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DECOMPOSITION AT THREE AQUATIC AND TERRESTRIAL SITES IN SOUTHERN LOUISIANA

A Thesis

Submitted to the Graduate Faculty of the Louisiana State University and Agricultural and Mechanical College in partial fulfillment of the requirements for the degree of Master of Arts

in

The Department of Geography and Anthropology

by Paul Max Bangs B.A., Louisiana State University, 2012 May 2014

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ABSTRACT

To compare decomposition potential variability in geographically distinct sites, this project deposited six pigs of comparable size to adult humans in pairs at three sites within southern Louisiana. These sites were: the east bank of the Mississippi River at Baton Rouge, "Red Pass" in the Bird Foot Delta near Venice, Louisiana, and the Grande Isle and Grand Terre Islands in the Gulf of Mexico. After partial decomposition of the three pairs of remains, one pig was placed onto land to simulate beaching by tidal action or recession of flood water. Each pig was allowed to continue decomposition. Despite geographic distance between the three sites, the Fresh stage lasted less than one day (Day 1), and the Bloat stage lasted six-seven days (Day 2-Day 7 or 8). The length of time remains lasted in the Active stage of decomposition varied with stage delineation becoming blurred in the aquatic pigs. Scavenging of remains was documented at every site. Alligators scavenged remains beginning on Day 9, completely removing the pigs on Day 10. Fish, including striped mullet (*Mugil cephalus*) schooled in masse around the pigs at Grande Isle, a new phenomenon to the forensic literature. Coyote (*Canis latrans*) scavenged the remains of the land pig at the Baton Rouge site, scattering bones and leaving evidence including a musk odor, footprints, and feces. The intensity of insect activity at the three sites varied, with the Baton Rouge site containing the most flies on the remains at a given time. The fly species present at the sites were largely congruent, with Chrysomya megacephala, Lucilia sericata, and Cochliomyia macellaria representing nearly all of the flies captured during the Fresh, Bloat, and Active stages. This study also shows that it is possible for feral pigs to be utilized by researchers as human surrogates of comparable weight by forensic researchers.

CHAPTER 1: INTRODUCTION

Forensic anthropologists understand that direct observation of decomposition under controlled circumstances can show trends that later can be applied to recovered human remains (Christenson 2004). Weather and geographic location have been shown to greatly effect decomposition (Komar 1998). As such, experimentation, identical in basic methodology but varying geographic location or seasonality, can help researchers predict unforeseen influences in decomposing human remains. Many of these studies vary little except by weather conditions and/or geographic location.

Beyond weather and geography, aquatic or terrestrial decomposition of remains can affect the speed of decomposition or clear delineation of stages (Ayers 2010). Remains that have been deposited on the surface of a warm climate terrestrial environment decompose relatively fast (Catteneo 2007). Insects quickly locate the remains and begin the process of colonization (Ament et al. 2004). Warm temperatures allow for a rapid expansion of the bacterial colonies that are present within the tissues of remains, while cooler temperatures will retard this expansion (Pinheiro 2006). An aquatic environment will slow overall decomposition. Water can be cooler than air, leading to a slowing of decomposition by bacteria. No necrophagous aquatic insects have been documented. As such, within submerged environments, this influential accelerant in decomposition is not present.

The effect on decomposition of the deposition of partially previously submerged, partially decomposed remains on land through natural forces in Louisiana has not been documented. Forensic investigators, with the current models of decomposition, would not have a published reference when establishing PMI.

To conduct an experiment of this nature, pig carcasses often are used to closely replicate human remains (Schoenly et al. 2007). Due to logistical difficulties, many of the previous experiments conducted in Louisiana have been unable to use pigs whose size accurately represents adult humans. Stillborn piglets, or rollover piglets (piglets that have been accidentally killed as a result of the sow "rolling over" it), have been used to replicate child-sized remains. Other institutions have published studies using large pigs which have been purchased from ranchers and euthanized (Sharonowski et al. 2008). A previously unexplored avenue has been culled adult pigs. Wildlife and Fisheries departments, which routinely cull feral pigs (Barrett et al. 1998; Coweled et al. 2012), may be willing to donate these pigs to forensic anthropology programs for use in experimentation.

This experiment will document decomposition in the autumn (September 27, 2013 – November 29, 2013) by observing the decomposition of pig remains at three geographically separate sites. Pairs of pigs will be used at each site. Both will be deposited in the water portion of the environment on Day 1. After partial decomposition has occurred, one pig from each site will be moved onto land and allowed to continue decomposing. The succession of the stages of decomposition according the model proposed by Anderson (2004), insect succession, and animal scavenging will be recorded and discussed. Additionally, this project stands as a case study for the use of culled feral pigs as human surrogates in forensic research.

CHAPTER 2: LITERATURE REVIEW

The term decomposition refers to the series of processes that occur within the remains of an organism after death. These processes break down the remains through either exploitation of nutrients by other organisms (Rothschild and Schneider 1997), or breakdown of the tissue as a result of the cessation of metabolism fueled homeostasis (Vass et al. 2002). Many of these processes are the result of the colonization of remains by microorganisms such as bacteria (Vass 2001) and fungi (Carter and Tibbett 2003). Scavenging of the body can be undertaken by animals including arthropods, mammals, and birds (Komar 1999). This consumption of remains can be an opportunistic behavior. In the case of the coyote (*Canis latrans*), the animal commonly preys upon live organisms and consumes carrion when available (Glimour and Skinner 2012). Alternatively, scavenging may be the exclusive feeding behavior of an organism, as is the case of the American Black Vulture (*Coragyps atratus*) (Reeves 2009) and many of the necrophagous insects. These organisms, if understood sufficiently, can hold forensic significance for an investigator. This significance can be beneficial and assist in establishment of a timeline since death (Turner and Wiltshire 1999). Many insect species, including the Hairy Maggot Blowfly (Chrysomya rufifacies) (Sukontason et al. 2005) and beetles of the family Histeridae (Centeno et al. 2002) can show, through either presence or life stage, time since death, known as the postmortem interval (PMI). Other species, including the Red Imported Fire Ant (RIFA, Solenopsis invicta) (Well and Greenberg 1994) and the Yellow Crazy Ant (Anoplolepis gracilipes) (Chin et al. 2009) can hinder proper estimation of PMI by eradicating evidence through the consumption of other indicative species.

Decomposition has been studied extensively to establish stages that human remains pass through as they decompose (Reed 1958). These stages have been described differently by

different researchers, with more or fewer stages described. These stages can be heavily influenced by factors including the size of the remains, the altitude in which the remains decompose, temperature, and if the deposition occurred in a terrestrial or aquatic environment. This section will discuss the stages of decomposition in both terrestrial deposition and shallow marine deposition (7.6m) as presented by Gail Anderson (2004), augmented with information from Haglund and Sorg (2002).

<u>Fresh</u> – Remains are considered to be in the Fresh stage immediately after death. Visual evidence of decomposition may not be observable immediately. No odor is associated with this stage of decomposition. Rigor mortis, the stiffening of the body as a result of chemical breakdown in the muscular system, begins and leaves the remains in this stage. Remains are considered out of this stage when the abdomen distends, indicating bloating.

Terrestrial Decomposition – Day 0-1 – Insect colonization, with the exception of cases in which access by insects is prevented, begins at this stage. Anderson notes that this colonization will begin at the natural orifices and at any wound sites. In a terrestrial system this stage usually progresses quickly, lasting a day or less.

Shallow Water Marine Decomposition – Day 0-3 – Remains can be buoyant during this stage. Anderson observed positive buoyancy of remains until the 18 hour mark, at which point the remains sank. Feeding by fish and small crustaceans begins at this stage and occurs on the entire body, not only at orifices and wound sites.

<u>Bloat</u> – This stage of decomposition is characterized by a distended abdomen. This distention occurs as a result of the colonization of the body cavity by naturally occurring bacteria in the digestive tract. During the life of the individual, these beneficial bacteria are restricted to specific

areas of the digestive tract. After death, these bacteria leave the intestinal tract. Bacteria colonize the rest of the body by utilizing the body's system of veins and arteries. The organs of the body, which are soft and nutrient rich, are broken down quickly. The gasses that are produced are trapped within the membranes of the organs, creating the classic distention.

Terrestrial Decomposition – Day 2-10 – Gasses expand the abdomen first, followed by the extremities. Insect colonization continues to advance.

Shallow Water Marine Decomposition – Day 3-11+ - In a marine environment this stage and the later active decay can overlap or be indistinguishable. Expansion of the body is observed, but can progress more slowly than in a terrestrial environment. Feeding by crustaceans and fish continues.

