject was assessed with a TBS of 16 with the head receiving a score of 6 and the trunk and limbs a score of 5 each.

Figure 26: PD1 Day 8 (144.92ADD: TBS 16)

On June 21st (day 9: 165.17ADD) the first bone exposure appeared, with a rib protruding from the wound site. The skin immediately around the wound was black in color with the region having more of a leathery appearance. There were some areas that still looked pinkish-white, but the percent of surface area of that hue was diminishing. Around the extrusion site, the skin was black to dark greenish-brown with the region inferior to the site maintaining a bloated state. A mass of 2nd and 3rd instar maggots were located under the skin around the extrusion site, yet no maggots were present at the initial wound site or in the facial region. The skin around the facial region was dark brown in color with minimal, with some bone exposure on the snout, and the color of the skin of the limbs ranged from a light tan to black throughout. By the following day, most of the flesh had peeled away from the bones but was still present. The skin around the head was

tan and leathery with the bones of the snout exposed. Most of the skin of the abdomen was dark brown to black and leather like in appearance with several ribs exposed. The limbs were dark red to black with no bone exposure. There were two large maggot masses present, one engulfing the inferior ⅔ of the subject, and a smaller one on the head just dorsally to the neck. The larger mass appeared to be operating under the skin where skin was present.

Figure 27: PD1 Day 11 (201.22ADD: TBS 20)

By the 24th (day 12: 216ADD) much of the active decomposition appeared to have ceased. The bulk of the skin remained and was tan to brown in color and leathery in appearance. No further bone exposure had taken place and a portion of the maggot masses on the head and torso had diminished. On June 25th (day 13: 233.33ADD) the skin had regained moisture due to the rainfall and the torso of the remains was covered in

foam. The skin of the proximal ⅔ of the hind limbs appeared to be going through a second state of putrefaction with the skeletal elements exposed but covered in a grey-green film. That skin which was not covered by the foam remained leathery in appearance and the subject was assessed with a TBS of 20. By the following day the foam had dissipated and many of the skeletal elements were exposed. A band of flesh remained attached to the vertebral column as well as to the ribs and cranium. From this point the flesh gradually receded with the subject reaching a terminal mummification point on July 2nd, 2011 (day 20: 380.92ADD) with a TBS of 29. The experimental trial was terminated on July 12th, 2011 with no change in the subject since the 2nd.

