SPATIAL AND TEMPORAL TRENDS OF BAT POPULATIONS AT DUNBAR CAVE STATE NATURAL AREA, MONTGOMERY COUNTY, TENNESSEE

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SPATIAL AND TEMPORAL TRENDS

OF BAT POPULATIONS

AT DUNBAR CAVE STATE NATURAL AREA,

MONTGOMERY COUNTY,

TENNESSEE

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To the College of Graduate Studies:

We are submitting a thesis written by Veronica Brook Mullen entitled "Temporal and Spatial Trends of Bat Populations at Dunbar Cave State Natural Area, Montgomery County, Tennessee." We have examined the final copy of this thesis for form and content. We recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science.

Andrew N. Barrass, Major Professor

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DEDICATION

This thesis is dedicated to my son Micah D. Logue, who was my motivation throughout this entire process.

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ABSTRACT

VERONICA BROOK MULLEN. Spatial and Temporal Trends of Bat Populations at Dunbar Cave State Natural Area, Montgomery County, Tennessee (Under the direction of ANDREW N. BARRASS).

Bats are of global conservation concern, mainly due to human associated disturbance and habitat destruction. Some bat species, once considered to be common, may become threatened or endangered in the near future. There is a lack of information regarding details of abundance, roost-site selection, site fidelity and seasonal cave use among local bat populations. The purpose of this study was to provide such details for the bat populations of Dunbar Cave. Specifically, to determine whether bat populations at Dunbar Cave have increased in abundance and diversity over time, as well as, to investigate cave chamber preference and spatial distribution of the local bat populations, within Dunbar Cave. Another goal of this study was to investigate additional bat species present in the areas surrounding Dunbar Cave through the use of acoustic monitoring. Dunbar Cave has been subjected to immeasurable amounts of human disturbance. In early 2010 the public was prohibited from entering Dunbar Cave, and since this time the bat populations may have begun to recover. Bats were captured at the cave entrance from May through August, and cave surveys were conducted throughout the year of 2011 and 2012. All captured individuals were banded and species, sex, age, and reproductive status were determined. A total of 473 bats were banded and four bat species were captured. At least ten additional species were detected through bio-acoustics. The tri-colored bat (*Perimyotis subflavus*) was the most prevalent species, and the majority of bats observed were adult males. Comparison of these results to those of previous studies

indicates that the bat population at this cave continues to increase. Bats are using Dunbar Cave year round. The highest in-cave occupation occurs during winter and spring, and three cave chambers are preferred. There is evidence for clumping or formation of population aggregations within at least three areas of the cave. Dunbar Cave serves as an important site for the local bat populations, and provides habitat for reproduction, hibernation, and colonization. Continued monitoring and additional research is needed at this site to reach more advanced conclusions regarding the social structure and roost preferences of the inhabiting bat populations.

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CHAPTER I

INTRODUCTION

Introduction and General Information

Bats are the second most speciose order of mammals, and arguably the most diverse (Glover & Altringham 2008). As the only mammals capable of flight, bats are able to disperse and traverse considerable distances. Bats occupy a wide geographical range and can be found on every continent but Antarctica. There are more than 1,100 recognized species of bat, comprising approximately 20 percent of all mammalian species (O'Shea et al. 2004). Bats provide several benefits to humans and ecosystems alike. Economically, bats serve important roles as pollinators, seed dispersers, and consumers of pest insects that may potentially damage a wide array of agricultural crops. A recent study predicted that the loss of bats in North America would lead to an annual agricultural loss of approximately $22.9 billion (Boyles et al. 2011). Regionally, that equates to between $4.8 and $6.6 million a year in agricultural cost (Boyles et al. 2011).

Locally, bat populations are threatened by many factors. Habitat destruction in the form of deforestation, as well as, damage to underground hibernacula and roosting sites through commercialism and vandalism greatly affect bat populations (O'Shea et al. 2003). Other threats to bat populations include the intake of pesticides through consumption of chemical laden insects, and wind turbine related mortality

(Boyles et al. 2011). In addition, cave-dwelling bats of the Eastern United States have recently suffered unprecedented mortality due to the emergent disease White Nose Syndrome (WNS) (Blehert et al. 2009; Boyles & Willis 2009; Frick et al. 2010).

White Nose Syndrome has been characterized as a condition of hibernating bats and was named after the white fungal growth observed on the muzzles, ears, and wing membranes of affected bats. White Nose Syndrome is caused by the cold-loving fungus, *Geomyces destructans*, (Gargas et al. 2009), which grows optimally between five and ten degrees Celsius, but can survive at temperatures of up to 20°C (Blehart et al. 2009). As a result, *G. destructans* thrives in bat hibernacula, such as mines and caves, which locally range in temperature between two and 14°C (Blehart et al. 2009). Once established, the hyphae of this fungus invade hair follicles and associated sweat and sebaceous glands, eventually breaching the basement membrane of the underlying tissue (Blehart et al. 2009; Gargas et al. 2009). Consequently, a cutaneous fungal infection occurs and produces physical irritation to bats in torpor, causing them to repeatedly arouse from hibernation in order to groom themselves. This increased activity during a normally inactive period leads to the premature depletion of fat reserves in affected individuals (Blehart et al. 2009).

Many endothermic animals enter prolonged bouts of decreased physiological activity, known as torpor, during periods of time when food may be scarce or unavailable. Torpor is characterized by a markedly reduced metabolic rate, as well as, a lowered body temperature (Wojciechowski et al. 2007). Insectivorous bats of temperate regions use both daily and hibernation torpor. Arousal from daily torpor occurs diurnally, while arousal during hibernation torpor is suppressed for prolonged periods of time (Willis & Brigham 2003; Wojciechowski et al. 2007). Hibernation torpor can last up to 80 days in many insectivorous bat species (Wojciechowski et al. 2007). During hibernation torpor, insectivorous bats spend 99% of their time in an inactive, lethargic state, while the remaining 1% is filled with bouts of temporary arousal to forage, drink water, or relocate within hibernacula (Boyles & Willis 2009). The energy used during these bouts requires a metabolic rate increase close to 400 times greater than that needed during torpor (Thomas et al. 1990). When infected with WNS, a bat arouses more frequently and for a longer time period, thereby depleting fat reserves at a quicker rate, and ultimately starving to death or dehydrating before the end of hibernation. White Nose Syndrome has also been known to cause scaring and necrosis of wing tissue in survivor bats. This may contribute to a decrease in foraging success during active summer months (Reichard & Kunz 2009).

White Nose Syndrome was first documented in 2006 in a cave in upstate New York (Blehart et al. 2009). Since this time, it has been documented in 19 states and four Canadian provinces (United States Geological Survey 2011). On average, approximately 70% mortality is observed among infected bat populations; however, in the most severely affected areas WNS has destroyed entire bat colonies (Gargas et al. 2009; Boyles et al. 2011). The United States Fish and Wildlife Service (2012) estimates that, since 2006, WNS has killed five to seven million bats in the eastern United States alone.

At least six species or subspecies of bats in the continental U.S. are listed as endangered (O'Shea et al. 2003; United States Fish and Wildlife Service 2011). The majority of these is either obligate cave-dwelling species, or are dependent upon caves for at least some portion of their lives (Harvey 1997; Briggler & Prather 2003). This has been true even before WNS was observed in the United States. It has been predicted, however, that due to the devastating effects of WNS at least one cave-dwelling bat species that was once considered common may become regionally extinct within the next thirty years (Frick et al. 2010). Additional species are being considered for federal listing as threatened or endangered species (United States Fish and Wildlife Service 2011). Because little is known regarding the spread of WNS, all nonessential human travel into caves and other known hibernacula has been banded on public lands at this time (Center for Biological Diversity, 2011).

There is growing concern regarding the status of bats in the United States. As a result, an interest in bat ecology and management aimed toward conservation of bat populations has re-emerged. Historically, bat management strategies were primarily observational based, and relied heavily on anecdotal evidence (Ellison et al. 2003; O'Shea et al. 2003). Bats possess certain natural history traits that make them very vulnerable to population declines. Many species give birth to only one young annually, and typically do not reach reproductive maturity until at least one year of age (Fenton 2003). Bats are the only mammal capable of flight, they are secretive and nocturnal, and therefore can be difficult organisms to study (Ellison et al. 2003; O'Shea & Bogan 2003). As a result, there remains a lack of basic behavioral, biological, and ecological information regarding details of roosting, foraging, and population demographics (Fenton 2003). Furthermore, little published information exists on abundance, site-selection, and seasonal site use by many bat species (Rabinowitz 1981; Sandel, et al. 2001;Briggler &Prather 2003).

Nevertheless, these animals are easily captured, taxonomically stable (i.e., easily identified to species with few recent changes), and fill a variety of ecological niches (Jones et al. 2009). In addition, bats are relatively long-lived, use an assortment of habitat types, and are sensitive to ecological change (Fenton 2003; Jones et al. 2009). All of these characteristics distinguish bats as excellent bio-indicator organisms. For example, Hickey, et al. (2001) used Vespertilionid bats as environmental indicators, by measuring the annual amounts of heavy metals accumulated in their fur. Additionally, because of their need for both a food source, as well as, a roost site within one area, bats can be used to assess the impact of habitat changes from urban to rural and wild situations (Sandel et al. 2001; Owen et al.2003).

Bats of the Southeastern United States are all insectivorous, and the majority of these belong to the family Vespertilionidae (Forsyth 1999), the most globally widespread and diverse family of bats (Graham 1994; Hester & Myers 2001). Vespertilionid bats generally display what is known as a fission-fusion reproductive strategy (Kerth 2008). Fission-fusion refers to the tendency that, depending on the season, these animals will either sexually segregate (fission), or form heterogeneous colonies (fusion). For example, during late spring and early summer, female Vespertilionid bats segregate into maternity colonies. Here gestation is completed and parturition takes place. The newly born young are nursed for roughly three to five weeks, or until self-feeding behavior is learned (Barbour & Davis 1969). Likewise, male bats segregate from the colony during summer and roost either singularly or among bachelor colonies (Barbour & Davis 1969; Tuttle 2006). By autumn, the young of the year, along with the reproductively active adults of both sexes, begin to congregate at the entrances to winter hibernacula.

This behavior is known as swarming, and was perhaps described best by Glover and Altringham (2008) who characterized swarming as, "*intense chasing flights in and around underground sites, by large, transient, multi-species bat assemblages*". Swarming generally occurs in autumn, but may take place anywhere from late July through November

(Rivers et al. 2006; Glover & Altringham 2008). The purpose of swarming is not entirely understood, however, there are currently two equally accepted hypotheses in regard to this event (Kerth et al. 2003; Parsons et al. 2003). One hypothesis suggests that the main function of swarming behavior is related to mating (Kerth et al. 2003; Parsons et al. 2003; Glover & Altringham 2008). Several species of bat attend swarming events, and individuals at these sites may vary from day to day (Parsons et al. 2003). As a result, swarming facilitates the maintenance of gene flow among bat populations (Kerth et al. 2003; Veith et al. 2004). It is also proposed that swarming serves as an opportunity for social learning in juvenile bats (Kerth et al. 2003; Parsons et al. 2003; Glover & Altringham 2008). During this time, the young of the year may be led to known winter hibernacula by adults, and are consequently given the opportunity to familiarize themselves with these areas (Kerth 2003).

As winter approaches, temperatures begin to drop, and insect populations become unavailable as food sources for insectivorous bat populations. Cave-dwelling bats of both sexes begin to secure roost sites within winter hibernacula and enter torpor. Some species roost singularly; scattered throughout cave chambers, crevices, or other underground sites, whereas others congregate into large aggregations. Copulation among Vespertilionid bats has been observed during both fall swarming, as well as, during hibernation (Cockrum 1955; Hill & Smith 1984). Sperm is stored by females over winter, and ovulation does not occur until arousal from torpor in the spring (Barbour &Davis 1969; Hill & Smith 1984). Fertilization takes place shortly after, and females begin to leave the hibernaculum. A male biased sex-ratio is generally observed among hibernating bat populations (Briggler & Prather, 2003), and has also been noted during the early months of swarming (Kerth et al. 2003). As the swarming season progresses, however, this ratio begins to approach the expected 1:1 ratio, as females arrive from maternity colonies (Cope & Humphrey 1977; Kerth et al. 2003).

Study Site

Research was conducted at Dunbar Cave State Natural Area in Clarksville, Montgomery County, Tennessee. This site is an 110 acre area located approximately one and a half miles northeast of downtown Clarksville. Dunbar Cave is the main feature of the park, and is one of the most prominent caves of the surrounding region (Tennessee Department of Environment and Conservation, 2011). Many community and recreational activities take place at DCSNA including hiking, fishing, picnicking, and viewing of wildlife.

Dunbar Cave was formed millions of years ago by the Red River cutting through limestone, which resulted in a lowering of the local water table (Tennessee Department of Environment and Conservation, 2011). As a result, water seeped through cracks and joints of the sinkhole plain above and down into the Red River water basin. This water was slightly acidic and over millions of years dissolved the limestone along its route creating the cave. The known passages and chambers of Dunbar Cave make up about eight miles and have been heavy explored (Matthews 2005). This particular cave, however, belongs to an extensive network of caves and sinkholes in the local vicinity, and it is likely that virgin passages and alternate entrances remain undiscovered (Matthews 2011; Tennessee Department of Environment and Conservation, 2011). Water from a variety of sources (i.e. the Red River, seepage from above ground, and underground springs) run throughout Dunbar Cave forming the River Styx, which exits as a cold, clear stream below the main entrance of the cave (Matthews 2005). This stream was impounded at some point, and back-water formed a 15 acre pond, known as Swan Lake (Figure 1).

Humans have been attracted to the constant stream flow and natural air conditioning of Dunbar Cave for thousands of years (Matthews 2005; Tennessee Department of Environment and Conservation 2011). Archeological evidence found near the entrance of Dunbar Cave dates back to as far as 10,000 years ago, and is believed to have originated from Paleo-Indian activity (Matthews 2005). In addition, petroglyphs have been found on several of the cave walls and are associated with the Mississippian culture (Simek et al. 2007). Consequently, Dunbar Cave has been recognized as an important archeological site (Simek et al. 2007).