<u>Active</u> – This stage is noted by the deflation of the body. The remains continue to decay, losing tissue to scavenging and bacterial digestion. Putrefaction expels chemicals from the body and creates a strong odor of decomposition.

Terrestrial Decomposition – Day 11-16 – At this stage, the carcass deflates. Chemicals released give the body a "wet" appearance. Insect colonization, except when inhibited, is extensive. Maggot masses can be noted, usually at orifices or wound sites. These masses can extend into the body cavity and create observable movement in the tissue.

Shallow Water Marine Decomposition – Day >11-30+ - Delineation between Bloat, Active, and Advanced stages may be difficult to establish, or may be present simultaneously. Anderson notes skeletonization of sections of the remains while maintaining gas-filled organs and positive buoyancy. <u>Advanced</u> – This stage is characterized by the drying of the remains. The decomposition that produced the "wet" look of active decay slows. Bones will be exposed during this stage.

Terrestrial Decomposition – Day 17-42 – At this stage, fly species such as family Calliphoridae, which have previously colonized the body, are no longer present. Flies, including family Piophilidae, and beetles, including the family Coleoptera, begin to colonize remains. Maggots, which have been feeding on the remains during active decay, leave the body en masse. The excrement produced by the maggots as they leave the body produces a death stain of discolored soil and dead vegetation that radiates from the body.

Shallow Water Marine Decomposition – There is no clear delineation between active and advanced decay. As such, this stage cannot be accurately described for this environment.

 \underline{Dry} – This stage is characterized by an end to most processes of decomposition. The remains have been reduced to bones, little soft tissue remains. If soft tissue is still present, it is desiccated.

Terrestrial Decomposition – Day 43+ - Beetle species, including members of the family Coleoptera, become the main insect species noted. Flies/maggots are no longer present. Incidental species noted. This stage shows little change over time and is usually when forensic research is terminated.

Shallow Water Marine Decomposition – Day 40+ - Some fauna can still be associated with remains. Adipocere may be noted, beginning at day 47. No clear delineation from previous stages. Remains may still float and have intact organs present.

Research that studies human decomposition reflects the multitude of variables that affect the rate at which human remains can decompose. As such, experiments are conducted that

replicate potential scenarios. Researchers then observe the decomposition in an attempt to observe predictable trends. These trends can be used to suggest the postmortem interval (PMI), or time since deposition of remains, to law enforcement when an actual timeline is not known (Simmons et al. 2010).

Much of the current decomposition research being done in the United States is conducted at forensic anthropology outdoor research facilities, known colloquially as "body farms." These enclosed facilities use indoor and outdoor environments that are under the exclusive control of an anthropology (or related) department. This oversight by researchers allows for a more precise control over variables during research. Often, these facilities have active body donation programs that allow for consistent use of human remains by the program (Bass 2004). These remains, when used in decomposition research, can demonstrate how a human body decomposes (Mann et al. 1990). As reported on the University of Tennessee website, after the conclusion of the study, remaining bones are collected and remain in the skeletal collection of the facility indefinitely.

For researchers who do not have access to human remains, pig (*Sus scrofa*) carcasses can act as adequate surrogates in decomposition research (Schults et al. 2006; Catts 1992; Byrd and Castner 2001). Pig bones have comparable thicknesses, relatively comparable shapes (with the exception of the skull and long bones), and are comparable to humans in number bones in the skeleton. The intestinal bacteria of pigs have been noted assisting in decomposition in much the same fashion as in humans. Pig skin and internal tissues have similar percentages of adipose tissue to humans. Pig skin is hairless or mostly hairless, with a texture and thickness similar to the surface of human skin (Anderson and VanLaerhoven 1996).

The method for the acquisition of the pigs used in research varies based on institutional policy. In cases in which protocol will not allow the killing of animals for research, rollover piglets, or stillborn piglets are predominantly used (Haefner et al. 2004; Pharr 2009). Rollover piglets are young animals that were born healthy, but because of accidental suffocation by the weight of the mother, die soon after death. Some institutions have protocols that allow pigs to be euthanized for use in research. In such cases, research animals are purchased from farms. The method of euthanizing the animals can vary as well. Pigs can be euthanized with mechanical asphyxia (Carvalho et al. 2000), a blow to the head (Carvalho et al. 2001), shot, or killed with a bolt gun. Care needs to be taken if a pig is to be shot. Pig skulls are thick and sloped; bullets can be absorbed or deflected, creating a less than humane method of euthanasia. A bolt gun is a device that uses an iron rod to accurately and humanely euthanize pigs in a farm environment.

The sizes of pigs used in research vary as well. If fetal pigs or rollover pigs are used, the remains can range from 1-2kg in weight. Other studies use pigs weighing 3kg (Keaton 2012), 10kg (Carvalho et al. 2000), 27.9kg (Schultz 2008), with a few using large pigs weighing 60kg (Schultz et al. 2006; Ururahy-Rodrigues et al. 2008). Researchers that euthanize animals for research can often pick animals with identical weights or features. This control over pig size can allow for more accurate observations of other factors. As of the writing of this thesis, spring, 2014, no published research examines the potential of using feral pigs as research subjects.

The following feral pig information comes from the Texas Fish and wildlife website and the Louisiana Wildlife and Fisheries websites, and is augmented by data from Sanders (1988). Pigs (*Sus scrofa*) are not native to the North American continent. Domestic pigs (*Sus scrofa domesticus*) were first brought from Europe in 1539 by the explorer Henry de Soto. His pigs were abandoned and became the first feral population. Successive waves of settlers over the next three centuries increased these wild numbers through the practice of free roaming pig herds. These same European settlers, accustomed to the sport of boar hunting in Europe, also released Eurasian Wild Boar (*Sus scrofa*) into the environment to act as game animals. Since then, populations of unrestricted swine have spread across much of the North American continent, including 39 states and four Canadian provinces. The two source populations of *Sus scrofa* and *Sus scrofa domesticus* have interbred, creating a new, hybrid population.

The average size of a feral pig is 50 kg - 75 kg. The largest feral pig recorded weighed more than 400kg. Pigs can reproduce at seven to eight months of age, with the average litter size between four to six offspring. Individual pigs within feral populations have an average of 1 ¹/₂ litters per year.

Feral swine are incredibly destructive to the environment. They are omnivorous and have been documented consuming garbage, deer corn, agriculture crops, birds, juvenile alligators and small dogs. Their instinct to root in the search for food destroys the roots of bushes and trees and intensifies erosion of the topsoil (Singer et al. 1984). Feral swine can be aggressive. The United States Fish and Wildlife Service documents five to seven fatalities a year from attacks by feral swine on humans. Indirectly, feral swine help spread diseases such as hog cholera, vesicular stomatis, and classical swine fever to domestic swine. Additionally, these feral populations can also carry diseases that can be transmitted to humans including tularemia, rabies, tuberculosis, pseudorabies, swine brucellosis, salmonellosis, leptospirosis, trichinosis, *Escherichia coli*-related illness, toxoplasmosis, and sarcoptic mange. These populations of swine are considered to act as "disease reservoirs" with the ability to perpetually re-infect domestic animals (Wood and Barrett 1979).

In response to destruction by feral swine, the Louisiana State Legislature passed a law in 2010 declaring feral swine "outlaw quadrupeds." This ruling, according to the Louisiana Wildlife and Fisheries website, granted unlimited day and night hunting from March 1 to September 1. *Sus scrofa* is the only species that can be hunted at night, has an unlimited bag limit, and can be hunted with automatic firearms. LDWF promotes trapping on private land by the landowner and assists when possible. During the summer of 2013 LDWF, in association with the USDA, conducted a systematic culling of pigs from public lands across Louisiana. Among the areas in which donated pigs can shed insight is in the behavior of and taphonomy produced by native animal species.

In a constant search for food, many animals have been observed opportunistically or exclusively feeding on animal remains (Wilson and Wolkovich 2011). This kind of feeding can impede a forensic investigators ability to gather necessary evidence. To better understand how these behaviors apply to investigations involving human remains, forensic scientists have conducted research on various animals from a forensic perspective. If these behaviors are understood, these observations could be accounted for or utilized to better predict PMI (Asamura et al. 2004). Of the notable scavengers native to Louisiana, the American alligator is possibly the least understood by forensic researchers.