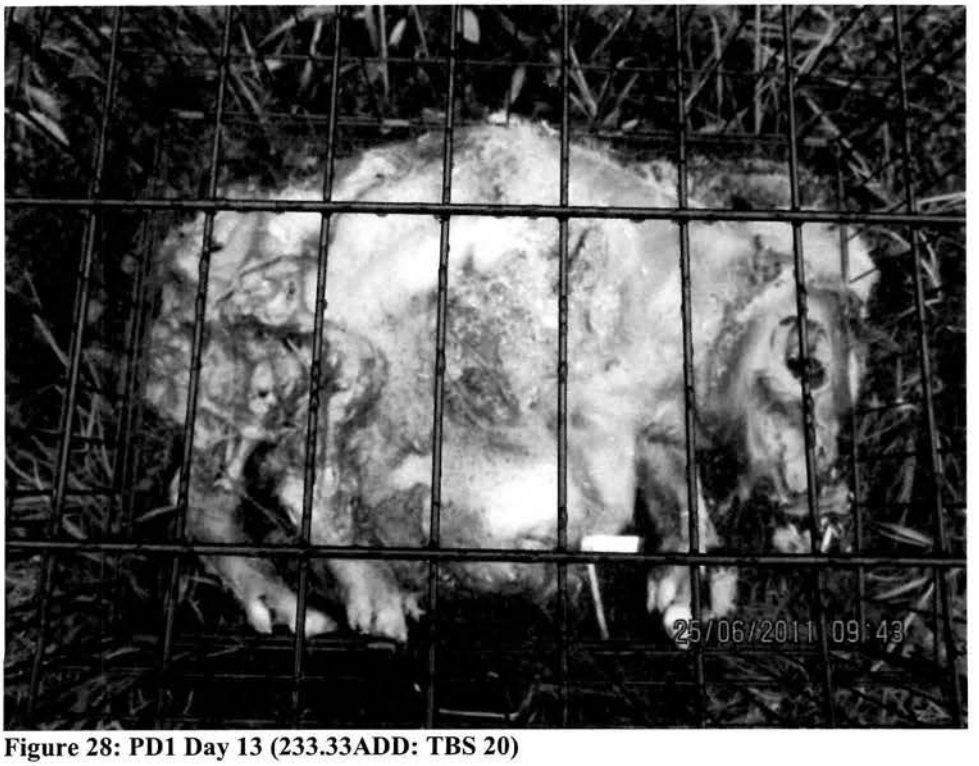


Figure 28: PD1 Day 13 (233.33ADD: TBS 20)

Figure 29: PD1 Day 30 (562.67+ADD: TBS 29)[5]

[5] This photo was taken 2 days after cessation of the experimental trial; the ADD had increased beyond what data has been collected, yet the TBS remained constant.

PU2

Figure 30: PU2 Day 1 (16.03ADD: TBS 3)

PU2 was placed out at the Holliston facility on June 13th, 2011 and was assessed with a TBS of 3. Decompositional changes began to appear in less than 24hrs with the skin turning pink-white with some slight bloating. The skin of the abdomen, while white with pink spots, did have a green tint. A large amount of blow-fly activity was present starting on June 16th (day 3: 47.33ADD), particularly at the wound site with additional groupings at the eyes, nose, and mouth. By June 17th (day 4: 67.67ADD) a large egg mass was found on the head and neck region, but was absent from the wound site. Abdominal organs, however, had begun to protrude from the wound site[6]. The abdomen

[6] It should be noted, that all of the subjects did eviscerate at approximately the same time; however, PU2 was the only subject were the abdominal organs became exposed via the wound rather than the abdominal

was bloated on this day with the skin remaining pinkish-white but with a green tint. On June 17th the subject was assessed with a TBS of 8 with the head and trunk receiving scores of 3 and the limbs a score of 2.

Figure 31: PU2 Day 6 (107.5ADD: TBS 8)

On June 18th (day 6: 107.5ADD) the subject was in full bloat with a heavy insect presence at the wound site. An egg and maggot mass was present at both the wound site and around the snout. By the following day, the skin around the left forelimb and neck was black to green in color and the cranial elements of the snout became exposed. The skin around the wound site was dark brown and leathery in appearance while the remaining abdominal skin was white to red in color and still fresh looking. By June 20th (day 8: 144.92ADD) the subject was in full bloat with the skin immediately around the

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wall. This was most likely due to the fact that either the diaphragm had been compromised, either naturally through the decompositional process or through an accidental incision.

wound black in color. That area was in itself surrounded by an area of skin that was brown and leathery in appearance. Almost half of the viscerocranium was exposed with the flesh peeling away from the cranium and the skin being black in color. The limbs were all tan to red in color with some skin slippage with the exception of the left forelimb which was black in color. The skin color at the fold between the right hind limb and the torso was also black and leathery. For June 20th, the subject was assessed with a TBS of 16 with the head receiving a score of 7, the abdomen a 4, and the limbs a 5.

Figure 32: PU2 Day 9 (165.17ADD: TBS19)

By June 21st (day 9: 165.17ADD) a large maggot mass was seen engulfing the wound site, extending along the ground and in between the hind limbs along the folds between the limbs and the trunk. More of the viscerocranium was exposed with the edges of the skin being dark brown to black in color. In between the forelimbs was a dark

red to black color with the limbs themselves being brown to tan and leathery in appearance. The distal aspect of all limbs was grey to black in color. Bloating appeared to have receded some and the subject was assessed with a TBS of 19. Due to the extensive maggot mass on the 22nd (day 10: 184.53ADD) little qualitative assessment was conducted, however by the 23rd (day 11: 201.22ADD) the complete cranium was exposed as was the majority of the skeletal elements of the remains. A sizable amount of dark reddish-brown skin remained on the neck region but appeared to be detached from the skeleton. Most of the inferior and distal portions of the remains were still engulfed in the maggot mass yet it was possible to visualize that predominantly bone remained. By June 26th (day 14: 254.31ADD) only dried bone remained with no real maggot activity left; the subject was assessed with a TBS of 35. Little flesh remained, but that was only found adhered to the containment unit rather than the remains.