Dunbar Cave State Natural Area also has a long history of commercial use (Matthews 2011). Many large events were held within the cave, and at times the inner chambers housed thousands of people. "*When* *brilliantly lighted with electricity through all of its caverns, byways, and magnificent halls, some three or four miles underground, it* (Dunbar Cave) *will furnish a day's entertainment and ample accommodations for 20,000 people*" (Matthews 2005). An electric lighting system was, indeed, installed in the cave and tours were given on a regular basis. Passageways and several central chambers were excavated on a large scale in order to hold these large groups of people. In 1948, then country music star Roy Acuff purchased the cave and surrounding property (Matthews 2005). A concession stand and amphitheater were constructed around the cave entrance, and concerts featuring well known country music artists regularly took place there (Figure 2).

Figure 1. Northeast view of the entrance to Dunbar Cave from Swan Lake.

Figure 2. Entrance to Dunbar Cave surrounded by the amphitheater and concession stand. Large events were once held at the entrance to Dunbar Cave. A dance floor was constructed within the natural amphitheater-like setting of the surrounding geology, and refreshments were served to the crowd.

By the early 1960's Dunbar Cave was no longer in its heyday, and, as a result, the property lay dormant for several years (Matthews 2005). During this period, vandals and amateur explorers frequented this site (Matthews 2005). Ammunition and other goods were stored there as part of a Civil Defense project during the Cold War (Matthews 2005). In the early 1970's, vandals broke into the cave and set fallout shelter supplies on fire (Matthews 2011). This fire burned for days. It has been reported that this event led to the death of thousands of roosting bats (Matthews 2011).

Anecdotal, historical and physical information implies that Dunbar Cave was once inhabited by thousands of bats (Matthews 2011). Although it is impossible to know exact numbers of species and individuals, it is evident that a large bat colony did occupy the cave at some point in the past. Some cave-dwelling bats form large, tightly-packed colonies during winter hibernation and maternity periods. As these bats roost they urinate and defecate in an upward direction. Such colonies generally number in the thousands, and so much waste is expelled from these animals that a large reddish-brown stain permanently remains in the area where they congregated (Barbour & Davis 1969). Large stains can be found on the ceiling of several chambers of Dunbar Cave.

Due to the regional history and proximity of the cave to the Red and Cumberland Rivers, it is generally believed that these stains were produced by a large colony of *Myotis grisescens* (Matthews 2005). *Myotis* *grisescens* prefer to form maternity colonies in large, high-ceilinged chambers of warmer caves, like Dunbar Cave (Hill & Smith 1984). Furthermore, *M. grisescens* maternity sites are more commonly formed in caves with standing, or softly flowing streams (Barbour & Davis 1969; Graham 1994). For these reasons, it is believed that Dunbar Cave was once home to a summer maternity colony of *M. grisescens*. This species is listed by the United States Fish and Wildlife Service (2011) as endangered.

Myotis grisescens was once one of the most abundant mammals within its range, but within the last fifty years these populations have been greatly reduced (Hill & Smith 1984). Because this species uses caves year-round, it is very vulnerable to habitat destruction. In addition, due to this species strict roost requirements, only 5% of remaining undisturbed caves are suitable for occupation (Graham 1994; Harvey et al. 1999). Currently, 95% of all hibernating *M. grisescens* populations are restricted to only eight caves within the eastern United States (Harvey et al. 1999) (Appendix A).

In 1973 Dunbar Cave and the surrounding property was purchased by the state of Tennessee and designated as a state natural area (Tennessee Department of Environment and Conservation 2011). Beginning in 1983, organized cave tours were offered by the park to attract the public and generate revenue for DCSNA, (Amy Wallace, DCSNA, Interpretative Specialist, August 10, 2011). The majority of these tours included groups of elementary school students, and, on average, was comprised of around 20 individuals (Amy Wallace, DCSNA, Interpretative Specialist, August 10, 2011).

Bat populations were not considered when determining cave tour schedules. Tours occurred throughout spring, summer, fall, and early winter, at a typical rate of at least ten tours a week (Amy Wallace, DCSNA, Interpretative Specialist, August 10, 2011). A study on the effects of commercial cave tours on bats found that there is, in fact, a positive correlation between the intensity of light and sound created by cave tours and bat activity (Mann et al. 2002). Human disturbance, vandalism, and commercialization of hibernacula, alter cave microclimates, and are principal factors in the decline of bat populations (Johnson et al. 1998).

Data do not exist in regard to bat populations at Dunbar Cave State Natural Area until late 2005 when Austin Peay State University began research here. At this time a "bat-friendly" cave gate had recently been installed. It is unknown whether bats had difficulty entering the cave prior to this installment. In addition, the species and number of individuals that may have been using Dunbar Cave prior to 2005 is unknown. Large colonies of bats that were reported as having once inhabited Dunbar Cave, were assumed to have been destroyed or abandoned this site, due to prolonged human disturbance. Since 2005 four species (*Eptesicus fuscus, Lasiurus borealis, Myotis lucifugus* and *Perimyotis subflavus*) have been documented either using Dunbar Cave or the surrounding areas. All of these are relatively common throughout the Southeastern United States and cover a wide geographic range (Appendices A & B).

In 2009, APSU research assistants were able to convince the park that public cave tours may have a negative impact on bat populations, especially during hibernation (Kurz 2011). As a result, beginning in 2009, the cave was closed to the public from November until March. In March of 2010, while conducting a routine cave survey, APSU research assistants discovered a little brown bat (*Myotis lucifugus*) infected with White Nose Syndrome inside the cave. Federal and state agencies were notified, and Dunbar Cave was closed to all human activities in order to reduce further spread of the *Geomyces destructans* fungus, as well as, to eliminate disturbance to an already compromised population. Since this time, only personnel affiliated with bat research and population monitoring have been allowed inside Dunbar Cave.

Purpose of this Study

The majority of bat research in the United States has focused on species legally classified as protected or "endangered" (Ellison et al. 2003). Monitoring programs for more common bat species are crucial for providing basic data, such as, habitat selection, landscape usage, and biological details. Such information is needed for conservation efforts (O'Shea et al. 2003).

The overall goal for this project was to compare abundance, species diversity, and other population demographics among the cave-dwelling bat populations of Dunbar Cave to existing data, in order to determine whether there have been significant changes in the population over time. More specifically, to investigate whether these populations had begun to recover from years of human disturbance and whether populations may have benefitted from the cessation of human recreational activities and closing of the cave to visitors since 2010. In addition, the surrounding landscape was surveyed acoustically to create baseline data for future studies focusing on species found using the areas around Dunbar Cave. Another major objective for this study was to determine if the individuals found within the cave were displaying preference in seasonal cave use, chamber selection, and roost-site selection within chambers.

Data collected during this study were added to Austin Peay State University and the Center of Excellence in Field Biology's data-base for the cave-dwelling bat populations at Dunbar Cave State Natural Area (DCSNA) as part of an ongoing research project. The maintenance of this data-base is essential to long-term monitoring by allowing APSU and the CFB to detect significant changes in bat population demographics over time. This information can then be used to assist state and federal agencies in cave restoration and future conservation efforts at this site.

Several general predictions were made based on review of existing data and scientific literature:

1. There will be an overall increase in abundance and species diversity among the bat population of Dunbar Cave over time.
2. There will be an increase in females among the swarming population, in comparison to previous years.
3. There will be cave chambers that are preferred by the bat population.
4. Bats will be observed aggregating in specific cave chambers.
5. Several bat species will be recorded using acoustic monitors that were previously not known to utilize this area.

CHAPTER II

METHODS AND MATERIALS

Introduction to Methods

The selection of methods used to study bat populations is dependent upon the target species, as well as, the season(s) that sampling is taking place. Bat activity at a particular site can vary dramatically, and the same assemblage of species may not be found at a given location from night to night (O'Farrell & Gannon 1999). Temperate bat species, in particular, display a wide range of deviation in behavioral patterns, both spatially and temporally and are difficult to observe in general (O'Shea et al. 2004; Weller & Lee 2007). Due to resource partitioning, variation in maneuverability, and differences in vocalizations between species or among individuals, not all bats are equally susceptible to any one particular assessment or capture method (Francis 1989; O'Farrell & Gannon 1999). Thus, each capture technique has inherent biases (Kuenzi & Morrison, 1998; O'Farrell & Gannon 1999). Therefore it is suggested that in order to collect the most representative sample, a combination of techniques be applied (O'Shea & Bogan, 2003; Flaquer et al. 2007).

A considerable amount of the knowledge of the biology and behavior of bats has been obtained through capture of individuals at roost-sites, water holes, and along foraging fly-ways through the use of mist-nets or harp traps (Francis 1998; O'Farrell & Gannon 1999). Although Kunz and Kurta (1988) described mist-nets as "the most commonly used devices for capturing flying bats", harp traps are light-weight and more easily erected on site (O'Farrell & Gannon 1999; Francis 1998). Moreover, when considering both the number and diversity of species captured, harp traps are found to be ten times more efficient than mist-nets, especially when targeting Vespertilionid bat species (Kunz & Kurta 1988; Francis 1989). Harp traps are also considered to be less stressful on captured bats (Flaquer et al. 2007). In a few instances, mist-nets have been found to be more successful at capturing larger bat species, which, in some cases, are able to use momentum to escape being captured in harp traps (Francis 1989). Nevertheless, both devices sample a very small area relative to the area that is used by free-flying and foraging bats. Bats are often able to avoid being captured after consecutive nights of netting and frequently become "net shy" (Kuenzi & Morrison 1998; Kunz & Kurta, 1988). Often the use of such devices results in sampling only a small portion of the Chiropteran fauna at a particular site, and some species may be completely missed (Francis 1989). For example, Francis (1989) noted that smaller species with lower frequency calls were netted more often than similar sized species with higher frequency calls. In addition, many forest-dwelling bat species fly at a height that requires several tiers of mist-netting (Kunz & Kurta 1998). Such materials are expensive and become difficult to monitor and maintain (Kunz 2003).

Recently, acoustic monitoring of bats, through the use of ultrasonic detectors, has been heavily incorporated into bat research. Insectivorous bats rely on echolocation to detect and capture prey, to sense items, and obstacles in their paths, as well as, to socialize or share information (Fenton 1988; Kunz 2003). In fact, bats have been considered one of the most vocal groups of animals (Fenton 1988). Individual bats or species produce distinct vocalizations and acoustic devices allow biologist to record and visualize these ultrasonic calls. Acoustic monitoring has become an especially useful technique since the emergence of White Nose Syndrome. This is mainly due to the fact that ultrasonic recording devices are nonintrusive and facilitate a more hands off approach to bat identification (O'Farrell & Gannon 1999).

First of all, ultrasonic microphones and recording devices, (i.e. acoustic bat detectors) are not detectable by bat populations and therefore can be used repeatedly at a site (Kuenzi & Morrison 1998). In addition, these devices may be used in areas where it is impossible to place netting and other capture devices (Kuenzi & Morrison 1998; Kunz & Kurta 2003). Ultimately, acoustic bat detectors permit sampling of bat populations at a larger area than nets or traps (Kuenzi & Morrison, 1998, Kunz and Kurta, 2003). The ability to identify bat calls to species allows a more complete inventory of bat population assemblages present within an area than by netting or other capture methods alone O'Farrell & Gannon 1999). Recording and analyzing bat vocalizations may also provide information on bat behavior and ecology (Fenton 1988).

It is suggested that capture techniques and acoustic monitoring be used in combination to obtain the most representative data (Kuenzi & Morrison 1998; Fenton 2003; Kunz & Kurta 2003; Flaguer et al. 2007). Therefore, a combination of four sampling techniques were used in this study, including; harp trapping, mist-netting, acoustic monitoring, and cave surveys. Additional approval from United States Fish and Wildlife Service (USFWS) was required to continue cave surveys at Dunbar Cave State Natural Area - a White Nose Syndrome positive cave. Permit ((TWRA #3070) (TDEC #2011-019)) restrictions were applied and the number of capture and survey events per season were limited. This study began in February of 2011, and was completed in October of 2012.

Federal and state agencies permitted seven cave surveys and four harp trapping events during the first year of research. During the second year of research, eight cave surveys were permitted, as well as, eight harp trappings. Banding of bats was allowed during two cave surveys each year, as well as, during all harp trappings. Acoustic monitoring and mist-netting had no permit limitations.

Cave Surveys

Cave surveys began mid-morning (e.g., 9:00 – 10:00 am) on each of the selected dates, and were generally completed within three to four hours. Detailed field notes were recorded during each cave survey. In order to prevent site contamination and the possible spread of White Nose Syndrome (WNS), researchers wore Tyvek® suits during cave surveys. Additional cave gear was used to ensure all participants safety, and National WNS Decontamination Protocol was strictly followed (United States Fish and Wildlife Service 2012b). A total of 15 cave chambers were examined during each cave survey (Appendix C). Historically, six cave chambers had been included as part of the State Natural Areas interpretive cave tour route (Amy Wallace, DCSNA, Interpretative Specialist, personal communication, August 10, 2011). Other cave chambers were not surveyed due to inaccessibility, safety concerns (e.g., rock slides or water traps), or consistent absence of bats. In an attempt to reduce disturbance to hibernating and day roosting bats, surveys were mostly observational.

Cave chambers were thoroughly searched for bats and the total number of individuals per cave chamber was documented. Bats were identified to species and roosting locations were recorded, by measuring the left or right distance of each individual from the center point of the chamber, as well as, roost height from the cave floor up the chamber wall. These measurements were then used to create a micro scale GIS map using the Arc Map 10, Geographic Information Systems software, and ultimately for spatial analysis.

During the cave surveys in which banding was permitted, roosting bats were removed from the cave wall by hand. These banding surveys took place on February 21st and April 1st of 2011, and March 30th and April 27th of 2012. Only bats within reach were removed from roost sites and any bats covered in condensation were left untouched. The presence of condensation typically indicates that a bat may be in a state of torpor (Harvey et al. 1999). Arousal may cause unnecessary energy expenditure in these individuals. Researchers wore leather gloves to prevent injury from bat bites, as well as, a pair of fresh latex or nitrile gloves over the leather gloves. Latex gloves were changed after the handling of each individual bat in order to prevent the spread of WNS and other potentially communicable diseases. Once in hand, each bat was sexed, aged, banded, and photographed using a Canon Rebel XS camera with a Canon EFS 18-55mm lens. Out of reach bats were also photographed with a Canon Rebel XS camera and a Canon EFS 75-300mm zoom lens.