The American Alligator (*Alligator mississippiensis*) is a large, carnivorous reptile that inhabits the freshwater and brackish water environments of the American southeast, including southern Louisiana (Tamarack 1984; Ryberg et al. 2002). These animals are usually slow and cautious, relying on their dark, armored skin to act as camouflage. Although alligators are born weighing .065kg, adults can weigh greater than 275kg (Woodward et al. 1995) and can exceed four meters in length. The largest confirmed alligator measured 5.84 meters, although the

average for modern, managed alligator populations is 2.5 meters. Alligator growth is highly dependent on temperature, with feeding behaviors and growth stopping during winter (Jacobsen and Kushlan 1989). Alligator diet varies based on size and sex but usually consists of snails, fish, small reptiles, birds, and amphibians (Delany and Abercrombie 1986). Large alligators that live in close proximity to neighborhoods have been documented consuming dogs and cats. Alligators are opportunistic scavengers and will feed on carrion when available. Attacks by alligators on humans are rare, but documented. Alligators that attack people are usually conditioned to associate people with food as a direct result of regular feeding. This feeding can be unintentional, including the dumping of trash in waterways that contain alligators. Often alligators are intentionally fed to attract them. Human deaths by alligators involve a quick lunging attack, usually when the individual is in the water or near the bank. The alligator will grab the individual and submerge. Cause of death is usually attributed to blunt force trauma and/or drowning. Alligators have been observed staying in close proximity to remains for extended periods of time before and after feeding. Alligators lack the ability to chew food. As such, feeding is accomplished by biting and ripping tissue, and spinning to remove tissue in a motion called "the death roll" (Fish et al. 2007).

Little research has been done as to the observable evidence of alligator attacks/scavenging on human bones, or the timeline for alligator scavenging once remains have been deposited. The published literature centers on cases in which people were seen shortly before, or during, an alligator attack (Langley 2005).

As a direct result of conservation efforts, alligator populations are experiencing an expansion within Louisiana. Seasonal hunting helps maintain alligator populations, but human/alligator contact is likely to increase as neighborhoods continue to expand into and

adjacent to wetland habitat. While the alligator acts as a scavenger in and around water, terrestrial scavenging is often carried out by coyotes.

The coyote (*Canis latrans*), also known as the American jackal, is a native canid species found across North America. Within Louisiana, the subspecies *Canis latrans frustor* is common in rural areas and, increasingly, inhabited areas. The coyote has seen a population explosion since the eradication of the grey wolf (*Canis lupus*) and the Red Wolf (*Canis rufus*) from Louisiana. This expansion of population has led to coyote sightings along roadways and neighborhoods, with some sightings far from wooded areas (Gese and Ruff 1996). The coyote is a social animal. Individuals are usually part of small packs consisting of a breeding pair and offspring. Packs usually number around six, but larger packs have been noted. These social bonds are hierarchical, although their bonds are looser than the pack structure of wolves (Messier and Barrette 1982).

The natural diet of the coyote consists of small animals such as mice, rabbits, and birds when available. These animals will opportunistically scavenge and have been observed stealing road kill. Attacks on domestic dogs and cats can be common in areas with dense coyote populations. Coyotes have been documented attacking people, although fatalities are rare.

Coyotes are nocturnal and territorial and will patrol a home range nightly in search of food (Bounds 1993). To claim the area against rival coyote packs, members will mark territory with a musk present in their urine. If remains of a large animal are present, coyote will heavily scent the area to claim the remains and make the area impalpable for other scavengers. When feeding on remains, small pieces will be removed and scattered around remains. This may be to allow members of the pack to eat separately, while still proximally close to remains. If remains

cannot be completely consumed, members of the pack have been documented returning on successive nights (Gese and Ruff 1997). Coyote are the most notable nocturnal scavenger, while vultures are the predominant diurnal scavenging animal in Louisiana.

The southeast United States in inhabited by two vulture species. The American Black Vulture (*Coragyps atratus*) and the larger Turkey Vulture (*Cathartes aura*) can be seen along roads, in farmland, and within the riparian wetlands adjacent to waterways. These birds are considered shy and will not land in areas with regular human foot travel. The diet of both species consists of carrion. Black vultures locate food with an acute sense of vision (McIlhenny 1939). Inversely, turkey vultures use a heightened sense of smell to locate food (Stager 1964).

In forensic contexts, vultures are important because they can quickly strip remains of soft tissues. Research has documented human remains skeletonized within twenty-four hours after deposition (Reeves 2009). Ongoing research aims to provide forensic scientists with the tools to differentiate between vulture-scavenged remains and remains that have completely skeletonized from the cycle of bacterial and insect processes (Pharr, personal communication).

Necrophagous insects can also be highly significant within forensic contexts. If factors such as temperature and humidity are taken into account, insect behavior and life cycles can be mapped with a high level of accuracy. Recognition of insects as "vanishing evidence" during the recovery process can greatly increase the precision of PMI establishment (Ament et al. 2007). In the following section the methodology for collection of this evidence, and several forensically notable insects will be discussed. Individual flies within a single species can vary in size. Overall size differences can be attributed to accessibility of food and time on remains as a maggot.

Maggots forced to pupate because of the disturbance of remains or skeletonization of remains will be smaller than individuals allowed to pupate naturally.

Chrysomya megacephala is a necrophagous fly species that only recently has been documented in Louisiana. This species arrives at remains throughout decomposition, often seen during the fresh and active stage. This fly is positively identifiable by black spiracles laterally and posteriorly to the head. Visually, this fly is distinctive from other Louisiana fly species by its large red eyes, which are quite pronounced compared to species including *Callitroga macellaria* (Pharr 2009).

Callitroga macellaria is a visually distinctive necrophagous fly in Louisiana. On the superior surface of the thorax, the fly has three dark lines. One line lies on the sagittal line, with one identical line laterally on either side (Byrd and Butler 1996).

Chrysomya rufifacies, the Hairy Maggot Blowfly (HMBF), is an invasive fly species originally from Australia. The maggots of this species are visually distinctive. Instead of having a smooth yellow outer surface, maggots of this species are darker, with rings of raised hair. The maggots of this fly species feed on remains in a similar fashion to the other fly species mentioned. Additionally, and more importantly for forensic investigators, HMBF maggots will predate smooth-skinned maggots cohabitating remains. In many instances, HMBF maggots can completely replace other species within a site. This removal of other species destroys evidence that would allow for an accurate PMI establishment (Catts and Goff 1992).

Flies of the genus *Lucilia*, known as the Green Bottle Flies, are quite commonly found on remains in Louisiana. These flies are difficult to identify as maggots. The adults, however, are a

distinctive metallic light green, and have a light spiracle on the lateral sides of the body immediately posterior to the head of the fly.

Flies of the family Piophilidae often are noted in forensic investigations in Louisiana. Maggots of species within this family are smooth skinned, having few physical differences from other flies. Behaviorally, these flies are unique. When disturbed, maggots of this family will launch themselves away from danger by holding the front and back ends of their body together, then springing themselves foreword. Additionally, it should be noted that these flies will not die if swallowed. Through a process known as intestinal myasis, maggots can pass through and exit the digestive tract unharmed, or burrow into the stomach or intestinal lining causing lesions (Friedberg 1981).

Some beetles of the family Histeridae, or Hister Beetles, are forensically significant within Louisiana. These beetles are often small and dark, with shiny exoskeletons. Hister beetles usually appear on remains in the later stages of decomposition. Adult beetles and larvae can be observed on dry remains long after flies and other insects have left. These beetles may not be immediately apparent at the site of remains. These insects will hide in the soil and under remains. If discovered, Hister beetles will fake death, allowing for easy collection. A special note should be made that some members of this family can be cannibalistic; some species will prey upon other beetles, including members of its own species. As such, collected beetles should be stored individually (Smith 1986).

Through globalization and the increase of goods and people across the globe, organisms have been intentionally or unintentionally transplanted to new areas. Examples of forensically significant species that have been introduced include the previously mentioned HMBF, the Red

Imported Fire Ant (RIFA) (*Solenopsis invicta*), and the Yellow Crazy Ant (*Anoplolepis gracilipes*). These species, like the other insects already discussed, have very specific behaviors that, if adequately researched, can be used to predict PMI. Unfortunately, evidence shows that in transplanted environments, the behavior of these species can be altered slightly or greatly. As such, these invasive species need to be researched by forensic scientists to fully understand their behavior and their role in decomposition. Some species, such as the HMBF, have existed in the United States for more than twenty years and are thoroughly researched (Catts and Goff 1992). Other species, including *Anoplolepis gracilipes*, have spread beyond their native ranges (Abbott 2005) and can now be found on the North American continent. As such, research in the new environment is non-existent and their forensic significance is unknown.

CHAPTER 3: MATERIAL AND METHODS

3.1 Study Sites

3.1.1 Baton Rouge site



Figure 1. Baton Rouge site.