Figure 33: PU2 Day 11 (201.22ADD: TBS 31)

Figure 34: PU2 Day 14 (254.31ADD: TBS 35)

PU3

Figure 35: PU3 Day 1 (16.03ADD: TBS 3)

This subject was placed at the Holliston facility on June 13th, 2011 and was assessed with a TBS of 3 since all aspects of the subject were fresh. Little activity was present until the 15th (day 3: 47.33ADD) when the subject began to show some signs of bloating. By the following day, the trunk was bloated and green in color and the organs had begun to eviscerate. Insect activity was present at the wound site and point of extrusion, but limited in the area of the snout and other natural orifices. The skin of the head and limbs were pink and white with a clear delineation of the abdomen from the fore- and hind limbs; the forelimb delineation mark is at the wound site. The subject was assessed with a TBS of 8 with the trunk receiving a score of 4 and the head and limbs scored as 2s.

On June 17th (day 5: 85.67ADD) there was a large egg mass on the chest between the fore limbs and a 1st instar maggot mass at the wound site. More of the abdominal organs had become expelled with the skin surrounding it dark brown in color. By the 19th (day 7: 127.19ADD) the abdominal organs reached their fullest extent of exposure and had begun to collapse and deteriorate. The skin around the site maintained a dark brown to black appearance, but the coloring had extended to the medial aspects of the hind limbs which had started to turn black in color. The right forelimb was also greyish-black in color though the left was still white. The skin in areas of folds had become tan and leathery in appearance with some skin slippage. PU3 was assessed with a TBS of 14 on the 19th, with the head receiving a score of 4 and the trunk and limbs scores of 5. The maggot mass was still present at the wound site, but had diminished in size; however, more maggots appeared on the abdominal organs than in previous days.

Figure 36: PU3 Day 4 (67.67ADD: TBS 8)

Figure 37: PU3 Day 7 (127.19ADD: TBS14)

By June 21st (day 9: 165.17ADD) the abdomen had collapsed, though the head and neck still appeared to be bloated somewhat. The skin around the wound site and on the hind limbs was overall tan and leathery in appearance. A pool of putrefactive fluid was present where the exposed organs had been, and there was a maggot mass that extended from the dorsal to the ventral aspects and formed approximately a six-inch "belt" encompassing the distal abdomen. Overall the skin was tanner in appearance than any other hue but there were still spots of pink and white. No bones were exposed and the subject was assessed with a TBS of 15.