Temperature was recorded at several sites within the cave. Originally, temperature was monitored with maximum-minimum mercury thermometers, and was recorded during each cave survey within the Twilight Zone of the Entrance chamber of Dunbar Cave, the stairway leading to the Counterfeiter's chamber, and the midway point of the Lots of Bats chambers. In October 2011 HOBO® data-loggers (Onset Computer Corporation, Bourne, MA, USA) became available, and were placed in various locations throughout the cave for continuous collection of cave chamber temperature data. A data-logger was kept in the Twilight Zone of the Entrance chamber, in the River Styx, and at the midway point of the Lots of Bats chamber from October 2011 until October 2012. Two additional data-loggers became available in February of 2012. From February of 2012 until May of 2012 one of these units was placed in the Hallway chamber and the other was placed in the back of the Spray Hall chamber. In June of 2012 these units were brought back into Dunbar Cave and placed in the Counterfeiter's chamber and in the Junction Room chamber. These areas were chosen because either existing data implied that the bat population frequented the specific chamber, or because the area was suspected to have a higher rate of temperature fluctuation based on proximity to the above ground environment or inflow of water.

For example, the Entrance chamber was chosen because it was suspected to have the greatest influx of ambient air and therefore the greatest influx in temperature. The River Styx was chosen, because it flows throughout many areas of Dunbar Cave and may influence overall cave temperature. The area between the two Lots of Bats chambers was chosen, because the highest number of individuals were consistently seen using this area of the cave, both historically and during this study (Appendix G). These three units remained in the same locations for one year, and were only removed for retrieval of data, and battery replacement. Two additional units became available in March of 2011, and were moved among four cave chambers between cave surveys. These chambers were chosen because bats were observed using these areas, and temperature data did not exist for these chambers.

Harp trapping and Mist-netting

From May through August of 2011 and 2012 an eight foot Cave-catcher 36"x44" G5 bat harp trap (Bat Conservation and Management, Inc. Carlisle, PA, USA) was placed in front of the cave entrance a total of twelve times in order to capture bats entering and emerging from Dunbar Cave. This particular harp trap was composed of a rectangular, double-frame of aluminum tubing approximately 2.4m high by 1.8m wide, supported by tripods on both sides. Low visibility Stren® fishing line (3.6kg test) was strung vertically from the frame, and approximately 2.5cm apart from each other. The harp trap was set up in front of the cave gate approximately thirty to forty-five minutes prior to sunset, and was normally taken down between midnight and two a.m. A 12'x20' utility tarp was cut into two sections and secured with duct tape to the cave walls in order to block off the areas of the cave entrance that were not covered by the harp trap. As bats emerged or entered the cave they were unlikely to detect the lines of the harp trap. As a result, bats would fly into these wires and then fall into a polyethylene bag suspended at the bottom of the trap where they were promptly removed and banded.

During 2011, mist-nets were also deployed in an attempt to survey additionally areas of Dunbar Cave State Natural Area. Due to the concern for the spread of White Nose Syndrome, state and federal agencies did not allow the use of mist-nets at the cave entrance or within the amphitheater area surrounding the cave entrance. Therefore, two mist-nets were set up over corridors and flyways. These sites consisted mostly of trails surrounding Swan Lake or within forested areas of the park. Throughout the duration of the study there were a total of ten mist-netting events.

Both double high and single high mist-nets were used during each mist-netting attempt. The double high system consisted of five interlocking aluminum poles, (36" long X 0.75" diameter, heavy duty 0.125 wall thickness) (Bat Conservation and Management, Inc. Carlisle, PA, USA), supporting a double panel Hot Foot Mist-net 40 (25'x10' each) (Hot Foot America, Hayward, CA, USA) on both sides. Holes were drilled into the top and bottom of six of these poles in order to attach a pulley system to both sides of the net. This allowed easy retrieval of captured individuals. The single high system was also controlled by a pulley system, however, a single 10'x40' Hot Foot Mist-net 40 (Hot Foot America, Hayward, CA, USA) was used. This net was erected using two; 1.5" wide, PVC pipes that were stabilized by iron umbrella stands. Mist-nets were not used in 2012, due to the lack of success with this method during the previous year.

Acoustic Monitoring

Acoustic monitoring was also conducted during harp trapping and mist-netting events. This was done in order to determine whether additional bat species were present, but may not have been represented in the captured sample. This data was also used to further build Austin Peay State University and the Center for Excellence in Field Biology's acoustic library. Two Anabat® bat detectors (unit numbers 80665 and 80685) (Titley Electronics, Perth, Australia) were used during each netting and trapping survey. Both acoustic devices were equipped with an Anabat® High Mount microphone (Titley Electronica, Perth, Australia). During harp trappings, both Anabat® units were placed so that the microphone was positioned at a 45° angle. One unit was placed on top of the old concession stand adjacent to the cave entrance, pointing out into the amphitheater area of the cave. The other unit was placed on the railing of the amphitheater facing Swan Lake. Both Anabat® units were also used during each mist-netting survey. Generally, both units were set at equal distances between the two nets. All acoustic data was analyzed with either Echo Class software version 1.0 (ERDC, Army Corps of Engineers, USA) or Bat Call Identification 2010 (Bat Call Identification, Inc. Springfield, MO, USA).

Animal Handling Procedures

Prior to the capture and handling of bats an Animal Use Protocol Permit (#11.007R) was obtained from Austin Peay State University and the Institutional Animal Care and Use Committee. Researchers designated to handle bats were all previously vaccinated against the rabies virus. The standardized procedures of Lamb & Wyckoff (2010) were followed, which meant that, at a minimum, species, sex, reproductive condition, age, and Wing Damage Index (WDI) were recorded for each individual captured.

Male reproductive condition was determined by presence of enlarged testicles, which signified that a male bat was reproductively active (Hester & Grenier 2005). From July through August female bats were examined for the presence of visible nipples. If nipples were not easily located the bat was deemed as non-reproductive (Hester & Grenier 2005, Lamb & Wyckoff 2010). If nipples were visually obvious on the female bat, they were then palpated to determine lactation status. If milk was expressed from the nipple upon palpation the bat was considered lactating, if not she was considered to be in a post-lactation state or a non-reproductive state (Hester & Grenier 2005).

Both age and Wing Damage Index were determined by stretching the wing over a white light. To determine age (adult vs. juvenile) the phalanges were examined. Phalanges of a juvenile bat possess an obvious growth plate between the first and second phalange, which will appear as a clear bulbous section when illuminated by the white light (Hester & Grenier 2005). Because White Nose Syndrome often establishes itself on the flight-membranes of bats, all captured bats were assessed and a Wing Damage Index value was assigned for each individual. This index ranges from a scale of zero to three with zero being minimal to no damage, and three signifying severe damage such as loss of flight membrane (Reichard & Kunz 2009).

To assess wing damage, all bats were simply held above a light while the wings were examined. The wing membranes of bats consist of two layers of epithelial tissue separated by a thin layer of underlying connective, muscular, and nervous tissues, as well as, blood and lymphatic vessels (Cryan et al. 2010). The wing membrane of a healthy bat appears supple and flexible. In many WNS affected individuals, however, the wing membranes have lost these characteristics (Reichard & Kunz 2009; Cryan et al. 2010). As a result of fungal invasion, the wing membranes of WNS affected bats may lose tone, strength, and elasticity, causing them to tear easily (Cryan et al. 2010). In addition, these weakened membranes often adhere to each other and may resemble crumpled tissue paper (Cryan, et al. 2010). As *G. destructans* invades the epidermis it may digest the underlying tissues and leave behind large areas of pallor, or irregular pigmentation (Cryan et al. 2010). During capture events, it was attempted to photograph wing membranes of all captured individuals. This was especially important for investigation of changes in WCI of recaptured individuals over time.

A split-metal aluminum alloy bat ring, 2.9mm narrow, (Porzana Ltd., East Sussex, UK) was placed on the forearm of each captured bat with the opening facing posteriorly, or overlying the patagia (wing membrane). Male bats were banded on the right forearm while females were banded on the left. This aided in identification of individuals during cave (hibernation) surveys. If a banded bat was seen roosting in the cave, but was still in torpor, or inaccessible, researchers were still able to sex the bat based on band location. Each band had an Austin Peay State University and the Center of Excellence for Field Biology initials imprint, as well as an F or an M to designate sex followed by a four digit number. Generally, bats were handled for less than ten minutes, and were not kept more than 30 minutes for processing in compliance with standard protocols (Lamb & Wyckoff 2010).

Data Analysis

Descriptive statistics were used to obtain overall population numbers and for preliminary data exploration for cave surveys, as well as, spring and summer capture events. Cave survey data was then grouped for both years into spring, summer, fall, and winter categories.

Spring surveys were designated as those taking place within the months of March, April, and May, summer included those surveys within June, July, and August, fall surveys fell within September, October, and November, and winter surveys were considered to be those that took place within the months of December, January, and February. Bar graphs of abundance per season, cave chamber usage, and sex ratios of banded bats per chamber were created using Microsoft Excel 2010. These figures were used to detect any major seasonal cave usage shifts within the bat population by season, between the two years of this study. Descriptive statistics were used to summarize temperature data collected from within cave chambers. All analyses of this type were undertaken through the use of Microsoft Excel 2010.

Species diversity for spring and summer capture events was determined by investigating population heterogeneity of the Dunbar bat community. Species diversity during swarming was of particular interest, because, during this time, a variety of species may congregate at the entrances of hibernacula at the same time (Glover & Altringham 2008). Because population heterogeneity involves two types of information ---species richness and evenness--- these two components were measured separately (Krebs 1999). For purposes of this study, species richness was defined simply as the number of species in the community (Krebs 1999). Species evenness was considered to be the relative contribution of each species to the total number of individuals (Mulder et al. 2004).

The reciprocal of Simpson's diversity index was used to investigate species diversity, which is recommended as the appropriate estimator for a finite population when field data include counts of individuals (Krebs 1999). The reciprocal of Simpson's diversity index was defined as:

$$1/\mathrm{D} = 1/\Sigma\ pi^2$$

Where 1/D = Simpson's reciprocal index, and *pi* = the proportion of species *i* in the community.

Simpson's Measure of Evenness was used to calculate species evenness, and this was defined as:

$$\mathrm{E}_{1/\mathrm{D}} = (1/\check{\mathrm{D}})/s$$

Where $\mathrm{E}_{1/\mathrm{D}}$ = Simpson's measure of evenness, Ď= Simpson's diversity index, and s = number of species in the sample. Both indices were calculated using Ecological Methodology statistical software, version 7.2 (Exeter Software, Setauket, New York, USA).

The sex-ratio of bats captured during harp trapping events were also evaluated using the Chi-square Goodness of Fit Test. Although all statistics for these analyses were performed using JMP 9 statistical software (SAS Institute, Inc. Cary, North Carolina, USA), the general Chi-square statistic was assumed as:

$$X^2 = \Sigma \text{ (observed-expected)}^2/\text{expected}$$

The results of this test were then evaluated to determine seasonal difference in cave use between sexes. In addition, these results were compared to historical data to determine whether the overall sex-ratio has fluctuated over time.

Banding of bats at Dunbar Cave began in 2009 (Kurz 2011). This data was useful for investigation of site-fidelity. In addition, by compiling banding data from 2009 – 2012 a mark-recapture study was incorporated into this project. All banding surveys, beginning in May of 2009 until August of 2012, were compiled and analyzed using Ecological Methodology statistical software, version 7.2 (Exeter Software, Setauket, New York, USA).

The Jolly-Seber Method was used to evaluate bat population abundance at Dunbar Cave (Krebs 1999). This particular model is designed for open populations. Open populations are those that are constantly changing in size, due to a variety of factors, including; birth, death, immigration, and emigration (Krebs 1999) The Jolly-Seber model is also designed for studies composed of more than three mark-recapture events (Krebs 1999). Furthermore, the Jolly-Seber Method is designed for samples of short duration separated by long durations of time (Krebs 1999). The time interval between samples need not be constant, and any number of samples can be accommodated (Krebs 1999).

The Cormack–Jolly-Seber (CJS) maximum likelihood model has been suggested to be the most applicable mark-recapture estimator for population parameters of bat populations (Kunz 2003; McCracken 2003).Because the Jolly-Seber method is designed for open populations; there is no assumption of absence of recruitment or mortality, as in many earlier methods. This model, however does assume the following (Krebs 1999):

1. Every individual has the same probability of being captured
2. Every marked individual has the same probability of surviving
3. Individuals do not lose their marks, and marks are not overlooked
4. Sampling time is negligible in relation to intervals between samples

The data obtained for this method were:

m_t = number of marked individuals in sample t

u_t= number of unmarked individuals in sample t

n_t = Total number of animals caught in sample $t s_t$ Total number of animals released after sample t (= n_t – number of accidental deaths or removals

m_{rt} = Number of marked animals caught in sample t last caught in sample r

R_t = Number of the s_t individuals released at sample t and caught again in some later sample

Z_t = Number of individuals marked before sample t , not caught in sample t, but caught in some sample after sample t

From these variables, an estimate of N (size of population at time of marking) is obtained. In addition, proportion of the population marked, probability of survival, and number of individuals joining were estimated by this model.

During cave surveys, when an individual bat was encountered, roost-site details were recorded, including; specific cave chamber, height from chamber floor, and direction and distance from the center of the cave chamber. The individual roost site for each bat encountered on each cave survey was entered into ArcGIS 10.0 (ESRI, Redlands, California, USA). A map was created for each survey using a general base map of the Dunbar Cave chambers surveyed (Appendix D). In order to determine whether specific cave chambers were being utilized more frequently by the residing bat populations a comparison of means was performed using JMP 9 statistical software (SAS Institute, Inc. Cary, North Carolina, USA). A pair-wise comparison was used to further determine differences between cave chamber usages.