The Baton Rouge site is located on the east bank of the Mississippi River immediately south of the intersection of River Road and Brightside Drive in Baton Rouge Parish, Louisiana. As seen in Figure 1, the site is bordered to the west by the Mississippi River. The eastern border of the site is the tree line that marks the beginning of the scrub and hardwood trees of the riparian wetland and natural levee. The yearly cycle of crest and recession of the river maintains a muddy bank between the tree line and the river, which measures approximately twelve meters in width when the river is lowest. This environment extends from the southern border of the Baton Rouge city limits to Greater New Orleans in Jefferson Parish. The Mississippi River water is relatively fast moving, maintaining a speed of 4.8 kilometers per hour, and quite turbid, maintaining 5cm visibility for much of the year. The water is fresh, maintaining a salinity of 0 parts per thousand (ppt).

3.1.2 Venice site

The Venice site consists of a small area of dry land on the property of the Targa Chemical Plant on Tidewater Road in the Bird Foot Delta, Plaquemines Parish. The site, as shown in Figure 2, is flat land surrounded by water on three sides. Two meter tall roso cane (Graminae sp.) grows at the periphery of the site, visually shielding it from the road. The Bird Foot Delta is the fan shaped area of land at the mouth of the Mississippi River, where the single stream of water branches off into several "fingers" as it enters the Gulf of Mexico. The site is a hybridized salinity zone, meaning its salinity varies from completely fresh in early spring, to brackish during the fall and winter. During the spring and early summer water may be entirely fresh, with saline levels maintained at 0 ppt. During the autumn and winter, the reduction of river water allows a backfilling of denser gulf water, creating a brackish environment of 8 ppt. Freshwater species, including channel catfish (Ictalurus punctatus) and American alligator (Alligator mississippiensis) can be seen year-round, as well as estuarine and gulf species including the Atlantic blue crab (Callinectes sapidus) and striped mullet (Mugil cephalus). The waterway used in this study is an inlet attached to the unnamed but unofficially titled "Red Pass."



Figure 2. The Venice site.

3.1.3 Grande Isle/Grand Terre Site

The Grande Isle/Grand Terre site is split between two Louisiana barrier islands. The Grande Isle part of the site sits on the eastern edge of the beach that acts as the northern edge of the Louisiana Department of Wildlife and Fisheries (LDWF) Station. The beach, as shown in Figure 3, is a mixture of naturally occurring yellow sand and imported concrete stones. The concrete stones are artificially deposited; the stones reinforce the beach for erosion control. The salt water, because of the steady current of river sediment from the east, is turbid, having 8cm of visibility for much of the spring, summer, and autumn months. The salinity of the water was measured to be 22ppt. Gulf fish including Atlantic croaker (*Micropogonias undulates*), speckled trout (*Cynoscion nebulosus*), and southern flounder (*Paralichthys lethostigma*), are commonly seen in the region, as well as striped mullet (Mugil cephalus), an estuarine fish species. Multiple dolphin species are often observed near the island. Because of the relative proximity to residents

and employees, the terrestrial part of the site is located a short distance northeast, on the island of Grand Terre. Grand Terre is a smaller, uninhabited island of similar proportions to Grande Isle. The portion of the island used in this study is the dirt road connecting the buildings of the abandoned LDWF station. The road, which can be seen in Figure 4, runs north to south and is bordered to the east and west by two meter tall grass and flowering bushes. No trees are present on the island. Coastal birds, including Brown Pelican (*Pelecanus occidentalis*), White Pelican (*Pelecanus erythrorhynchos*), and Thayer's Gull (*Larus thayeri*) are common. Footprints belonging to raccoons (*Procyon lotor*) and the droppings of eastern cottontail (*Sylvilagus floridanus*) have been observed on the island.



Figure 3. The Grande Isle site.



Figure 4. The Grand Terre site.

3.2 Experimental Animals

The pigs used in this experiment were obtained through donations by LDWF and USDA employees through the ongoing culling of feral swine from rural Louisiana. The collection was carried out through a combination of trapping within baited corral traps and night hunting with the use of infrared sensing cameras. Two pigs, which were collected but not used in this study, were donated on June 15, 2013. BRPIG1 and VPIG2 were donated on July 1, 2013. BRPIG2 and VPIG1 were donated on July 15, 2013. GIPIG1 and GIPIG2 were donated on August 1, 2013, and August 7, 2013, respectively. The pigs were weighed and measured before deposition at the sites. The measurements, as shown in Table 1, show that four of the pigs were similar in size and weight. The two heavier pigs, labeled GIPIG1 and GIPIG2, were used at the same site to maintain consistency of sizes at the respective sites.

Pig I.D.	Length	<u>Height</u>	<u>Weight</u>
BRPIG1	119cm	69cm	53kg
BRPIG2	127cm	66cm	54kg
VPIG1	112cm	69cm	54kg
VPIG2	122cm	74cm	55kg
GIPIG1	152cm	81cm	113kg
GIPIG2	163cm	82cm	119kg

Table 1. Pig measurements and weights.

Until the beginning of the study, the pigs were kept frozen in the freezer of the Gross Anatomy Laboratory within the Louisiana School of Veterinary Medicine (LSVM) on the Baton Rouge campus of LSU. On September 14, 2013 the pigs were moved to the East Baton Rouge Coroner's Office at 4030 T B Hearndon Ave, Baton Rouge, Louisiana. The pigs were placed in the refrigerators to thaw. The core temperatures of the pigs were all measured to be above 0°C on the seventh day in the refrigerator (September 21, 2013). After removal from the refrigerators the pigs were transported to the researcher's residence and shaved using Oster Pet clippers. Figure 5, seen below, shows the shaving of VPIG2 by the researcher. Shaved portions as well as shaved portions are visible. Because the fur of some of the pigs had blood or mud matted to it, the clipper could not shave every pig completely. The pigs were transported to the study sites outside of the study racks. The racks and caging were prepared on site.



Figure 5. Shaving a pig.

3.3 Field Protocols

The methodology for the fieldwork of this experiment was slightly modified from Anderson (2009). During the fieldwork of this project, the pigs were disturbed as little as possible. The high turbidity of the water prohibited use of underwater cameras, as utilized by Anderson (2009). To monitor the decomposition of pigs while in the water, each pig was removed from the water, documented, and returned to its original position. The pigs were observed once a week to reduce the overall impact to the decomposition process. The remains were unclothed for this study.

As shown in Table 3, each pig was brought to the three sites on Day 1 and placed in position. The sites were visited again on Day 8 and observed. On Day 10, the methodology called for one pig from each site to be moved from the water onto land and allowed to continue decomposing. Following this movement, each site would be visited once a week for the

remainder of the project. Additionally, the Baton Rouge site would be visited every day. The pattern of visitation shown by Table 2 continues, with visitations to the Venice site and Grand Isle/Grand Terre site scheduled to occur on Days 17, 24, 31 and so forth until termination of observations.

	1			
Date	Day #	Venice site	Grande Isle/	Baton Rouge site
			Grand Terre site	
Sep 27, 2013	Day 1	Visit site/Place	Visit site/Place	Visit site/Place
		pigs at the site	pigs at the site	pigs at the site
Oct 4, 2013	Day 8	Visit site	Visit site	Visit site
Oct 6, 2013	Day 10	Visit site, move	Visit site, move	Visit site, move
		one pig to land	one pig to land	one pig to land
Oct 7, 2013	Day 11			Visit site
Oct 8, 2013	Day 12			Visit site
Oct 9, 2013	Day 13			Visit site
Oct 10, 2013	Day 14			Visit site
Oct 11, 2013	Day 15			Visit site
Oct 12, 2013	Day 16			Visit site
Oct 13, 2013	Day 17	Visit site	Visit site	Visit site

Table 2. Schedule for visitation and movement of pigs.

Each pig was placed on a rack constructed with a multiple plastic restaurant trays. Because of the varying size of the pigs, some racks were constructed of two trays, while others required three. Figure 6 below shows BRPIG1 on the completed rack on Day 1. Because of the smaller size of BRPIG1, its rack was constructed with only two trays. The trays were connected with 75kg test zip ties and 1.5cm nylon rope. The rack was covered on three sides with extruded wire fencing. The wire was connected to the rack with four black, 45kg test zip ties, as seen in Figure 5. The purpose of this wire was to prevent theft of remains by large carnivores, as noted by Anderson (2009). The sites at Baton Rouge and Venice were in areas commonly inhabited by American alligators, while Grande Isle/Grand Terre is the habitat for small to medium sized sharks. This wire would allow scavenging while preventing theft. It should be noted that the diamondback terrapin (*Malaclemys terrapin*) is a protected turtle species in the brackish water of coastal Louisiana. In an effort to prevent accidental mortality of scavenging animals within the cages of this experiment, two ends of each wire cage were left open.



Figure 6. BRPIG1 on the completed rack.