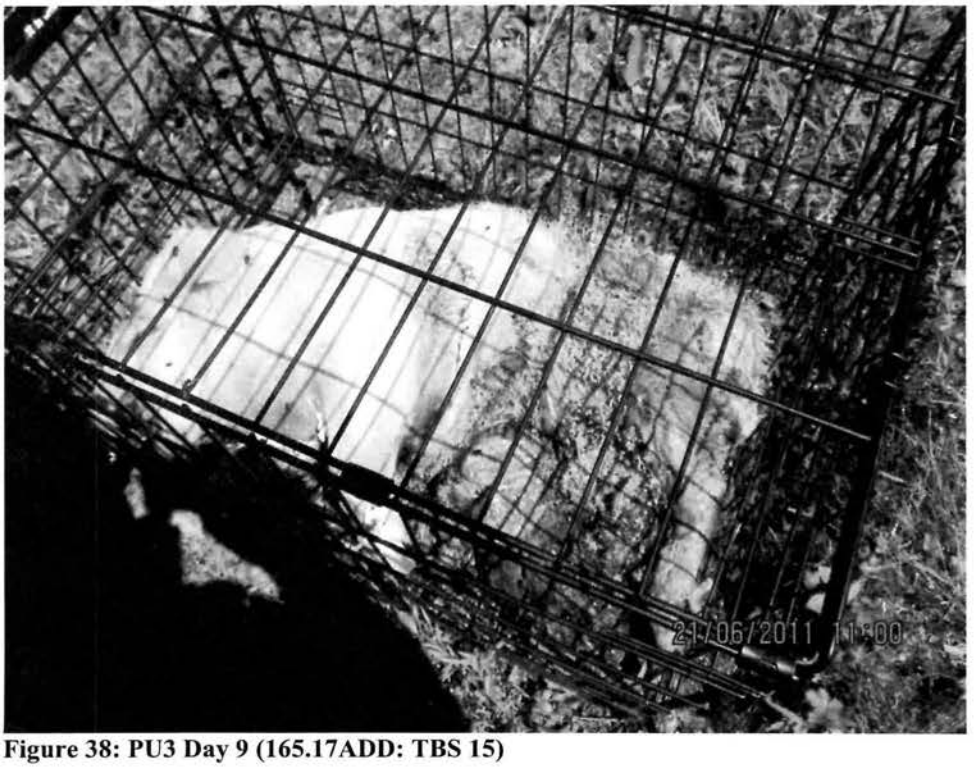

Figure 38: PU3 Day 9 (165.17ADD: TBS 15)

On June 23rd (day 11: 201.22ADD) the subject had fully collapsed with a large maggot mass engulfing much of the neck, thorax, and proximal abdomen. The skin was tan and leathery in appearance with bone exposure in all regions of the body. The skin of

the head, while attached, had lost its form and adhered to the contours of the skull. Much of the area around the remains was covered in putrefactive liquid as was the internal aspect of the subject. By the following day, much of the skeleton had been exposed but was covered in the putrefactive liquid. There was a wide band of tan, leathery skin extending from the occiput to the tail and encompassed both scapulae which on the 25th (day 13: 233.33ADD) was soft and pliable and almost gelatinous in appearance. By June 26th (day 14: 254.31ADD) the skin re-dried out and the subject reached a point of terminal mummification with a score of 34 with the head receiving a score of 13 and the limbs a score of 10, because they were completely devoid of skin and the skeletal elements were dry, and the trunk was assessed with a score of 11 because those elements that were exposed were dried, and only that bit of mummified tissue remained.

Figure 39: PU3 Day 14 (254.31ADD: TBS 34)

ND1

Figure 40: ND1 Day 1 (16.03ADD: TBS3)

Subject ND1 was placed out at the Holliston facility on June 13th, 2011 and was assessed with a TBS of 3. Decompositional changes were seen less than 24hrs later, with the abdomen becoming bloated and green in coloration with pink and white patches throughout the remains. By June 16th (day 4: 67.67ADD) there was a large blowfly presence with most of the insects congregating near the wound site, snout, and eyes. On the 17th (day 5: 85.67ADD) the subject was in full bloat with the abdominal organs exposed. The skin of the forelimbs was greyish-black as was the area around the neck. The skin of the snout was light tan in color and the hind limbs were still fresh. There was an egg mass located just ventrally to the wound site as well as a mass in the mouth, nose,

and eyes. On the 17th, the subject was assessed with a TBS of 12 with all three regions receiving a score of 4.