Three chambers (The Spray Hall chamber, the dry Lots of Bats chamber, and the 79-10 chamber) were chosen, because prior research of this nature had been conducted at this site, and measurements for these chambers already existed. X, Y, and Z coordinates were recorded for each individual observed during each cave survey within these three chambers. X coordinates measured the length of the chamber, Y coordinates measured the width of the chamber, and Z coordinates measured the depth of the chamber. These measurements then created a three dimensional cell in which statistical habitat analysis could be applied (Buckland 1993).

The standardized Morisita Index of Dispersion was used to determine whether individuals were clustering within Dunbar Cave. Two cave surveys were chosen from both 2011 and 2012, and were used to calculate separate indices for each of the three chambers. All analysis was completed using Ecological Methodology, statistical software, version 7.2 (Exeter Software, Setauket, New York, USA).

This index was based on the standardized formula:

$$I_p = n \, [\, \Sigma x^2 - \Sigma x / (\Sigma x)^{2} - \Sigma x]$$

Where I_p = Morisita's index of dispersion, n = sample size, Σx = sum of the quadrat counts, and Σx^2 = sum of quadrat counts squared.

CHAPTER III

RESULTS

Harp Trapping

Twelve complete harp trapping events (and one incomplete) were conducted throughout the study. A total of 443 bats were captured. One hundred and thirteen bats were captured during the four harp trapping events of 2011, and 330 bats were captured during the eight harp trapping events of 2012. Thirty four of the 443 total captures were recaptured individuals. Therefore a total of 409 individuals were banded during harp trappings (Table 1). Individuals M0074, M0069, M0247, and M0370 were recaptured on more than one occasion (Table 2). Individuals M0148, F0099, M0143, and M0011 were originally banded in 2009 (Table 2). All other recaptured bats were originally banded at some point within the two years (2011-2012) of this study.

Banding of bats at Dunbar Cave State Natural Area was initiated in 2009, and there were few recaptures during the initial surveys (Kurz 2011). Therefore, abundance could not be estimated by entering each individual capture event into the Jolly-Seber population model (Krebs 1999; Kurz 2011). As a result, data from 2009 and 2010 were pooled, and this time period was considered to be the "initial capture event". The capture events that took place during this study (2011-2012) were pooled by season to avoid gaps in recapture data (Hargrove &Borland 1994; Krebs 1999). Total abundance (N) was estimated as 398.7+/-199.0 bats (Table 3). Probability of survival was estimated as 0.605+/- 0.500 and the estimate of recruitment or number of individuals joining was 273.6+/-222.2 (Table 4).

Table 1. Total number of bats captured, banded, and recaptured during harp trapping at DCSNA

Date	Number of Individuals Banded	Number of Recaptured Individuals	Total
May 31, 2011	6	0	6
June 28, 2011	19	0	19
July 27, 2011	57	4	61
August 11, 2011	26	1	27
May 18, 2012	14	2	16
May 30, 2012	4	2	6
June 14, 2012	3	0	3
June 29, 2012	12	3	15
July 11, 2012	34	3	37
July 26, 2012*	4	2	6
July 30, 2012	28	3	31
August 8, 2012	61	10	71
August 23, 2012	141	4	145
Total	**409**	**34**	**443**

*Indicates an incomplete survey

Table 2. Recaptured bats from harp trapping at DCSNA

Date Banded	Date Recaptured	Initial Age	Sex	Band Identification	Initial WCI	Recapture WCI
June 28, 2011	July 27, 2011	J	M	M0074*	0	0
February 21, 2011	July 27, 2011	A	M	M0202	0	0
May 31, 2011	July 27, 2011	J	M	M0069*	0	0
August 12, 2009	July 27, 2011	J	M	M0148	0	0
July 27, 2011	August 11, 2011	J	M	M0247*	0	0
June 2, 2009	May 18, 2012	J	F	F0099	0	0
July 27, 2011	May 18, 2012	J	M	M0247*	0	0
July 27, 2011	May 30, 2012	J	M	M0247*	0	0
May 31, 2011	May 30, 2012	A	M	M0080	0	1
July 27, 2011	June 29, 2012	J	M	M0232	0	0
July 27, 2011	June 29, 2012	J	M	M0247*	0	0
June 28, 2011	June 29, 2012	A	M	M0061	0	0
July 27, 2011	July 11, 2012	J	M	M0059	0	1
June 28, 2011	July 11, 2012	A	M	M0074*	0	1
March 30, 2012	July 11, 2012	A	M	M0394	0	1
June 29, 2012	July 26, 2012	A	M	M0322	0	0
August 11, 2011	July 26, 2012	J	M	_0037	0	0
July 11, 2012	July 30, 2012	A	M	M0336	0	0
May 30, 2012	July 30, 2012	A	M	M0314	0	0
May 31, 2011	July 30, 2012	J	M	M0069*	0	1
February 21, 2011	August 8, 2012	A	M	000790(TWRA)	0	0
July 11, 2012	August 8, 2012	A	M	M0401	0	0
July 11, 2012	August 8, 2012	A	M	M0345	0	0
July 11, 2012	August 8, 2012	A	M	M0385	0	0
April 27, 2012	August 8, 2012	A	F	F0158	0	0

August 12, 2009	August 8, 2012	J	M	M0143	0	0
April 27, 2012	August 8, 2012	A	M	M0355	0	0
June 28, 2011	August 8, 2012	J	M	M0070	0	0
July 11, 2012	August 8, 2012	A	M	M0348	0	0
April 27, 2011	August 8, 2012	A	M	M0370*	0	0
April 27, 2011	August 23, 2012	A	M	M0370*	0	0
July 30, 2012	August 23, 2012	A	M	M0409		0
August 12, 2009	August 23, 2012	A	M	M0011	0	0
August 8, 2012	August 23, 2012	A	F	F0173	0	0

N=34

* indicates multiple recaptures of individuals _ indicates a dremeled band identification number (All male bands were used upon completion of the 2011 capture events. Sex identifiers were dremeled from remaining female bands and used as "unisex" bands until new bands were received.

WCI = Wing Condition Index

Individual M0069 = *Eptesicus fuscus*

Individual 000790 = *Myotis septentrionalis*

All other individuals = *Perimyotis subflavus*

Wildlife biologist from Tennessee Wildlife Resources Agency accompanied APSU/CEFB researchers during the February 21, 2011 cave survey. Six bats were banded with TWRA identification bands, at this time.

Table 3. Total number of bats captured per pooled time period (n), proportion of population marked per capture event (α), and estimated total abundance (N) by Jolly-Seber stochastic population model.

Period of Capture	n	α	N	Std. Error of N
1 (2009-2010)	121	---	---	---
2 (May/June 2011)	25	0.038	193.1	574.7
3 (July/August 2011)	88	0.045	1304.4	1008
4 (May 2012)	22	0.217	300.5	255.3
5 (June 2012)	18	0.211	224.8	149.2
6 (July 2012)	73	0.095	398.7	199
7 (August 2012)	216	0.046	---	---

Table 4. Rate of survival (φ) and number of individuals joining population (B) estimated by Jolly-Seber stochastic population model

Period of Capture	**Φ**	**Std. Error of φ**	**B**	**Std.Error of B**
1 (2009-2010)	---	---	---	---
2 (May/June 2011)	0.061	0.051	955.2	1356.5
3 (July/August 2011)	1.808	0	-292.8	455
4 (May 2012)	0.455	0.353	54.1	141.4
5 (June 2012)	0.568	0.536	262.7	205.1
6 (July 2012)	0.605	0.500	273.6	222.2
7 (August 2012)	---	---	---	---

Based on total number of individuals captured per harp trapping event, data indicated, that more bats were captured during 2012 than 2011 (113 vs. 330; Figure 3). The data could not be assessed in this manner, however, because sampling periods were uneven. Therefore, mean number of bats were compared between years in order to determine if there was in fact a difference in abundance. The residual data from harp trapping events conducted during this study were normally distributed (Shapiro-Wilk Goodness of Fit, $p = 0.6795$, $\alpha = 0.05$), but due to small sample sizes, variance was unequal between years (Levene's test of equal variance, $p = <0.001^*$, $\alpha = 0.05$). Therefore, the nonparametric Kruskal-Wallis rank sum analysis was used to determine that there was no statistically significant difference between mean number of bats captured ($p = 0.8852$, $\alpha = 0.05$, $df = 1$) for both years of this study (2011-2012).

Data collected during 2009-2010 (Kurz 2011) consisted of a total of 121 bats that were captured as a result of seven harp trapping events. Again, due to an uneven number of capture events between previous studies and this study, it was difficult to determine whether or not there was a true difference between sampling events and numbers of bats captured. An analysis of means was used to determine if there had been an overall increase in abundance from 2009 to the present. Data, again, were not normally distributed (Shapiro-Wilk Goodness of Fit, $p = 0.0011^*$, $\alpha = 0.05$). The Kruskal-Wallis rank sum analysis ($p = 0.4532$, $\alpha = 0.05$, $df = 3$) determined that there was not a statistically significant difference between the number of bats captured from 2009 to 2012.

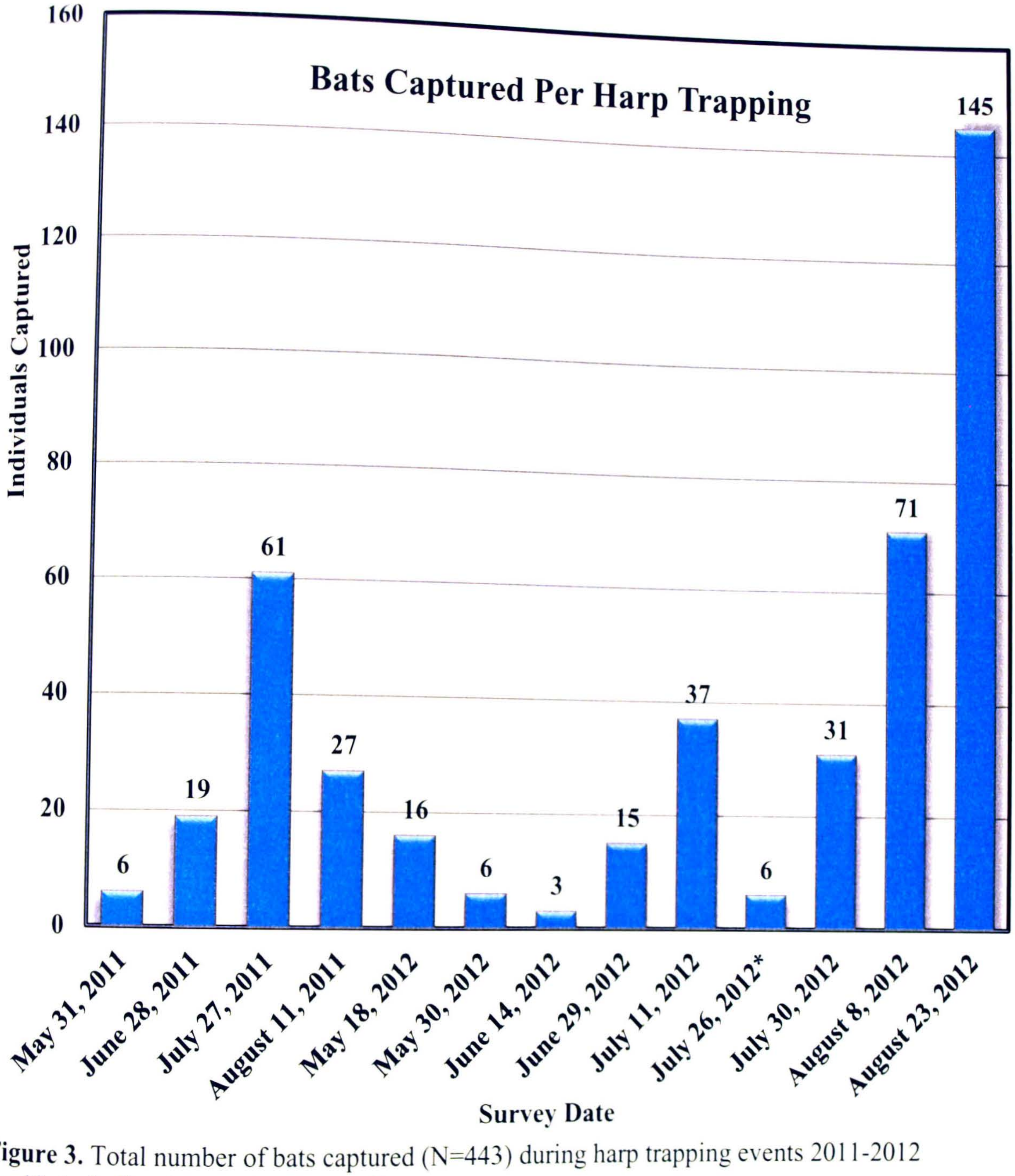


Figure 3. Total number of bats captured (N=443) during harp trapping events 2011-2012 (n=13, including incomplete survey*).

A total of four bat species were captured during harp trapping events at Dunbar Cave. The majority (N=425; 95.94%) were *Perimyotis subflavus* (Table 5). The other three species captured (*E. fuscus* (2.93%), *M. septentrionalis* (0.90%) and *M. lucifugus* (0.23%)) were observed in low numbers (Table 5). Simpson's index of diversity was calculated as 0.079. This index ranges from 0 (low diversity) to almost 1 (Krebs 1999). The reciprocal of Simpson's diversity was determined as, $1/D = 1.085$. The reciprocal of Simpson's diversity ranges from 1 to s, s= the number of species in the sample (Krebs 1999). In this form Simpson's index of diversity can be interpreted as the number of equally common species required to generate the observed heterogeneity of the sample (Krebs 1999). Simpson's measure of species evenness, which ranges from 0 (low value) to 1, was calculated as 0.271. Both indices were relatively low due to the predominance of one species of bat.