Each pig was placed on the rack and photographed. The wire cages were constructed at the location. The pigs were carried into waist/chest deep water (water measuring 1.5 meters in depth, approximately 4 meters from shore). Iron rebar rods (1.75 meters in length, 2 centimeters in diameter) were hammered into the sediment to act as an anchor. Each rack was tied to a rod with 1.5 centimeter nylon cord, reinforced with 75 kilogram test zip ties. Flies had continual access to each pig in the study. Air and water temperature, water salinity, weather conditions, average wind speed, and photographs were taken at each site upon visitation.

At each site Moultrie 155 Wildlife Cameras were placed facing the pigs. These cameras were installed at the site to optimally record visitation by scavenging animals, investigating humans, and allow documentation of the stage of decomposition. Upon successive visitations to the sites, the site around the land pig was mapped on grid paper through techniques consistent with forensic recovery. Movement of the remains from previous visitations was noted. After deposition on land by the researcher, the land pig (BRPIG1) would be visited every day. This allowed the Baton Rouge site to act as the guide to contrast the environments of Venice and the barrier islands.

Flies were collected with fine mesh insect nets. Captured flies were placed in plastic restaurant cups. After returning to the lab, flies were euthanized with ethyl acetate gas and pinned to Styrofoam and identified. Maggots were collected with soft tweezers and preserved in a solution of 70/30 ethyl alcohol/water solution. Beetles, when present, were collected. Beetles were stored individually, euthanized with ethyl acetate, and pinned. Adult flies and beetles were initially identified by Paul Bangs and Lauren Pharr. When possible, identification was made at the species level.

In the case of water pigs (BRPIG2, VPIG2, GIPIG1) any disarticulated bones were collected upon discovery. Disarticulated bones of land pigs BRPIG1 and GIPIG2 were allowed to remain *in situ* to record terrestrial scatter patterns. After completion of the project, remaining bones were collected, cleaned, and indexed. Postmortem trauma, when observed, was noted, photographed, and analyzed.

CHAPTER 4: RESULTS

The results of this study will be presented in four sections. The sections titled "Weather" and "Stage of Decomposition" will address the six pigs simultaneously, while "Scavenging" and "Insect Activity" will be broken up into subsections that will describe each pig individually. This delineation is necessary because the scavenging and insect activity documented during this study varied considerably, as will be shown through the use of figures and descriptions.

4.1 Weather Data

Weather data shown in Figure 7 was taken from the Louisiana State University Southern Regional Climate Center website (srcc.lsu.edu), as documented by the nearest substations to the sites. As shown by the three lines representing the changes in temperatures over time, the difference in temperature was minimal. The Grande Isle/Grand Terre and the Venice site had the most similar temperatures, while the Baton Rouge site was the most different. The site with the warmest average temperature fluctuated between the Venice site and the Grande Isle/Grand Terre site during most of the study, with the Baton Rouge site remaining cooler than the other two by at least one degree Celsius. The only exception to this occurred on October 29, 2013, when the Baton Rouge site was recorded showing an average temperature one degree higher than Grande Isle/Grand Terre. The presence of Tropical Storm Karen in the Gulf of Mexico caused local temperatures in southern Louisiana to drop between October 4, 2013, and October 6, 2013, with temperatures returning to a seasonal norm on October 10, 2013. Despite the geographic distance in the three study sites, this drop in temperature can be seen in Figure 7 affecting the three sites simultaneously.



Figure 7. Average site temperatures

4.2 Stage of Decomposition

As noted in Table 3 below, the stages of decomposition for the six pigs were documented every day. On days in which a site was not visited by the researcher, determination of the stage of decomposition was made through analysis of wildlife camera photos of the remains.

Each pig was in the Fresh stage on Day 1. By Day 2, each pig exhibited distention of the abdomen, signaling the Bloat stage. This inflation of the body cavity with gas increased the buoyancy of each pig, increasing the amount of surface area accessible to flies. Also, the amount of rolling of the body was reduced, meaning that the side of the body exposed above the surface of the water remained more consistent than during the Fresh stage.

The Bloat stage lasted seven to eight days. GIPIG1, GIPIG2, BRPIG1, and BRPIG2 exited Bloat and entered Active decomposition on Day 8 when the abdomens of these four pigs opened below the water line, exposing internal organs. To illustrate this, the abdomen of GIPIG1 on Day 9 is shown in Figure 8. The exposed sub dermal skin and adipose tissue below the water
line can be seen as well. The lack of scavenging damage to these organs suggests that this was a result of the stage of decomposition, not aquatic scavenging alone.



Figure 8. GIPIG1 in situ on Day 9.

The Venice site pigs remained bloated until the morning of Day 9, when the first indicators of active decomposition were noted. These indicators included skin slippage on the abdominal wall and the opening of the abdomen, exposing internal organs. Although it was not documented by the wildlife cameras, employees of the Targa plant observed American alligator scavenging beginning on the evening of Day 9.

Four pigs, BRPIG1, BRPIG2, GIPIG1, and GIPIG2 continued to decompose beyond the Active stage. GIPIG2 was transported to the land site of Grand Terre on Day 12. In the days following this deposition, the remains desiccated quickly, entering the Dry stage on Day 18. As

Figure 9 shows, remains appeared to be mummified, with much of the tissue remaining. The tissue had become stiff, turning from a pale grey color to dark brown. Successive visitations to the site showed little change to the remains. After the visitation on Day 33, weekly visitations were suspended. The site was visited once more on Day 62, showing no change to the remains.



Figure 9. GIPIG2 on Grand Terre Island on Day 26.

GIPIG1 remained in its original position in the water at Grande Isle. The indicators for the stage of decomposition became blurred, with no clear delineation between the Active stage and the Advanced stage. The skin and fat became exposed, as well as the bones of the limbs and rib cage. The bones of the limbs became skeletonized and disarticulated between Day 12 and Day 19, with the skull disarticulating between Day 20 and Day 26. The remains completely disappeared the evening of Day 26.



Figure 10. BRPIG2 on Day 16.

BRPIG1 and BRPIG2 entered the Active stage of decomposition on Day 8, the same day as GIPIG1 and GIPIG2. BRPIG2, which remained in the water throughout the entirety of its decomposition, entered the Advanced stage on Day 15. As a result of the lack of buoyancy noted during the previous stages, the remains sunk below the surface of the water while remaining in the water column (not resting on the sediment). The skin and adipose tissue were completely removed during this stage. The axial skeleton surrounded the soft tissue of the internal organs, which remained largely unscavenged. Between Day 15 and Day 17 little progression of decomposition was noted. The only taphonomic change seen by the researcher was portions of the exposed soft tissue began to separate from the main body of the remains. As seen in Figure

10 above, these pieces of soft tissue remained near the body. The remains were removed by increased water level associated with a rain event on the evening of Day 17.

BRPIG1 was moved onto land on Day 10 while in the Active stage of decomposition. On Day 14 brown putrefactive fluid began to coat the remains, signaling the start of Advanced decomposition. This liquid produced a significant stain on the ground surrounding the remains. The remains continued to show indicators of this stage until removal by scavengers on the evening of Day 20. The disarticulated bones that remained were considered to be in the Dry stage. They were observed daily, with no change recorded, until Day 37, when observations were terminated.

Date	Day #	VPIG1	VPIG2	GIPIG2	<u>GIPIG1</u>	BRPIG1	BRPIG2
9/27/2013	Day 1	Fresh	Fresh	Fresh	Fresh	Fresh	Fresh
9/28/2013	Day 2	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
9/29/2013	Day 3	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
9/30/2013	Day 4	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
10/01/2013	Day 5	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
10/02/2013	Day 6	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
10/03/2013	Day 7	Bloat	Bloat	Bloat	Bloat	Bloat	Bloat
10/04/2013	Day 8	Bloat	Bloat	Active	Active	Active	Active
10/05/2013	Day 9	Active	Active	Active	Active	Active	Active
10/06/2013	Day 10		Active	Active	Active	Active	Active
10/07/2013	Day 11			Active	Active	Active	Active

Table 3. Observed Stages of Decomposition

Table 3. (Continued)