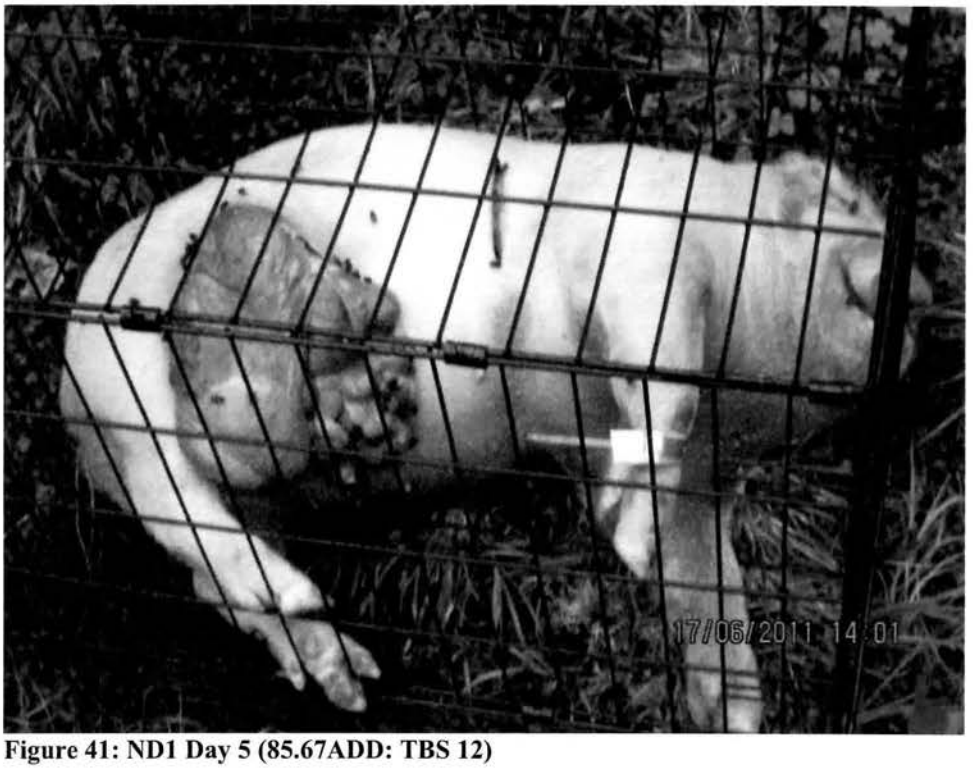

Figure 41: ND1 Day 5 (85.67ADD: TBS 12)

On June 19th (day 7: 127.19) the subject remained in full bloat with the wound relatively unchanged. The skin around the ventral surface of the neck and thorax was dark red to greyish-black, as were the forelimbs. The skin around the snout and face was tan and leathery with blowfly activity located in the mouth, eyes, and nose. The abdominal organs were fully exposed, had collapsed, and were deteriorating. There was insect activity at that site with the skin surrounding the organs being black in color. The subject was assessed with a TBS of 16 with the head receiving a score of 6 and the trunk and limbs scores of 5. By the following day, the exposed organs were covered in a maggot mass, and there was a mass found within the nose and eyes. The skin, overall,

had turned a dark red, particularly the area of the distal trunk and the hind limbs. Skin slippage was present on the hind limbs but not on the fore. The skin around the snout was tan and leathery but still maintained its form and the nose was a dark red, almost maroon, and dry. There was still little activity around the wound site save that the edges were drying some. The subject was assessed with a TBS of 17 with the head and trunk receiving scores of 6 and the limbs a 5.

Figure 42: ND1 Day 8 (144.92ADD: TBS 17)

By June 21st (day 9: 165.17ADD) the subject had progressed to a TBS of 19. The bloating had collapsed and much of the skin was turning dark red. There was still a large maggot mass present with little activity near the wound site, however, the edges of the wound site and the extrusion site have blended. No bone had really been exposed at this point, but the outline of ribs could be seen. The skin around the snout was still tanned

and leathery, and still maintained its form. Insect activity in the facial region had ceased and shifted to the abdomen. By the 23rd (day 11: 201.22ADD) the subject was predominantly bone. A large maggot mass remained, engulfing the vast majority of the remains, and what bones were present were very wet and greasy. What skin remained was light tan and leathery in appearance, however the extent of the maggot mass prevented a thorough qualitative assessment. By the following day, however, it was clear that mostly bone remained with a band of flesh adhering to the vertebral column and some of the ribs. The bones were still greasy and wet in appearance and the subject was assessed with a TBS of 30.