Table 5. Number of bats by species captured at DCSNA during harp trapping events

Species	**Year**		**Total**
	2011	**2012**	
Perimyotis subflavus	102	323	425
Eptesicus fuscus	9	4	13
Myotis septentrionalis	2	2	4
Myotis lucifugus	0	1	1
Total	113	330	**443**

Harp trapping events, n=13

Three hundred and fifty five male bats and 86 female bats were captured during this study (Table 6). Ninety four male bats and 17 female bats were captured as a result of the four harp trapping events of 2011. During 2012, 261 male bats and 69 female bats were captured. Few to no females were captured during May and June of both years (Table 6). Because of this, the Chi-Square Goodness of Fit test was not appropriate for analysis of these data. Instead Fisher's Exact Test was used, and the observed male to female ratio (355:86) did not fit the expected 1:1 ratio ($p = 0.001$, $\alpha = 0.05$, $df = 7$). When considering only those harp trapping events in which bats of both sexes were captured, sample size assumptions were met. Still, Chi-square Goodness of Fit analysis concluded that there was a lack of fit between the observed (156:43) and expected (1:1) male to female ratio ($p = <0.001$, $\alpha = 0.05$, $df = 4$).

Overall, the male to female ratio was approximately four male bats to every one female bat. No female bats were captured in May and June of 2011 (Figure 4). In 2012 only three females were captured during the four harp trapping events that took place in May and June (Figure 4). During July and August of both years, female bats begin to appear in the population (Figure 4). During August of both years the male to female sex ratios were the closest to fitting the 1:1 ratio (Figure 4).

The mean number of female bats captured during harp trapping from 2009-2010 (Kurz 2011) were compared to the mean number of female bats captured during harp trapping events from this study. Because there was a low number of females captured over the past four years (N=118), and because these data were counts, all data were transformed using the square root transformation. The residual data were normally distributed (Shapiro-Wilk Goodness of Fit, p=0.2965, α=0.05), and variances between the samples were equal (Levene's test of equal variance, p=0.0730, α=0.05).

To determine whether there had been a change in abundance of female bats among the sample population a one-way Analysis of Variance (ANOVA) was used. There was no significant difference found between the mean number of female bats captured between years (F=1.3403, df = 3, p=0.3280, α=0.05, R^2 Adj.=0.0849) (Table 7). A one-way ANOVA was also used to compare mean number of female bats captured during harp trapping between the years of this study (2011vs. 2012). No statistical significance was found (F=2.6918, df=1, p=0.1618, α=0.05, R^2 Adj.=0.2199).

Table 6. Sex ratios of bats captured during harp trapping at DCSNA

Date	Males	Females	M:F ratio (relative to 1)
May 31, 2011	6	0	6:0
June 28, 2011	19	0	19:0
July 27, 2011	54	5	10.8:1
August 11, 2011	15	12	1.25:1
May 18, 2012	13	3	4.3:1
May 30, 2012	6	0	6:0
June 14, 2012	3	0	3:0
June 29, 2012	15	0	15:0
July 11, 2012	36	1	36:1
July 26, 2012*	6	0	6:0
July 30, 2012	26	5	5.2:1
August 8, 2012	59	12	4.9:1
August 23, 2012	97	48	2.0:1
Total	**355**	**86**	**~4.1:1**

Table 7. Mean female bats captured during all harp trapping at DCSNA

Level	n	Mean	Std Error	Lower 95%	Upper 95%
2009	3	5.23448641	0.7169009	0.1124932	17.9809922
2010	2	7.06661206	1.075369	0.071289	25.4984602
2011	4	3.48953608	0.5377289	0.0313998	12.6657692
2012	3	16.3333181	0.7169009	4.363921	35.9268372

Data used for one-way ANOVA between years 2009 through 2012.

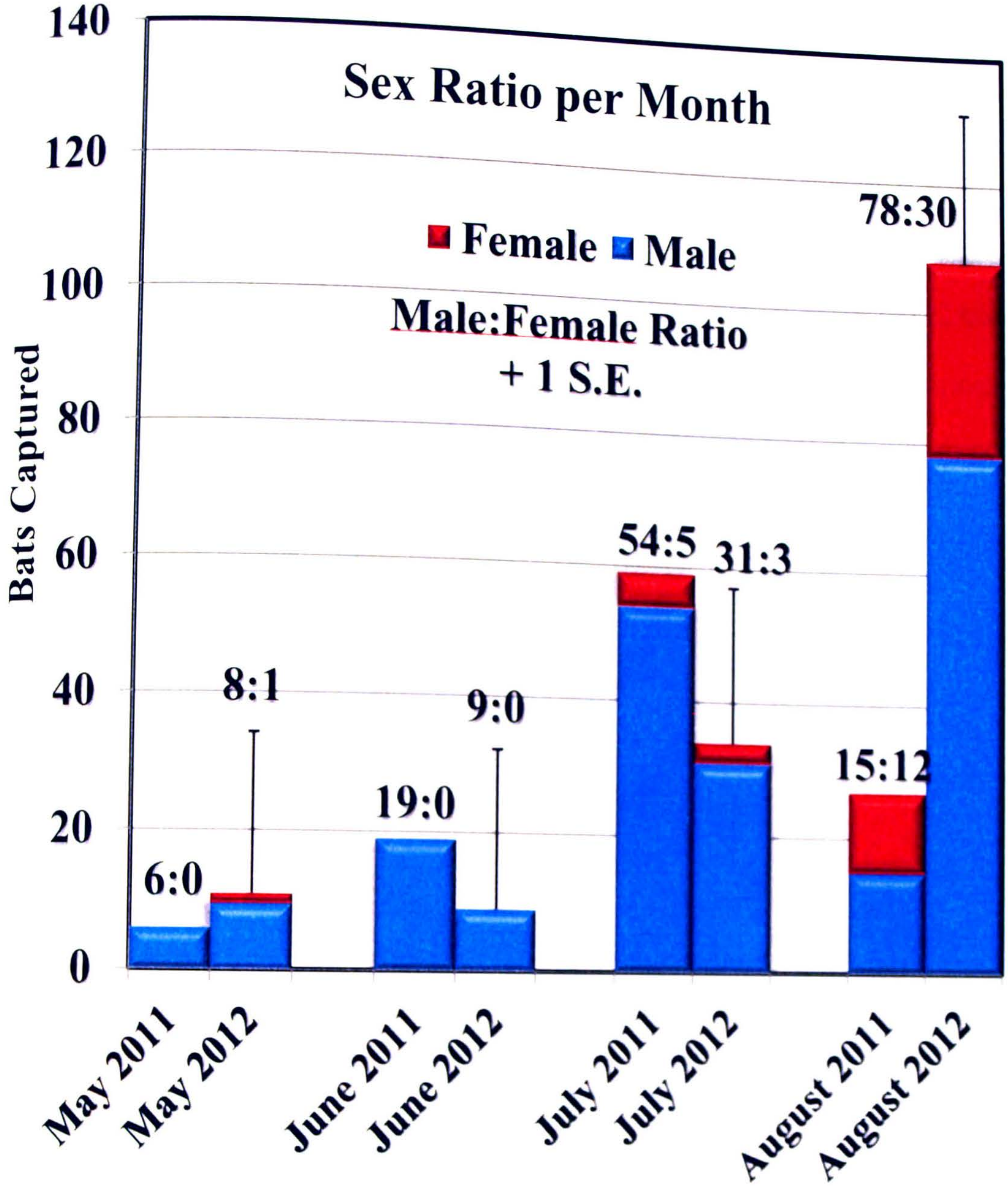


Figure 4. Mean male to female sex ratios of all bats captured during harp trapping events (n=4, 2011, n=8, 2012).

Three hundred and forty two adults and 100 juveniles were captured as a result of the 12 harp trapping events at Dunbar Cave throughout the course of this study (Table 8). In 2011 the adult to juvenile ratio was 1:7 with a total of fourteen adult bats and 98 juvenile bats captured during the four harp trapping events (Table 8). In 2012, however, a total of 328 adult bats and only two juvenile bats were captured as a result of 12 harp trapping events (Table 8). The overall ratio of adults to juveniles for 2012 harp trapping events was 164:1 (Figure 5). The average adult to juvenile ratio for both years of bats captured over the course of this study was 3:1 (Table 8).

Table 8. Adult to juvenile ratios of bats captured during harp trapping at DCSNA

Date	Adult	Juvenile	A:J (relative to 1)
May 31, 2011	4	2	2:1
June 28, 2011	0	19	19:0
July 27, 2011	3	58	0.05:1
August 11, 2011	7	19	0.37:1
May 18, 2012	16	0	16:0
May 30, 2012	6	0	6:0
June 14, 2012	3	0	3:0
June 29, 2012	15	0	15:0
July 11, 2012	37	0	37:0
July 26, 2012*	6	0	6:0
July 30, 2012	31	0	31:0
August 8, 2012	70	1	70:1
August 23, 2012	144	1	144:1
Total	**342**	**100**	**~3:1**

Capture events = n=13, including incomplete capture events*

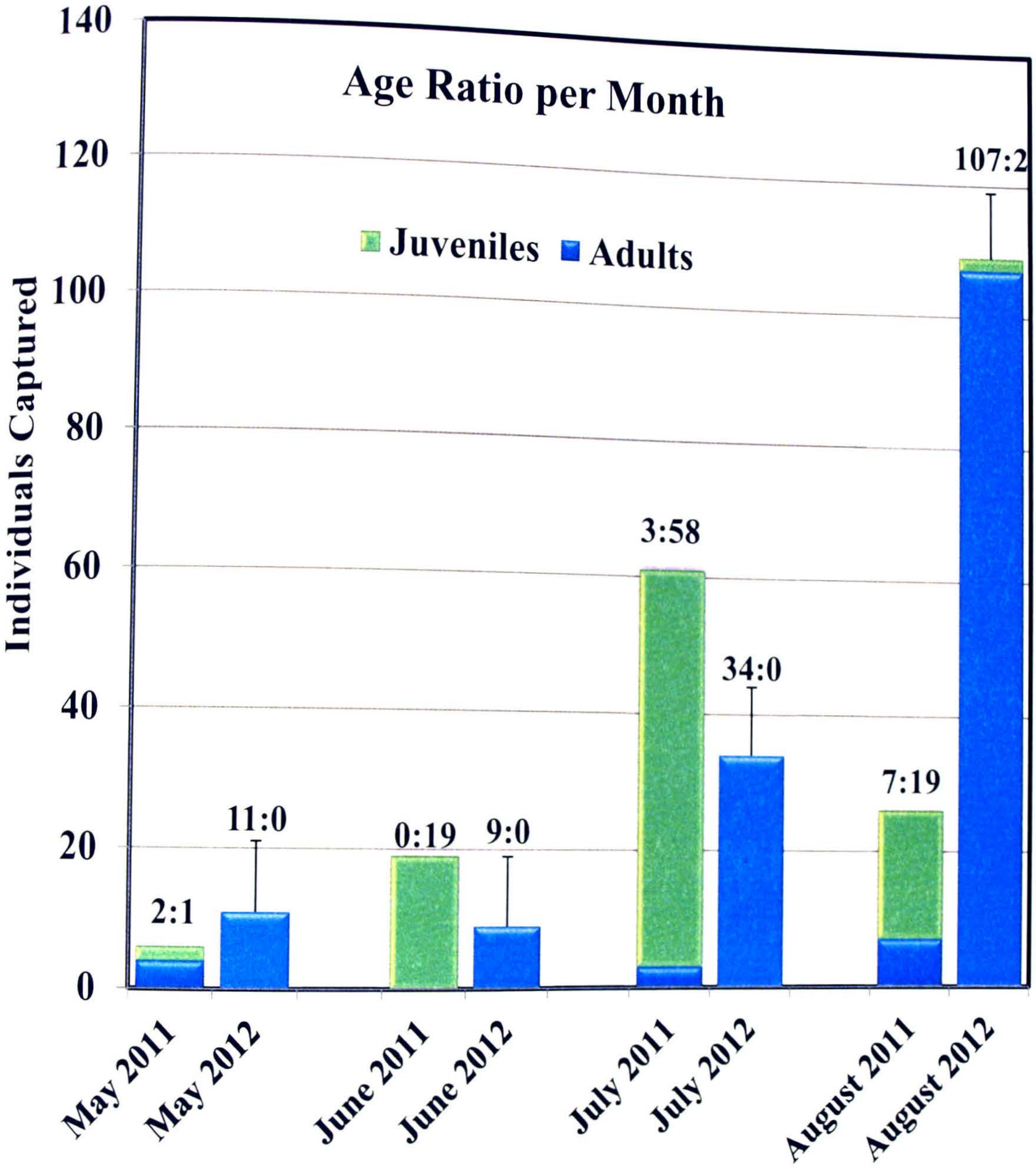


Figure 5. Average adult to juvenile age class ratios of all bats captured during harp trapping events (n=4, 2011, n=8, 2012).

Rate of recapture for each harp trapping event varied between months (Figure 8). Overall, approximately 8.3% of all individuals within the sample population were recaptures (34 recaptured individuals to 409 non-banded individuals). Thirty one of the recaptured individuals were *Perimyotis subflavus* (Table 2). Individual TWRA 00079 was an adult male *Myotis septentrionalis* that had been originally banded during the February 21, 2011 cave survey, and was recaptured in August of 2012 (Table 2). Individual M0069 was an adult male *Eptesicus fuscus,* originally banded in May of 2011 as a juvenile (Table 2). This individual was recaptured during July of both 2011 and 2012 (Table 2). Only three recaptured individuals were female, and none of these bats were recaptured multiple times during harp trappings (Table 2). Approximately 41.2% (14 of 34) of all recaptured individuals were originally banded as juveniles (Table 2).