Date	<u>Day #</u>	VPIG1	VPIG2	GIPIG2	GIPIG1	BRPIG1	BRPIG2
10/08/2013	Day 12			Active	Active	Active	Active
10/09/2013	Day 13			Active	Active	Active	Active
10/10/2013	Day 14			Active	Active	Active	Active
10/11/2013	Day 15			Active	Active	Advanced	Advanced
10/12/2013	Day 16			Active	Active	Advanced	Advanced
10/13/2013	Day 17			Active	Active	Advanced	Advanced
10/14/2013	Day 18			Dry	Active	Advanced	
10/15/2013	Day 19			Dry	Active	Advanced	
10/16/2013	Day 20			Dry	Active	Advanced	
10/17/2013	Day 21			Dry	Active	Advanced	
10/18/2013	Day 22			Dry	Active	Dry	
10/19/2013	Day 23			Dry	Active	Dry	
10/20/2013	Day 24			Dry	Active	Dry	
10/21/2013	Day 25			Dry	Active	Dry	
10/22/2013	Day 26			Dry	Active	Dry	
10/23/2013	Day 27			Dry		Dry	
10/24/2013	Day 28			Dry		Dry	
10/25/2013	Day 29			Dry		Dry	
10/26/2013	Day 30			Dry		Dry	
10/27/2013	Day 31			Dry		Dry	

Table 3. (Continued)

Date	Day #	VPIG1	VPIG2	<u>GIPIG2</u>	<u>GIPIG1</u>	BRPIG1	BRPIG2
10/28/2013	Day 32			Dry		Dry	
10/29/2013	Day 33			Dry		Dry	
10/30/2013	Day 34			Dry		Dry	
10/31/2013	Day 35			Dry		Dry	
11/01/2013	Day 36			Dry		Dry	
11/02/2013	Day 37			Dry			
11/29/2013	Day 62			Dry			

4.3 Scavenging

4.3.1 Venice Site

During the initial deposition of VPIG1 and VPIG2 at the Venice site, multiple scavengers were observed in the area. Several vultures, later identified as American black vulture (*Coragyps atratus*) were photographed, as well as American alligator (*Alligator mississippiensis*), channel catfish (*Ictalurus punctatus*), Atlantic blue crab (*Callinectes sapidus*), and small minnows (*Gambusia sp*). No scavenging was observed the day of deposition of the remains.

Employees of the Targa plant noted alligator scavenging beginning in the evening of Day 9. Upon arrival at the Venice site on the morning of Day 10, multiple American alligators were again seen in the area. One, measuring approximately 170cm in length, was positioned directly adjacent to the two racks. After retrieval of the two racks from the water, this alligator remained in that position until the end of the daily observations. Figure 11 shows the alligator at the site. VPIG1 had been removed from the rack by scavengers. The nylon cords which had held the pig to the rack were not broken, suggesting that the pig had been dismembered during the removal process. VPIG2 showed extensive postmortem trauma to the posterior half of the remains, with less extensive postmortem trauma to the face and forelimbs. The remaining limbs had become partially skeletonized, but remained articulated and retained a normal range of motion.

The posterior half of VPIG2 was largely missing. As Figure 12 shows, the hind limbs, organs, vertebral column, and skin from the right side of the body were missing. The remaining skin of the left side of the body extended thirty-two centimeters beyond the last present vertebra. The last vertebra did not show trauma to the bone. Two ribs, as indicated with arrows in Figure 13, show complete fractures to their sternal ends.

Fish of the genus *Gambusia* were seen feeding on the soft tissue of the remains. Immature Atlantic blue crabs (*Callinectes sapidus*) were also noted within the remains after removal from the water.

The remains were removed from the land site on the evening of Day 10. The American alligator is the only scavenger in the Venice area that would have the strength to remove remains of that size (Dr Andrew Nyman, personal communication).

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Figure 11. Alligator (center) near the pig remains (far left) on Day 9.



Figure 12. VPIG2 on Day 9.



Figure 13. Body cavity of VIPIG2 on Day 9. Broken ribs indicated with arrows. 4.3.2 Grande Isle/Grand Terre Site Scavenging

Terrestrial scavenging animals were not readily noticeable at the Grande Isle/Grand Terre site. There were no vultures observed on powerlines or in trees as there had been at the Venice site, and coyotes are not known to travel between the barrier islands. Atlantic blue crab (*Callinectes sapidus*) are known to be in the area, but these were not noted on Day 1.

Beginning on Day 6, a large school of striped mullet (*Mugil cephalus*) was observed near the two pigs. This school did not scavenge directly from the remains. Instead, the fish were filter feeding by sucking tissue particles from the top of the water. This school numbered approximately one hundred individual fish from Day 6 through Day 20, and thirty individuals from Day 21, until the disappearance of the remains on Day 26. The striped mullet school did not dessipate when the two pigs were retrieved and returned to their positions. The fish moved a short distance away from the researcher but stayed within approximately two meters of the pigs. When the pigs were brought to the shore line, the school followed, coming within two meters of the beach, feeding continuously. The striped mullet school was observable from shore by the feeding behavior of the fish. To siphen water, the fish push their mouthes partly out of the water as they swim. This behavior, as seen in Figure 14, can be observed from a distance, even when the water is choppy. Additionally, the feeding behavior produces a unique sucking sound that signals the presence of the school.

Striped mullet are common in the area and do form schools of multiple individuals, but this school was abnormally large. This phenomenon had not been seen previously by the LDWF Biologists who witnessed the experiment (Zach Hammer, personal communication).



Figure 14. The striped mullet school feeding.

On Day 12 GIPIG2 was removed from the original position in the water near Grand Isle and deposited at the land site on Grand Terre. No scavenging animals were observed on the island. On Day 19 GIPIG2 was revisited. Several sets of bird tracks were observed near the remains, as well as bird droppings on and around the remains. Figure 15 and Figure 16 suggest the presence of vultures on the island of Grand Terre because:

- 1. Bird tracks were consistent in size and distance with vulture tracks.
- 2. Urea showing a diagonal spray pattern, a behavior seen in vultures.
- 3. A cavity on the exposed side (left side) of the body cavity, with internal soft tissue removed.
- 4. Possible evidence of feeding was observed, with little disarticulation/scattering of bones.

Figure 15 shows raccoon tracks, indicated with two white arrows. Diagonally directed urea in Figure 15 is indicated with two yellow arrows. Figure 16 shows tracks, indicated with three white arrows, which are consistent with vulture tracks. Additionally, it should be noted that raccoon tracks were observed on the main road of the island, but the tracks did not approach the remains. After the visitation on Day 19 there was no new evidence of visitation by animals.



Figure 15. Bird urea and raccoon tracks near the Grand Terre site on Day 19.



Figure 16. Possible vulture tracks near the Grand Terre site on Day 19.

4.3.3 Baton Rouge Site Scavenging

The water portion of the Baton Rouge site had little scavenging. Small fish (*Gamusia sp.*) were noted around the remains of both BRPIG1 and BRPIG2, but as was seen in Figure 10, much of the available soft tissue remained unscavenged. Larger fish, turtles, crayfish, and alligators are known to exist at the site, but were not observed scavenging.

BRPIG1 was redeposited on land on Day 10. Multiple coyotes began visiting the site and scavenging remains during the nights. The remains were visited every night between Day 8 and Day 20, with scavenging of remains occuring on Day 10, Day 11, Day 12, Day 13, Day 14, Day 15, Day 16, Day 18, and Day 20. Evidence of coyote scavenging varied by day and included:

- 1. Movement of the main body of remains (Days 11, 12, 13, 14, 18)
- Footprints around remains as well as coming from and going to the tree line (Days 11, 12, 13, 14, 15, 16, 17, 18, 20)

- Coyote trail near leading into the woods from the site of remains, first noted on Day 15.
- 4. Fresh tooth marks on remains, consistent with dragging/tearing (Days 11, 12, 13, 14, 15, 18)
- 5. Musk odor, a result of coyote scent marking, first noted on Day 11.
- 6. Dispersal of disarticulated bones
- 7. Breaking/Scratching of the ends of bones (seen in figure 25 and figure 26)
- 8. Removal of remains from the site.
- 9. Photographic evidence of coyote visitation/scavenging by the game cameras.

Figure 17 and Figure 18 are photos taken by the motion-sensing wildlife camera on the evening of Day 10 and the morning of Day 13, respectively. Figure 17 shows a profile of one of the coyotes which allows for a positive identification of species, ruling out the possibility of the scavengers being feral domestic dogs. Figure 18 shows a coyote biting and attempting to drag the remains.

The photos consistently recorded two coyotes feeding, standing, and, in the case of Figure 18, eating at the site. This suggests that the coyote pack consisted of only two individuals, and that once the carcass was located, the coyotes intentionally returned nightly. It should be noted that on the night of Day 14 the camera recorded a coyote visitation to the site followed by visitation by individuals not associated with the experiment, and a return of the coyotes after the individuals had left.



Figure 17. Two coyotes at the Baton Rouge site.



Figure 18. A coyote biting and attempting to drag remains.