Figure 43: ND1 Day 13 (233.33ADD: TBS 30)

On June 26th (day 14: 254.31ADD) most of the bone had dried, but there was still the skin present along the trunk; the subject was assessed with a TBS of 31. By June 28th

(day 16: 298.33ADD), most of the remaining skin had deteriorated and the remaining bones had started to become bleached. The subject attained a terminal TBS score of 35 on June 29th (day 17: 321.69ADD) when all of the skin had detached from the bones and all of the bones had dried.

Figure 44: ND1 Day 16 (298.33ADD: TBS 34)

NU2

Figure 45: NU2 Day 1 (16.03ADD: TBS 3)

Subject NU2 was placed out at the Holliston facility on June 13th, 2011 and was assessed with a TBS of 3, given that all regions were fresh. Beginning signs of bloat could be seen by the next day, with even greater signs of decomposition being seen by the 15th (day 3: 47.33ADD). By then, the abdomen was bloated with a greenish hue, and the abdominal organs had begun to eviscerate. The limbs and head were white and pink tints and were both assessed with regional scores of 1. There was blow- fly activity at the natural orifices, but little at the wound site. By June 16th (day 4: 67.67ADD) the abdomen had continued to expand with a darkening of the greenish tint. The abdominal organs became even more exposed which resulted in more insect activity at that site. An

egg mass could be seen in the nose and mouth, but little activity (either insect or egg) could be seen at the wound site.

Figure 46: NU2 Day 4 (67.67ADD: TBS 8)

On June 18th (day 6: 107.5ADD) the subject had progressed to a TBS of 9. The green coloration has extended to the chest region, between the forelimbs and along the neck. The abdominal organs continued to expel but had lost their overall form, becoming more of a congealed mass. There was an egg and maggot mass present in the nose and mouth and blowfly activity at the extrusion site. However, the wound site was still without any blowfly or maggot activity and there was little by way of decompositional changes.

On June 19th (day 7: 127.19ADD) the greatest change in decomposition could be seen. The face and right forelimb had begun to turn grey in color and a darkening of the

nose and a blackening of the flesh around the mouth. The neck had remained pinkish in hue but was turning redder in color, and the left forelimb was becoming dry and leathery. The abdomen was white save for the skin around the extrusion site which was a reddish-brown and leathery in texture. The abdominal organs had continued to expand but were beginning to deteriorate. There was a large maggot mass present along the right (ground) side of the remains which extended up, along the fold around the forelimbs and to the wound site. The subject on the 19th was assessed with a TBS of 13 with the head and limbs receiving scores of 4 and the abdomen a score of 5. The following day, the remains had been entirely covered in a large maggot mass, but what flesh could be seen was dark brown and leathery in appearance.

Figure 47: NU2 Day 7 (127.19ADD: TBS 13)

Figure 48: NU2 Day 8 (144.92ADD: TBS 19)

By June 21st (day 9: 165.17ADD) the bulk of the maggot mass had left the remains. The remaining tissue had begun the mummification process, becoming dry, leathery, and brittle with little moisture and practically no bone exposure. By the following day, some of the bones of the snout could be seen, but little else of the skeleton. On the 24th (day 12: 216.47ADD), the day's rain added a bit of moisture to the remaining flesh, causing a return of a maggot mass to the remains, though not as dense as before. Some of the flesh had restarted the putrefaction process, and the bones of the right forelimb were exposed. Overall, however, the skin remained brown and leathery, though it maintained that moistened state until the 26th (day 14: 254.31ADD). From the 26th until July 1st (day 19: 359.39ADD), little by way of changes to the remains could be seen save a gradual rise in bone exposure. The bones of the limbs became devoid of flesh

and had dried out, and more of the skull became exposed. The distal aspects of 4 ribs had likewise become exposed. By July 1st the subject had reached a point of terminal mummification with a TBS of 31; the head receiving a score of 11 and the abdomen and limbs receiving scores of 10 each.