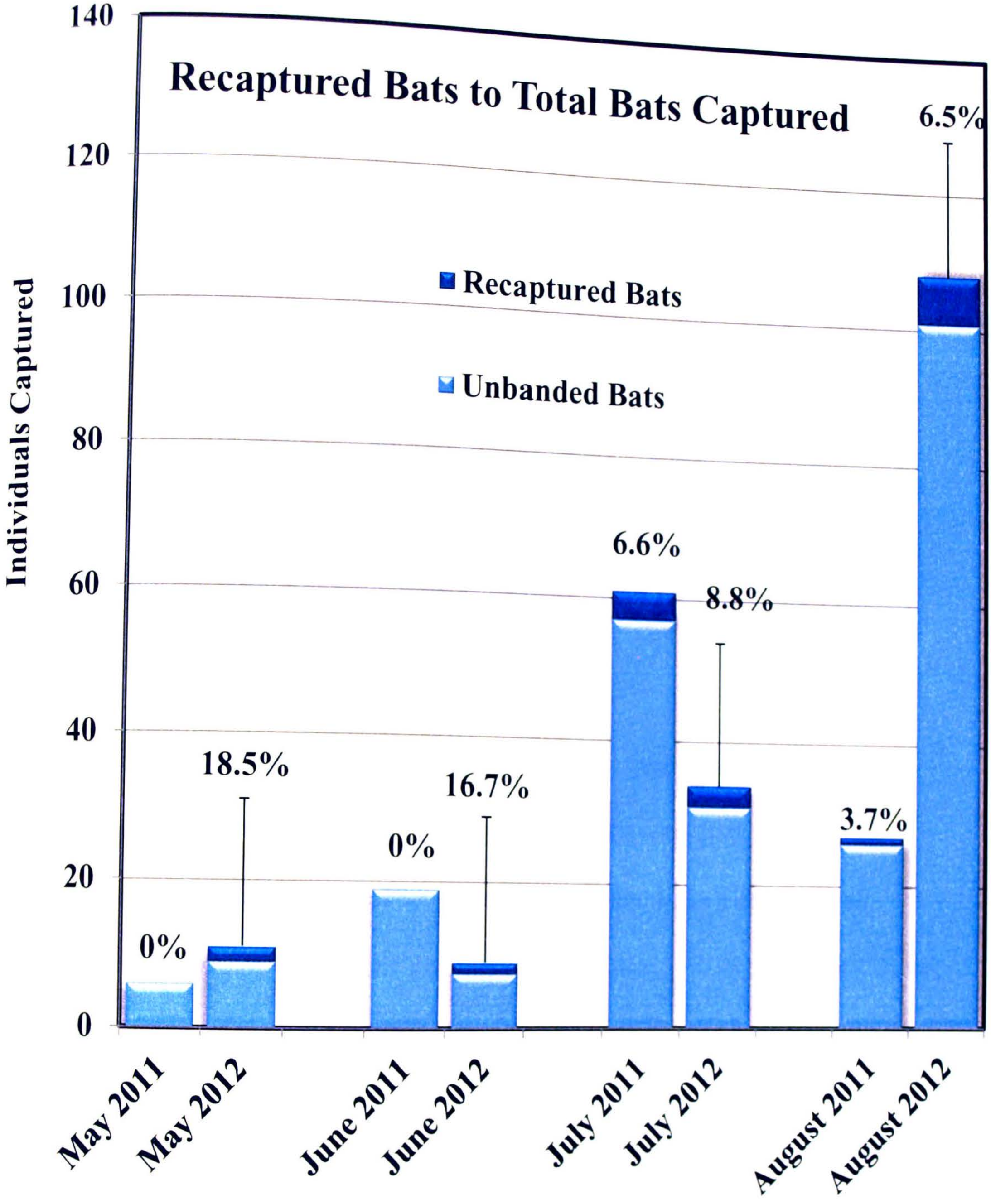


Figure 6. Percent of recaptured individuals (N=34) to total number of captured individuals (N=409) per month during harp trapping at DCSNA (2011-2012).

Cave Surveys

A total of 16 (15 complete and one incomplete) cave surveys were completed throughout the duration of this study (Table 9). Originally, seasons were defined as winter (December, January and February), spring (March, April and May), summer (June, July and August), and autumn (September, October and November). This was adjusted, however, because three surveys occurred late within the original seasonal category, and, therefore, were grouped into the following season. Specifically, the August cave surveys for both years occurred toward the end of the month, and therefore were grouped into the mean number of bats observed during autumn. This was also the case for the May cave survey of 2012, which was grouped into the mean number of bats observed during summer.

Bats were observed within the cave throughout the year; however, cave use appeared highest during winter and spring (Figure 7). Data were normally distributed (Shapiro-Wilk Goodness of Fit, $p=0.1076$, $\alpha=0.05$), and there was no difference in variance between the samples (Levene's test of equal variance, $p=0.3022$, $\alpha=0.05$). A one way ANOVA was used to find that there was a difference among mean number of bats observed within the cave per season ($F=19.3509$, $df = 3$, $p=0.0001$*, $\alpha=0.05$, R^2 Adj.=0.7973).

The highest in-cave occupancy was observed during early April of both years (Table 9). Tukey-Kramer HSD was used for pair-wise comparisons between seasons. Cave occupation was significantly higher in winter and spring compared to summer and autumn (Table 10). There was no significant difference, however, between mean number of bats observed among winter and spring seasons (Tukey-Kramer HSD, $p=0.2862$,

$\alpha=0.05$) or among summer and autumn seasons (Tukey-Kramer HSD, $p=0.8534$, $\alpha=0.05$

(Table 10).

Table 9. Number of bats observed, number of banded bats, and number of un-banded bats per cave survey at DCSNA.

Date	Previously banded	Un-banded	Total
February 21, 2011	8	52	60
April 1, 2011	7	63	70
May 12, 2011	2	44	46
June 8, 2011	0	2	2
August 23, 2011	1	9	10
October 18, 2011	3	13	16
December 8, 2011	3	27	30
January 10, 2012*	2	14	16
January 27, 2012	5	46	51
March 30, 2012	8	45	53
April 12, 2012	4	74	78
April 27, 2012	6	46	52
May 23, 2012	2	20	22
June 21, 2012	0	1	1
August 28, 2012	1	8	9
October 23, 2012	2	24	26
n=16			

* indicates an incomplete survey

Mean # total individuals observed = 33.9

Mean # previously banded observed = 3.4

Mean # un-banded observed = 30.5

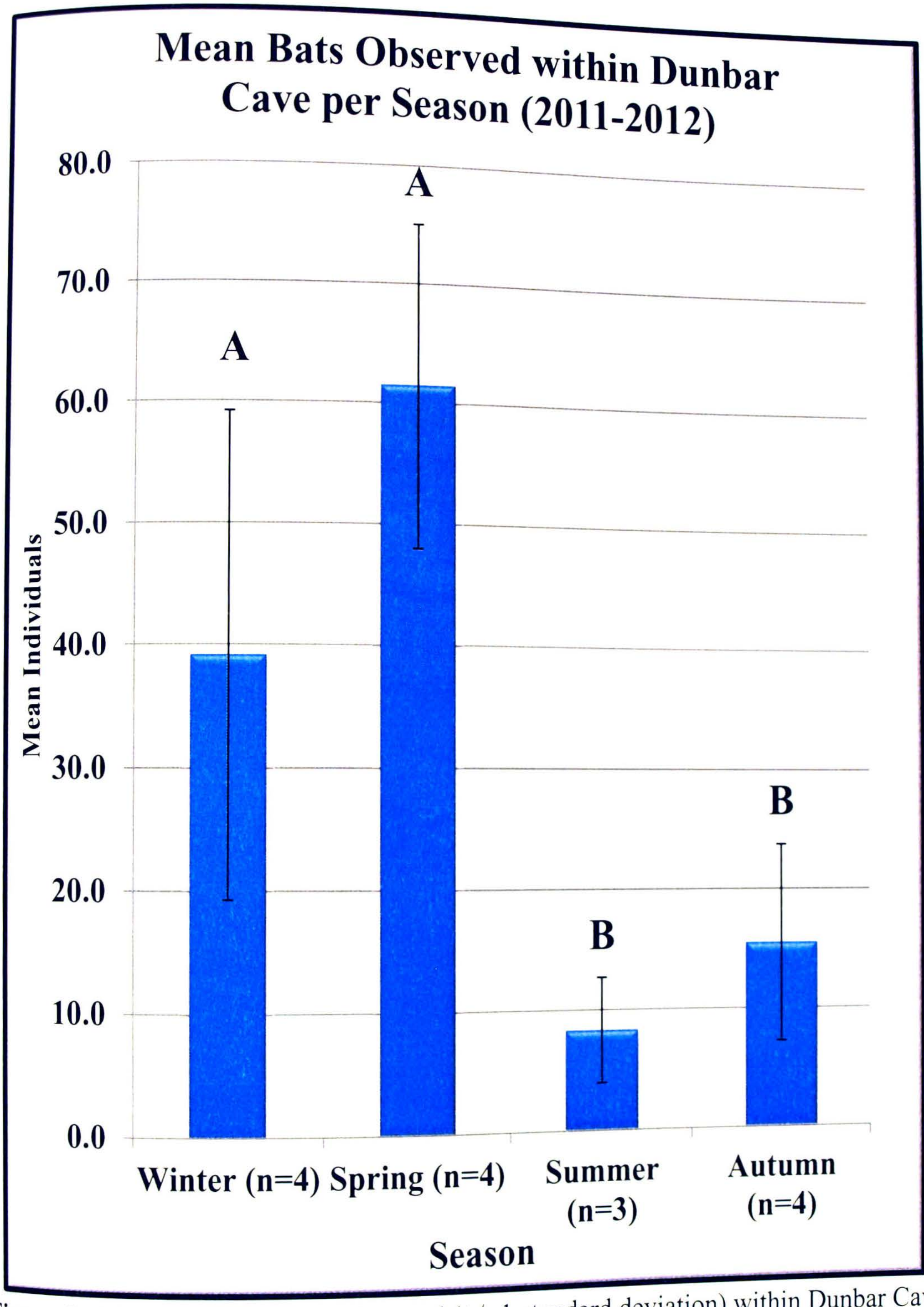


Figure 7. Mean number of bats observed (+/- 1 standard deviation) within Dunbar Cave per season, throughout the study. Means sharing letters did not show a statistically significant difference (Tukey-Kramer, HSD).

Table 10. Pair-wise comparisons between seasonal mean numbers of bats observed within Dunbar Cave chambers

Level	- Level	Difference	Std. Err Difference	Lower CL	Upper CL	p-Value
Spring	Summer	55.4	8.67445	29.2942	81.5058	0.0003*
Spring	Autumn	48.15	7.96799	24.1703	72.1297	0.0004*
Winter	Summer	39	9.69833	9.8128	68.1872	0.0093*
Winter	Autumn	31.75	9.07196	4.4479	59.0521	0.0220*
Spring	Winter	16.4	8.67445	-9.7058	42.5058	0.2862
Autumn	Summer	7.25	9.07196	-20.052	34.5521	0.8534

*Indicates significant results (95% confidence intervals) for Tukey-Kramer HSD

To determine whether there was a preferred cave chamber among the bat population, the mean number of bats observed within each chamber was compared. Overall, the data were not normally distributed (Shapiro-Wilk Goodness of Fit, $p=0.0498$, $\alpha=0.05$), and variance was not equal between groups (Levene's test of equal variance, $p=<0.001$, $\alpha=0.05$). A Kruskal-Wallis rank sum analysis was used to determine that there was no statistically significant difference between mean number of bats observed among various cave chambers ($p=0.1467$, $\alpha=0.05$, $df=14$). Three chambers (The Entrance, and both wet and dry Lot of Bats (LOB) chambers) did stand out, however, upon visual investigation, as consistently housing the highest number of individuals, throughout the duration of this study (Figure 8).

Data on seasonal cave chamber usage was not appropriate for statistical analysis. This was mainly because data consisted of counts of individuals per chamber, and many of these chambers were unoccupied throughout all or most of the year. Therefore, these data were assessed visually. The Entrance chamber, as well as, both wet and dry Lots of Bats chambers appeared to consistently house the highest mean number of individuals throughout all seasons (Figure 9).

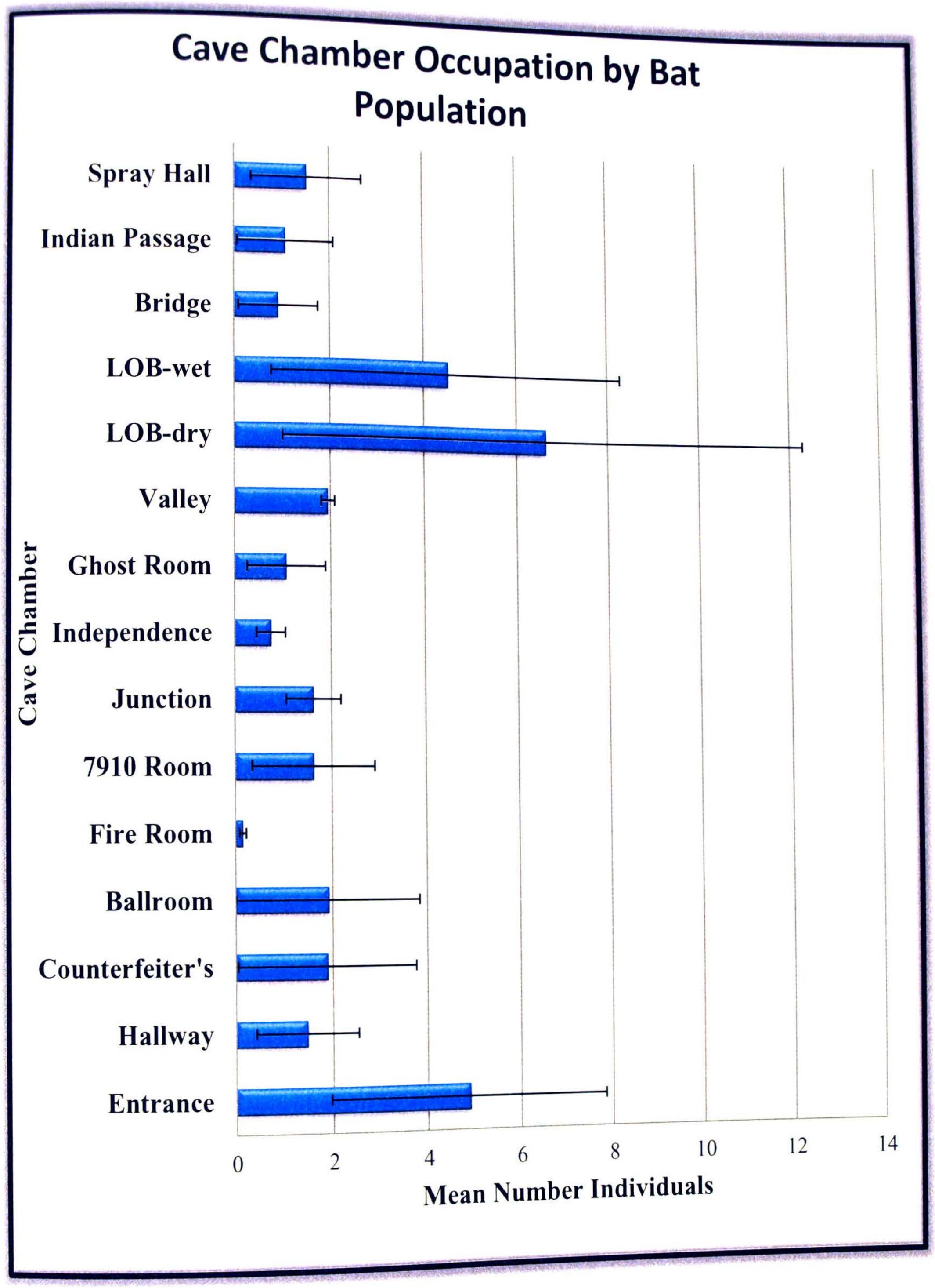


Figure 8. Mean (+/- 1 standard deviation) number of bats observed per cave chamber during all cave surveys (n=16) over the duration of this study.