Figures 19 through 24 below show the effects of coyote scavenging on the remains of BRPIG1. These drawings show the movement of the remains to the southeast in the direction of

the tree line. A small dot at the four meter line on the X axis shows the original position of the pig, as noted the previous day. Figure 20 shows the new position of BRPIG1 after two nights of scavenging. The remains had been moved only slightly toward the treeline, but had been turned considerably. Of the eighteen bones collected after the termination of this study, a total of seven had been removed by the fourth night of scavenging. As the remains were consumed, these rib bones were removed and dragged a short distance from the remains, chewed on, then discarded. These disarticulated rib bones would not be continually chewed on, moved, or removed after their initial placement by the coyotes. After removal of the anterior portion of BRPIG1, no other activity was noted at the site by scavengers. The positions of bones as shown by the map of Day 21 are identical to their positions at the termination of the study.



Figure 19. The Baton Rouge land site on Day 11.



Figure 20. The Baton Rouge land site on Day 12



Figure 21. The Baton Rouge land site on Day 13.



Figure 22. The Baton Rouge land site on Day 14.



Figure 23. The Baton Rouge land site on Day 18.



Figure 24. The Baton Rouge land site on Day 21.

Postmortem coyote scavenging was observed on the collected bones of BRPIG1. The distal ends of twelve ribs and one thoracic vertebra showed gnaw marks. The marks, seen in Figures 25 and 26, have destroyed the sternal ends of the ribs. This destruction of the rib ends

prevents aging the individual based on sternal rib ends, as outlined by Iscan et al. (1984). The rib bones in both figures have been adjusted to show the length of bone missing.



Figure 25. Undamaged sternal rib end (above) and damaged rib end (below).



Figure 26. Undamaged sternal rib end (left) and damaged sternal rib ends (center and right).

4.4 Insect Activity

Each pig was initially exposed to insect activity when the bag containing the individual pig was opened after arrival to the site. The insects collected on the first day were captured before placement of the pigs in the water. On successive visits, the pigs were removed from the water, and flies were captured while the pigs were on land. During the Fresh and Bloat stages of decomposition, trapped gasses within the remains allowed for significant bouyancy. Portions of the pigs remained above water, allowing for fly colonization. Upon entrance to the Active stage of decomposition, the pigs lost some (but not all) bouyancy. As a result, portions of the pigs remained above water, but splashing from waves and rolling of the pigs prevented consistent dry spots necessary for fly colonization.

4.4.1 Venice Site Insect Activity

Upon the arrival of VPIG1 and VPIG2 to the Venice site, flies began to appear. This fly activity was minimal, with one fly appearing every three minutes. This slow appearance of flies lasted for the entire first visit, with only eleven flies captured. As Figures 27 and 28 show, *Cochliomya macellaria* represented all but one fly, identified as *Chrysomya megacephala*.

As a result of scavenging by American alligator (*Alligator mississippiensis*) VPIG1 was missing after the visit on Day 1. Figure 28 shows that the flies captured during this second visitation represent *Lucilia sericata*, *Cochliomyia macellaria*, and *Chrysomya megacephala*. As a probable result of continued scavenging, VPIG2 was removed from its position on land at the Venice site after visitation by the researcher on Day 10.



Figure 27. Insects collected from VPIG1.



Figure 28. Insects collected from VPIG2.

4.4.2 Grande Isle/Grand Terre Site Insect Activity

Immediately after the arrival of GIPIG1 and GIPIG2 to the Grande Isle site, flies began to appear. This fly activity was minimal, with one fly appearing every four minutes. The five flies captured during the visitation on Day 1 were identified as *Cochliomyia macellaria*. Although flies were observed on both pigs, even flying from one pig to the other, flies were only captured

from GIPIG1. As Figures 29 and 30 show, *Cochliomyia macellaria* and *Chrysomya megacephala* were the only flies observed during the Fresh through Active stages at the site.

GIPIG1, which was transported to the land portion of the site on Grand Terre on Day 12, showed activity from flies of the family Piophilidae, first observed during the third visitation on Day 18. As the figure shows, the next visitation, occuring on Day 25, showed limited fly activity, consisting of only *Cochliomyia macellaria* and Piophilidae. The next visitation, occurring on Day 32, showed no insect activity.



Figure 29. Insects collected from GIPIG1.

Figure 30 shows the insects collected from GIPIG2, the pig that remained in the water at Grande Isle for the duration of the study. The third visitation, occuring on Day 12, was the first visitation in which this pig was not bouyant enough that a portion of the pig remained perpetually above the surface of the water. As such, insect data following the Day 8 visitation represent fly activity that began only once the pig was removed from the water. *Cochliomyia macellaria* and *Chrysomya megacephala* were the only flies captured from this pig.



Figure 30. Flies collected from GIPIG1.

4.4.3 Baton Rouge Site Insect Activity

Immediately after the arrival of BRPIG1 and BRPIG2 to the Baton Rouge site, flies began to appear. This site showed greater fly activity than Venice and Grande Isle, with approximately fifteen flies arriving every minute. As Figure 31 shows, flies catpured were identified as belonging to the species *Cochliomyia macellaria*, *Chrysomya megacephala*, and a single fly from the family Muscidae. After the third visit on Day 10, flies were captured from BRPIG1 every day. Insect activity for the early stages was predominantly represented by *C. megacephala*, *C. macellaria*, and *L. sericata*. After the entrance of BRPIG1 into the Advanced stage of decomposition, flies of the family Piophilidae began to visit the site. Beetles from the family Histeridae appeared beginning on Day 15. Starting on Day 15, a steady decline in insect activity was observed at the site. Day 21 marked the first day with no fly activity. As the figure shows, beginning on Day 24, a mass emersion of young flies from around the site of BRPIG1 from the soil began. Thousands of flies were observed drying their wings on the grasses and bushes surrounding the site. These flies, as the figure shows, were identified as *C. megacephala*, *C. macellaria*, and a single fly from the family Muscidae. This emersion represents the only

insect activity at the site past Day 21. Total number of emergent flies at the site began to decline after Day 25. On Day 27, no insect activity was recorded at the site.



Figure 31. Insects collected from BRPIG1.



Figure 32. Insects collected from BRPIG2.

BRPIG2 remained in the water after the removal of BRPIG1 on Day 10. As stated in the methodology for this project, the stage of decomposition of BRPIG2 would be noted on a daily basis, but collection of insects would occur every seven days. As a result of the shrinking of remains, BRPIG2 came loose from the rack and became stuck near shore. As seen in Figure 10, the soft tissues of the remains were stationary just below the surface of the water. This position did not allow for insect colonization. The remains were removed from the site by water currents on Day 17, one day before collection was scheduled to occur. As shown in Figure 32 below, the insect activity around BRPIG2 consisted of *Chrysomya megacephala*, *Chochliomya macellaria*, and *Lucilia sericata*, consistent with BRPIG1.

On Day 15 Red Imported Fire Ants (*Solenopsis invicta*) were observed removing maggots from BRPIG1 at the Baton Rouge site. The ants, seen in Figure 33 below, had not been seen at the site previously. The mound was not seen. Three maggots were observed being removed by the ants.



Figure 33. Red imported fire ants (Solenopsis invicta).

CHAPTER 5: DISCUSSION

Forensic scientists observe the effects of different variables on decomposition through repeated experiments. Variations in weather conditions, including temperature, can accelerate or retard decomposition. Until this experiment, no published article has simultaneously used research sites more than one hundred miles apart. This project shows that although average daily temperatures may vary within a few degree points, the succession of the stages of decomposition varies little as remains advance from Fresh to Bloat, and from Bloat to Active stage of decomposition. Each pig remained in the Fresh stage for less than twenty-four hours after deposition. Anderson (2004) published a timeline allowing zero to three days for the remains to exit the fresh stage. On Day 1, the average temperature at the Venice site was 26°C, the average temperature at the Grande Isle site was 26.5°C, and the average temperature at the Baton Rouge site was 24.5°C. This project shows that between 24.5°C and 26.5°C, remains in the Fresh stage of decomposition at the surface of water will enter the Bloat stage within twenty-four hours after deposition. Anderson (2004) shows that the Bloat stage in shallow water marine environments lasts from three to eleven days. The six pigs observed in this project follow this outline; the Bloat stage lasted six or seven days, with the Venice site pigs remaining in the Bloat stage for one day longer than the other four.

As has been noted by Anderson (2004) and Anderson (2009), stages of decomposition beyond the Bloat stage in aquatic environments become difficult to delineate. The beginning of the Active stage was clearly observed at the three sites; the skin of the abdomen ruptures, exposing the soft tissue, white dermis and adipose tissue. This opening allows the gasses to escape, reducing the buoyancy of the remains. These remains did continue to float to varying degrees well after loss of a majority of soft tissue.