Figure 49: NU2 Day 16 (298.33ADD: TBS 29)

NU3

Figure 50: NU3 Day 1 (16.03ADD: TBS 3)

Subject NU3 was placed out at the Holliston facility on June 13th, 2011 and assessed with a TBS 3 as all regions were fresh. Decompositional changes were seen by the following day with the forelimbs and head turning red and the abdomen having a greenish tint; no real bloating, however, is present. By the 16th (day 4: 67.67ADD) the subject had become more bloated and the abdomen had become green in color. The distal forelimbs were red with the proximal being more white in color and the hind limbs remaining pink. The neck was red as was the nose and snout which was drying, yet the rest of the head was grey. Blowfly activity was seen at the natural orifices at the head and wound site and there was an egg mass located in the nostrils. The beginnings of

extrusion could be seen just superior to the right hind limb, and there was blowfly activity located at the site.

Figure 51: NU3 Day 4 (67.67ADD: TBS 7)

By the 17th (day 5: 85.67ADD) bloating had continued with the abdomen turning an even darker green. The abdominal organs had become eviscerated with blowfly activity located on the exposed organs. Little activity, however, existed at the wound site. The face had maintained grey coloring and the egg mass had developed into a 1st instar mass. On the 19th, the extruded organs had reached their greatest exposure amount and had deteriorated to the point where putrefactive liquid had begun to purge. The subject had reached its maximum point of bloat with the abdominal skin almost white in color. The skin around the extrusion had turned dark brown and leathery, with 50% of the edges turning black. The skin around the exposed organs was dark red in color with

some skin slippage. There was a large amount activity around the wound site with an egg mass located inside the wound, though little decompositional changes had taken place around the edges. The skin in the area of the chest between the forelimbs was red in color with the skin of the limbs themselves being grey to black. The skin of the left forelimb had split and there was a maggot mass located in the skin. The skin of the face had become tan and leathery and had begun to separate from the skull, though it maintained its form. There was a maggot mass located in the mouth with the surrounding skin black in color. The nose was a dark red and there was a small mass located in the nostrils. The subject was scored with a TBS of 15 with the head receiving a score of 6, the trunk a 4, and the limbs a 5.

Figure 52: NU3 Day 7 (127.19ADD: TBS 15)

On June 20th (day 8: 144.92ADD) a large maggot mass had engulfed the remains. The skin of the left forelimb had turned black and had separated from the bone with the skin of the right forelimb being tanner in color and was adhering to the bones. The left hind limb was dark red to black in color but had still maintained its form while the skin of the right hind limb was black with some bone exposure present. The skin around the rest of the remains was turning dark brown and leathery, though photographs make it hard to see due to the excessive amount of white fur. The subject was assessed with a TBS of 19 with the head and trunk scored as 7s and the limbs as a 5. By the following day, most of the maggot mas had left the remains to pupate with only a few remaining. The skin had become dark red to black and had desiccated and there was still little bone exposure. On the 23rd (day 11: 201.22ADD) maggots were still present, yet dissipating. The skin had deteriorated, but what skin remained was dark brown to black and dried. The bones of the limbs and many of the ribs were exposed, yet greasy, and the skull was mostly exposed. By the following day most of the bones had become exposed with a band of skin extending from the superior aspect of the skull, along the vertebrae, to the tail. Most of the exposed bones were dry and the subject was assessed with a TBS of 31. On June 27th (day 15: 275.97ADD) the subject reached a terminal TBS of 35 with all of the bones exposed and dried. What skin that remained had adhered to the cage rather than the bones.

Figure 53: NU3 Day 11 (201.22ADD: TBS 29)

Figure 54: NU3 Day 15 (275.97ADD: TBS 35)

LIST OF JOURNAL ABBREVIATIONS

J. Agr. Entomol.	Journal of Agricultural Entomology
USHoRJ Committee	U.S. House of Representatives Judiciary Committee
G. P. O.	Government Printing Office

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