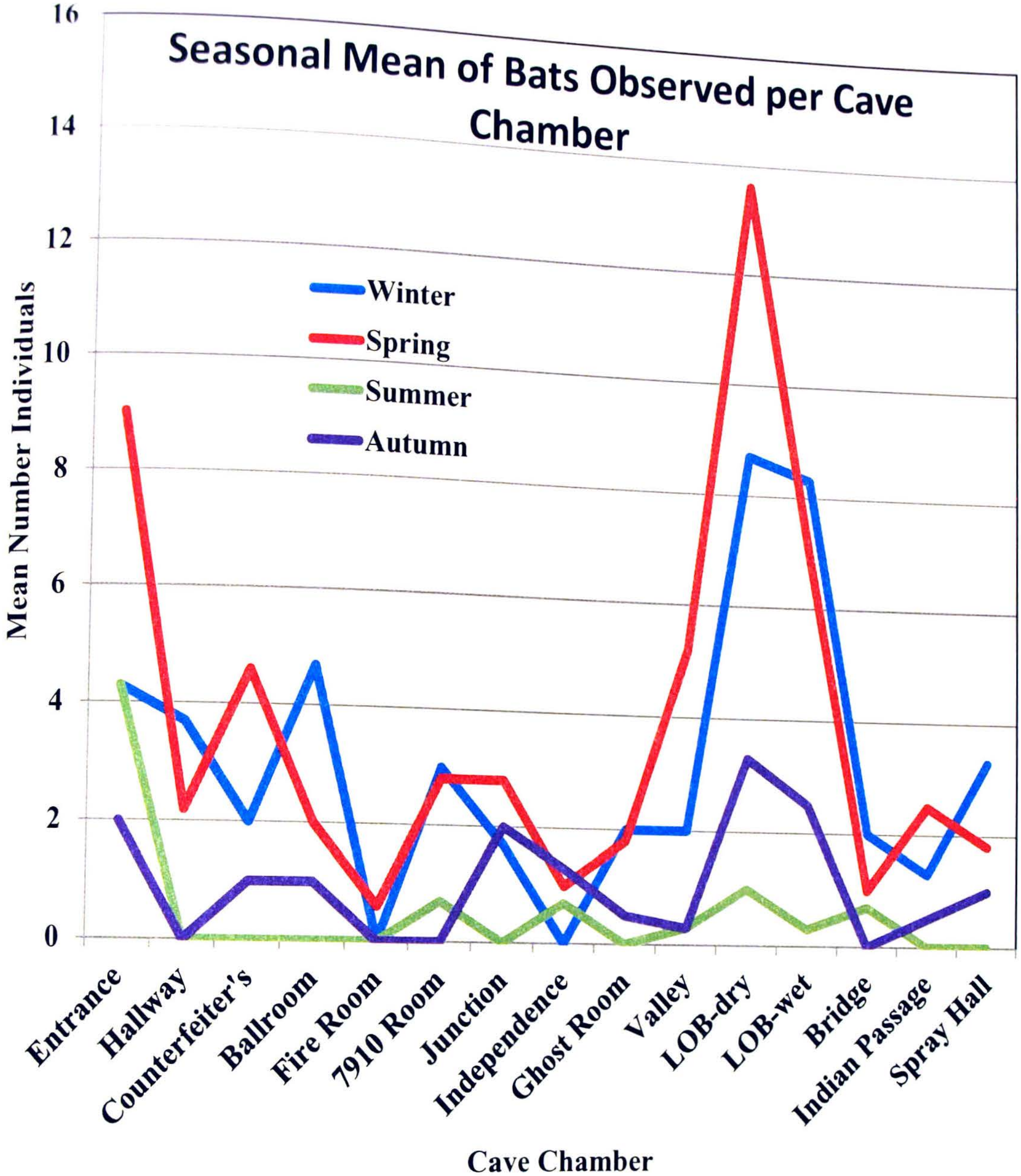


Figure 9. Seasonal mean number of bats observed for cave chambers for all cave surveys (n=16), 2011 and 2012.

Monthly mean temperature was recorded for seven cave chambers. Three of these chambers were monitored consistently over the course of one complete year. The Entrance chamber experienced the largest fluctuations in temperature (Figure 10). The average temperature range for the Entrance chamber was recorded as 8.9-13.4°C. The River Styx displayed little fluctuation in temperature (<1°C), ranging between 14.5 and 15.2°C. The area monitored between the two Lots of Bats chambers remained within 0.4°C (13.3°-13.7°C) throughout the year (Figure 10).

Four additional cave chambers were also monitored. Due to the lack in availability of data logger access, temperatures were recorded for shorter periods of time. The Hallway and Spray Hall chambers were monitored from March 30, 2012 until May 23, 2012 (Figure 11). During this time, the Hallway chamber displayed a < 1°C fluctuation (12.4-13.3°C). The Spray Hall chamber ranged from 13.6-13.7°C. The Junction chamber and the Counterfeiter's chamber were monitored from June 29, 2012 until October 23, 2012 (Figure 12). Unfortunately, the battery in the data logger placed in the Junction chamber lost charge after approximately two months of monitoring. Both of these cave chambers showed a very small variation in temperature (Figure 12).

The Entrance chamber displayed the highest fluctuation in temperature, comparatively (Figure 10). These fluctuations were consistent with external, seasonal temperature fluctuations. The lowest temperatures were recorded between December 2011 and January 2012 (Figure 10). Still, the temperature within this chamber did not drop below 8.9°C, while outside mean temperature was reported around 5.5°C (National Oceanic and Atmospheric Administration 2012). The highest temperatures in this chamber were recorded during July and August of 2012, but never exceeded 13.5°C (Figure 10).

Temperature was also recorded for the Hallway Chamber from March 2012 through May 2012 (Figure 11). This chamber is adjacent to the Entrance chamber, and therefore, most likely, experiences a certain amount of air flow from the above ground environment. The temperature in this chamber stayed around 13.0°C for the majority of the time that temperature was monitored here (Figure 11). Temperature did drop less than one degree in March of 2012, and was recorded at 12.4°C (Figure 11).

The wet and dry Lots of Bats chambers, the Spray Hall chamber, the Junction chamber, and the Counterfeiter's chamber all displayed steady ranges in temperature that did not fluctuate more than 0.5°C (Figures 10, 11, 12). Furthermore, the temperature in all of these chambers remained at 13.0+/-0.7°C. This is the usual temperature for most caves of this region (Matthews 2011). The temperature of the River Styx did not fluctuate as much as expected, but instead, remained constant within one degree Celsius (Figure 10). The recorded temperature was, however, consistently around two degrees higher (14.5°C-15.2°C) than any of the monitored cave chambers.

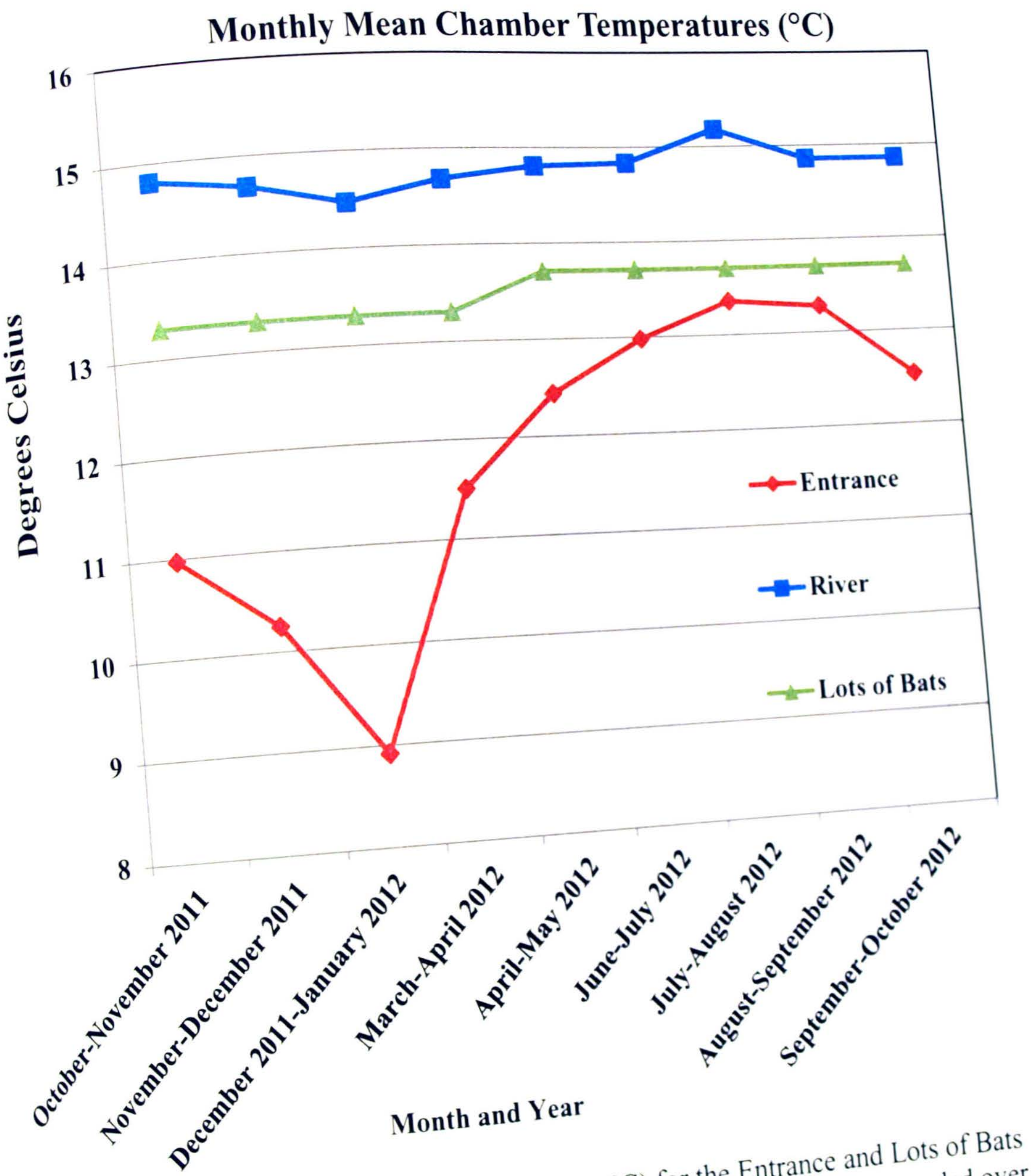


Figure 10. Monthly mean temperatures recorded (°C) for the Entrance and Lots of Bats (wet and dry) chambers, as well as, the River Styx site. Temperature was recorded over the course of one year.

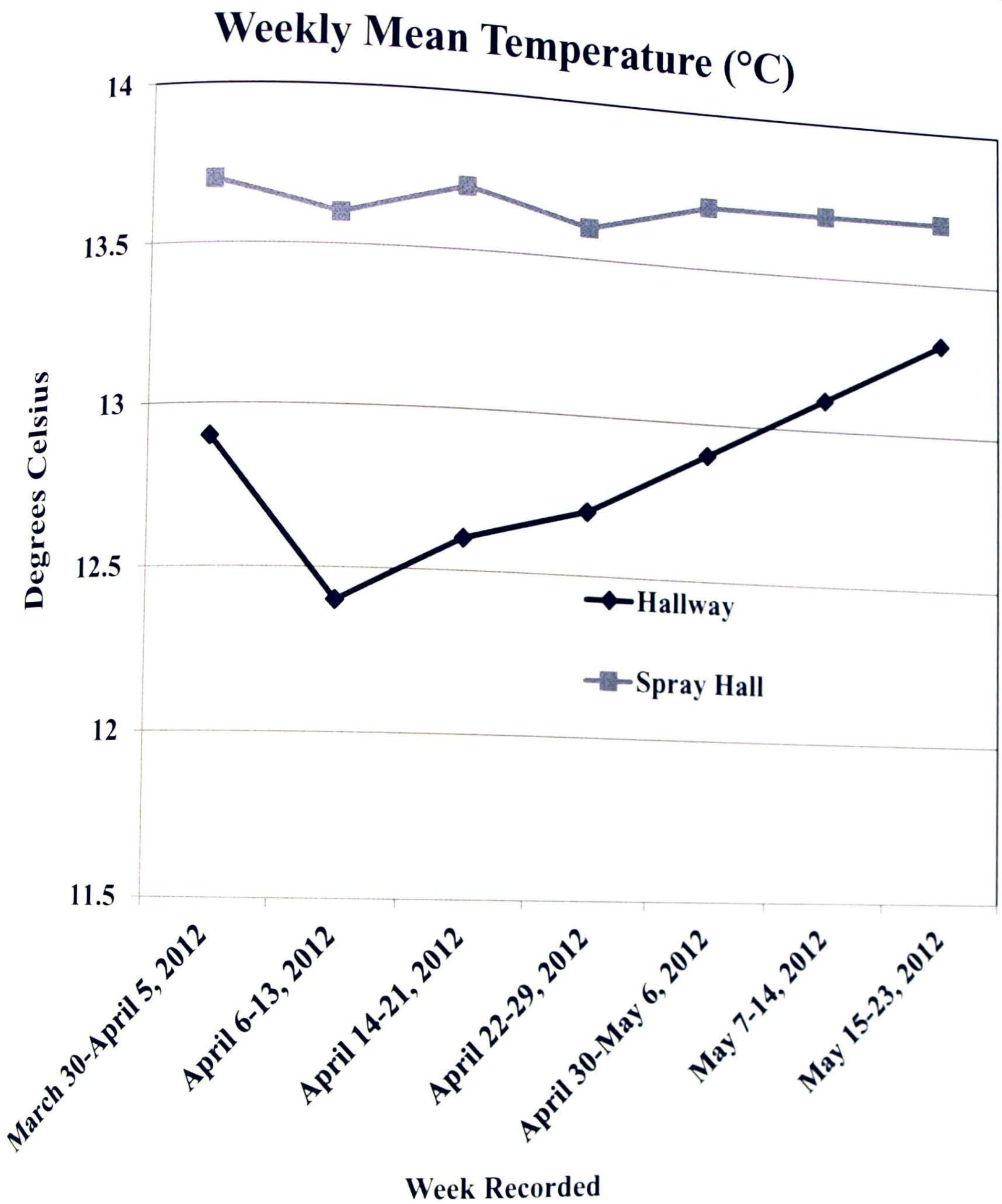


Figure 11. Weekly mean temperatures recorded for the Hallway and Spray Hall chambers. Temperature was recorded over the course of approximately two months.