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Advanced decomposition was seen only in one of the two pigs that finished decomposing on land. BRPIG1, the Baton Rouge site pig that was moved onto land on Day 10, entered the advanced stage on Day 15, as evident by the putrefactive liquid that began covering the remains and staining the ground beneath it. GIPIG2, the pig placed on land on Grand Terre, appeared to skip the Advanced stage completely. The pig desiccated within six days of replacement, leaving much of its soft tissue in a mummified state. Although the temperature of the Baton Rouge and Grand Isle/Grand Terre sites did not vary considerably during this time, temperature around the GIPIG2 may still have been higher. GIPIG2 was placed on an old road to decompose. That road consisted of hard-packed limestone and mud, with no cover from the sun. Although the temperature was taken upon every visit to the site, because the temperature was not taken near the pig by a recording device, temperature fluctuations may have gone unrecorded.

BRPIG2 and GIPIG1 were allowed to remain in the water to completely decompose. Their decomposition was markedly different; GIPIG1 lost most of its internal soft tissue as a result of scavenging. As a result, the axial skeleton, surrounded by a thick layer of adipose tissue and dermal skin, remained on the ribcage until the removal of the remains on Day 26. Repeated visitation by the researcher showed only a reduction in the overall size of the remains due to persistent scavenging, not an advance in the stage of decomposition. Conversely, BRPIG2 lost its skin and adipose tissue by Day 15. With the exception of a few small fish identified as *Gambusia sp.*, no aquatic scavenging was noted at the aquatic portion of the Baton Rouge site. This allowed the remains to retain much of the original soft tissue while showing signs of Advanced decomposition. This categorization is tenuous, however; the skull of both BRPIG2 and GIPIG1 disarticulated within one day of each other (Day 15 and Day 14, respectively) even though

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BRPIG1 was considered to be in Advanced decomposition while GIPIG1 remained in Active decomposition.

Scavenging at the three sites progressed very differently as each site hosted unique scavengers. The Venice site was visited by multiple alligators that completely removed pig remains. Although the anterior half of VPIG2, which contained the skull, forelimbs, chest cavity (missing most of the internal organs) and the skin of the left side was recovered, this portion was removed within twenty-four hours after placement on land. Observations made during this study show that alligator scavenging can leave marks on the bone of remains. As shown in Figure 10, two ribs were broken near their sternal ends. This is consistent with documented case studies of alligator damage to bone. Harding and Wolf (2006) show that long bones will be broken as alligators tear tissue from the main body of remains. The final thoracic vertebra and other exposed ribs did not show damage. As shown by this study, the twisting motion used by alligators to remove tissue from remains may not leave postmortem evidence on disarticulated bones. This means that if remains are only partially scavenged, then allowed to continue decomposition, evidence of the prior alligator scavenging may not be evident on the bone.

The alligator scavenging observed during this study began on Day 9, the same day that the remains entered the Active stage of decomposition. Informally, alligators have been said to wait "a week" with remains before beginning to scavenge them. This behavior, although potentially supported here, has not been discussed in the literature.

The Grande Isle portion of the Grande Isle/Grand Terre site was characterized by fish scavenging of GIPIG1 and GIPIG2. The fish that feed directly off of the remains did not cause significant trauma to the tissue; no bite marks were noted. Instead, the subsurface hair and skin

of the remains were slowly and uniformly removed, giving the submerged skin a white color. As the pigs lost buoyancy and began to roll within the water column, the entirety of the remains took on this white, scavenged look. Once the internal soft tissue had been removed on Day 16 the remains began gradual reduction in size over time. After termination of the study, bones of the water pig GIPIG1 do not show obvious scavenging trauma.

Indirect fish scavenging included the presence of an unusually large school of striped mullet. This behavior is undocumented in the literature. The fish were readily noticeable by their mouths surfacing from the water, the sucking sound the fish made as they fed which could be heard from shore and their consistent presence within two meters of the remains even after disturbance by the researcher. Because these fish were feeding on bits of tissue coming off of the remains, this behavior could be used to find sunken or hidden remains during rescue operations, mass disasters that involve coastal floods or storm surges, or when searching for intentionally submerged remains.

The Grand Terre portion of the sight showed evidence of possible vulture scavenging. To date, no study has investigated the possibility for vulture visitation or scavenging on a Louisiana barrier island. Presence of these birds on the island should be unlikely. Grand Terre is treeless and is too small to provide a stable food supply of dead animals for a population of scavenging birds. If it is shown that these birds can be present in this environment, forensic investigators would know to be mindful of the alteration of evidence when collecting remains.

The water portion of the Baton Rouge site was unique in its relative lack of scavengers. Soft tissues, as shown in Figure 10, should be the first tissues removed by scavengers, as was seen at the Grand Isle site. Instead, these tissues remained unscavenged. The slow reduction in the size of the remains seemed to be driven mainly by tidal action slowly removing pieces of tissue and bone through mechanical processes.

The land portion of the Baton Rouge site was dominated by a pack of coyotes that repeatedly visited the site. As noted previously, coyotes are commonly seen in the neighborhoods and wooded areas around Baton Rouge, and packs like the one recorded likely patrol the length of the riparian wetland from Baton Rouge to Greater New Orleans. These covotes quickly located and scavenged remains, accelerating decomposition. The two covotes in this study were able to drag remains which originally weighed 53 kilograms, attempting to pull it from the easily noticeable place near the river. These coyotes left telling evidence of their presence; a musk odor, coyote fecal matter, and dog-like foot prints were noted beginning early in the study. Scavenging began by removing the soft tissue on the abdomen and exposing the muscle tissue of the right thigh. This removal of tissue was identifiable by tooth marks and discoloration of freshly exposed tissues. Coyote scavenging continued into advanced decomposition, even on remains containing significant maggot masses. Although individual bones appear to be scattered randomly around the site, the main portion of the remains was consistently dragged toward the tree line. By Day 16 a coyote trail became visible with the direct line of the drag marks noted.

The disarticulation noted during the observations of BRPIG1 are partially consistent with the findings of Reeves (2009) and Haglund et al. (1989). The hind limbs and forelimbs were scavenged first, as well as the soft tissue of the abdominal cavity. Against the previous literature, the cranium remained articulated to the remains until the portion containing it and the forelimbs was removed from the site. The difference in findings between this study and the previously noted publications may lie in the thickness of the neck of pigs to that of humans.

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Flies were attracted to the six pigs with varying intensity depending on the site. The Baton Rouge site had a much higher concentration of flies around the pigs after deposition. The Venice site and Grande Isle/Grand Terre site, however, saw much slower colonization by flies. This discrepancy was seen through the duration of the study. Species of flies noted appeared to be relatively consistent between the sites. *L. sericata*, *C megacephala*, and *C. macellaria* were the dominant fly species for the Fresh, Bloat, and Active stages. Members of the family Piophilidae appeared during the later portion of the Advanced stage of BRPIG1 and GIPIG1 as overall numbers of the previous three species declined.

The large emergence of young flies, which occurred at the Baton Rouge site between Day 24 and Day 26 coincides with the peak of fly colonization at Day 14. During this emergence, PMI based on the bones present at the sight may have been difficult to estimate. The presence of the emerging flies, however, adheres closely to the established timeline for fly growth to adulthood as established by Gabre et al. (2005).

This project examined three geographically distinct sites in Louisiana to determine how decomposition can vary. Based on the temperature recordings, the three sites were similar throughout the study, with the recorded variation having little direct impact on decomposition. The intensity of insect activity at the sites differed, with the Baton Rouge site having more flies present at a given time than either the Venice site or the Grande Isle/Grand Terre site. Despite this difference, the fly species represented remained consistent. Scavenging at the three sites was markedly different. At the Venice site, American alligators completely removed the remains after the visitation on Day 10. At the Grande Isle site, striped mullet marked the location of the aquatic remains, while evidence suggests possible vulture visitation of the land remains on Grand Terre. At the Baton Rouge site almost no aquatic scavenging was noted. This left decomposing

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remains with much of the original soft tissue. Coyotes scavenged the Baton Rouge land remains, scattering bones eventually removing the main portion of the remains from the site. Further research could explore the potential visitation of Louisiana's two vulture species to the barrier islands in the Gulf of Mexico. Further observations of remains in environments with American alligators could show if the delay between deposition and scavenging reported here is a typical behavior.

CHAPTER 6: CONCLUSION

This project provides additional data pertaining to decomposition in southern Louisiana. The pigs observed show decomposition consistent between the three sites studied through the Active stage. For necrophagous insects, the data provided show insect appearances based on stage of decomposition, showing a relatively uniform succession between the sites. Animal scavenging varies greatly based on location, as shown. Further research could focus on the delays between deposition of remains and alligator scavenging. A measurable delay could provide an additional marker for the establishment of PMI.
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