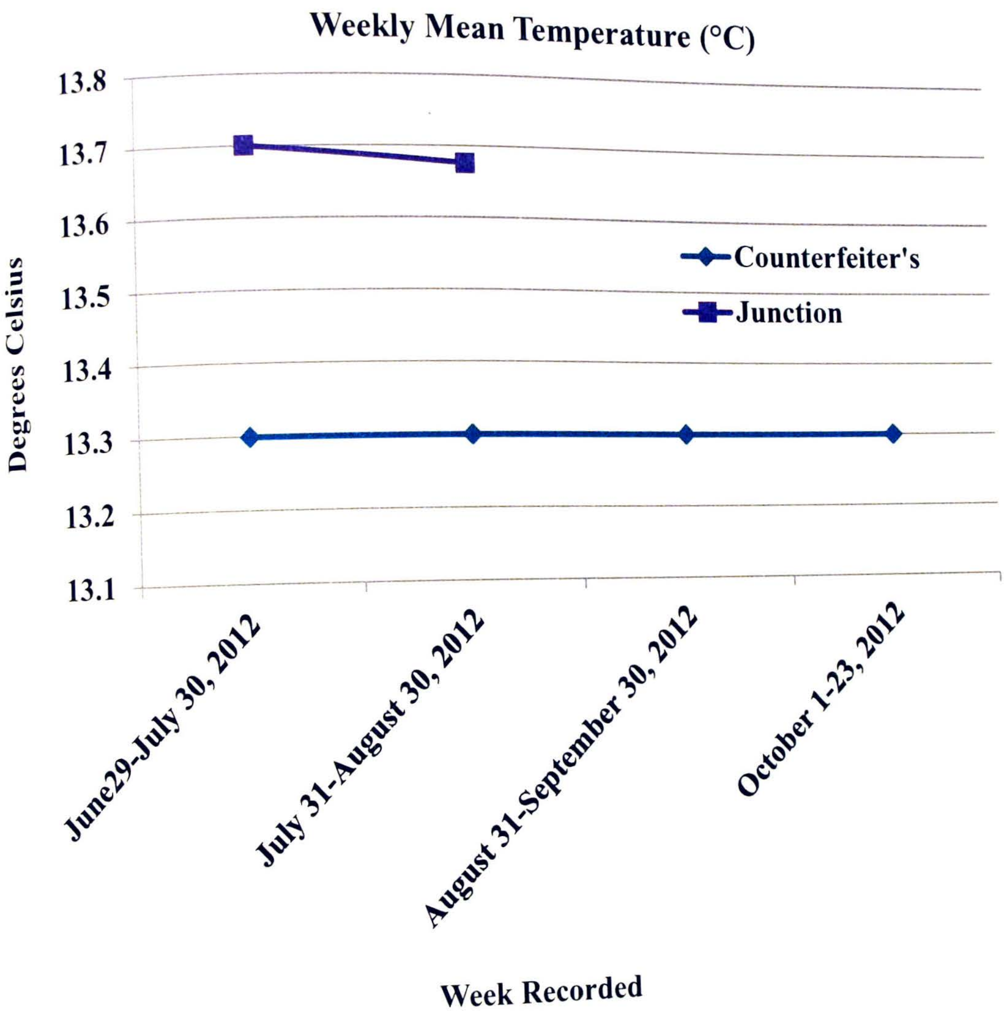


Figure 12. Weekly mean temperatures recorded for the Counterfeiter's and Junction chambers. Temperature was recorded over the course of four months. After two months of recording, the data logger in the Junction chamber exhibited battery failure.

It was often unknown if individual bats observed during cave surveys were the same individuals using the cave or specific chambers over time. For this reason, the overall mean number of individuals per species was used to calculate species diversity. *Perimyotis subflavus* was the most prevalent species documented using Dunbar Cave throughout the duration of this study (Table 11). *Myotis lucifugus* and *Myotis septentrionalis* were observed in low numbers (Table 11). Simpson's index of diversity was calculated as 0.696. The reciprocal of Simpson's diversity was determined as, 1/D = 1.436. Simpson's measure of species evenness, which ranges from 0 (low value) to 1, was calculated as 0.479.

Table 11. Bat species observed during cave surveys

Survey Date	*Perimyotis subflavus*	*Myotis lucifugus*	*Myotis septentrionalis*
February 21, 2011	58	0	2
April 1, 2011	66	0	4
May 12, 2011	46	0	0
June 8, 2011	2	0	0
August 23, 2011	10	0	0
October 18, 2011	16	0	0
December 8, 2011	30	0	0
January 10, 2012	13	2	0
January 27, 2012	46	5	0
March 30, 2012	53	0	0
April 12, 2012	74	0	0
April 27, 2012	52	0	0
May 23, 2012	20	0	0
June 21, 2012	2	0	0
August 28, 2012	9	0	0
October 23, 2012	27	0	0
Overall Mean	**32.8**	**0.44**	**0.38**

A total of 69 individuals were banded during the five cave surveys in which banding was permitted (Table 12). Forty five males and 24 female bats were banded within the cave (Table 12). The male to female ratio did not fit the expected 1:1 ratio (Fisher's exact test, $p=0.6844$, $\alpha=0.05$, $df=4$). The majority of bats banded ($N=63$) were *Perimyotis subflavus* (Table 13). Sixteen of the bats captured by hand during banding surveys within the cave were individuals that had originally been banded at an earlier date (Table 14). Individuals M0127 and M0171 were observed on more than one occasion (Table 14). Percent of banded to total number of individuals observed varied per cave survey. Overall, approximately 9.9% of all bats observed within the cave at any given time were previously banded individuals (Figure 13).

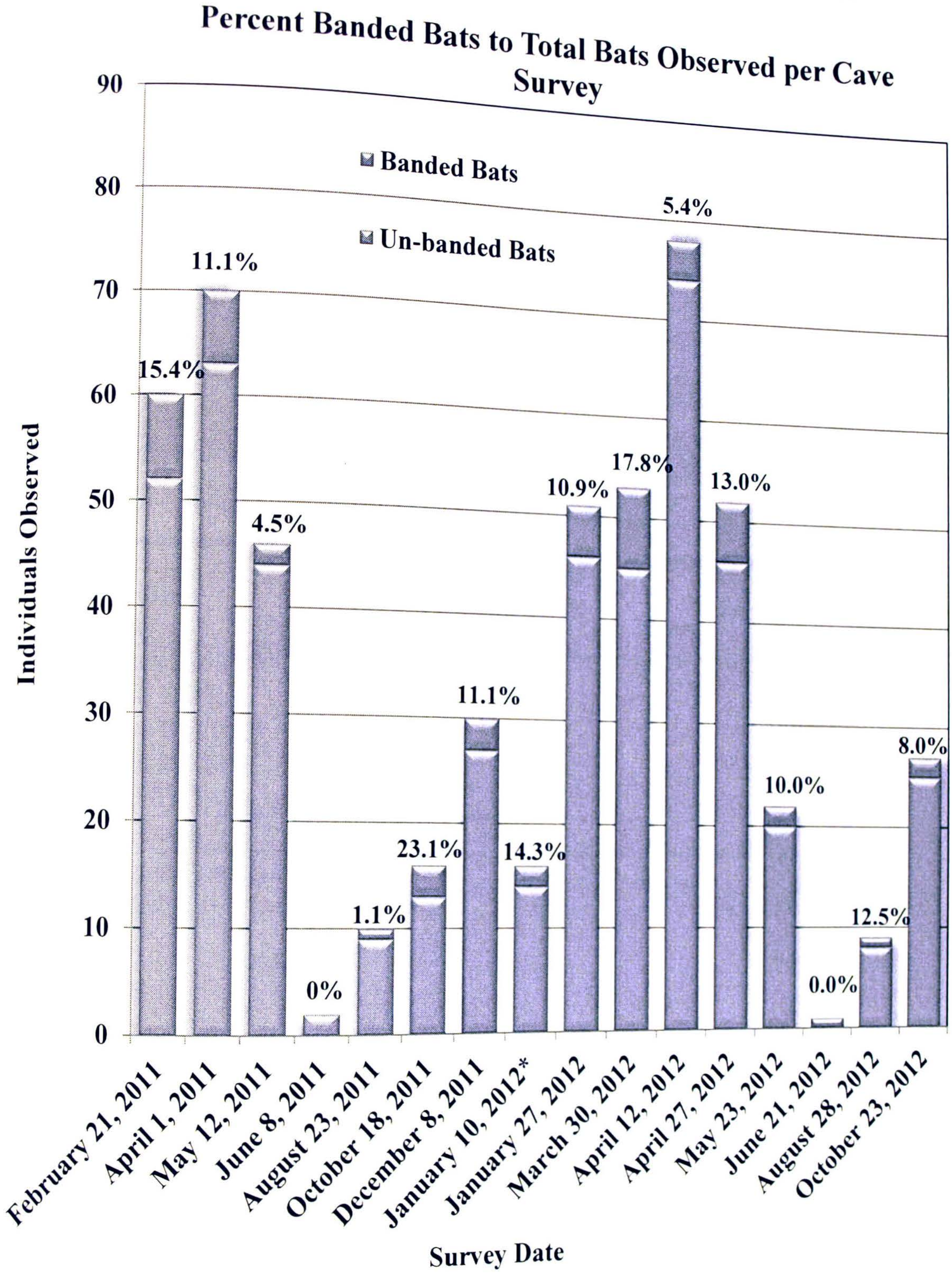


Figure 13. Percent of banded to total number of individuals observed during cave surveys at DCSNA (2011-2012).

Table 12. Total number of bats banded and sex of banded bats during cave surveys at DCSNA

Date	Number of Individuals Banded	# Males	#Female
February 21, 2011	8	4	4
April 1, 2011	6	5	1
January 10, 2012*	8	5	3
March 30, 2012	17	10	7
April 27, 2012	30	21	9
Total	**69**	**45**	**24**

*indicates an incomplete cave survey

Table 13. Total number of bats per species banded during cave surveys at DCSNA .

Species	**Year**		
	2011	**2012**	**Total**
Perimyotis subflavus	9	54	63
Myotis lucifugus	0	1	1
Myotis septentrionalis	5	0	5
Total	14	55	**69**

Banding was allowed during 5 cave surveys (4 complete and 1 incomplete)

Table 14. Bats recaptured during cave surveys at DCSNA

Date Banded	Date Recaptured	Initial Age	Sex	Band Identification	Initial WCI	Recapture WCI
April 29, 2010	February 21, 2011	A	M	M0183	0	0
April 29, 2010	February 21, 2011	A	M	M0167	0	0
August 12, 2009	February 21, 2011	J	M	M0124	0	0
August 12, 2009	February 21, 2011	A	M	M0127*	0	0
May 18, 2009	February 21, 2011	A	M	M0044	0	0
April 29, 2010	April 1, 2011	A	M	M0171*	0	0
April 29, 2010	May 12, 2011	A	M	M0171*	0	0
February 21, 2011	October 18, 2011	A	F	000785(TWRA)	0	0
September 28, 2009	January 10, 2012	A	M	M0099	0	0
August 12, 2009	January 10, 2012	J	M	M0133	0	0
July 27, 2011	March 30, 2012	J	M	M0214	0	0
April 29, 2010	March 30, 2012	A	M	M0157	0	0
April 29, 2010	March 30, 2012	A	M	M0168	0	0
August 12, 2009	March 30, 2012	J	M	M0120	0	0
August 12, 2009	March 30, 2012	A	M	M0127*	0	0
August 12, 2009	April 27, 2012	A	M	M0018	0	0

N=16

* indicates multiple recaptures of individuals

Individual M0099 = *Myotis lucifugus*

All other individuals = *Perimyotis subflavus*

Spatial analysis within specific cave chambers was performed in order to provide quantitative data of bat populations within Dunbar Cave, and to determine whether bats were uniformly or unevenly distributed. Because there were a higher number of individuals observed within the cave during spring surveys, spatial analyses focused on this season.

The cave surveys of February 21, 2011, April 1, 2011, March 30, 2012, and April 12, 2012 were chosen, because more individuals were observed during these times (Table 11). The 73-10 and Lots of Bats chambers displayed both heterogeneous and homogeneous distribution among the bat population (Table 15). The bat population within the Spray Hall chamber consistently displayed a heterogeneous distribution during both months of 2011 (Table 15). This chamber was not investigated during 2012 cave surveys, because only one individual was observed during both the March and April surveys (Appendix E).

Table 15. Morisita's Standardized Index of Dispersion

Date	Chamber	I_δ
February 21, 2011	73-10	7.89
April 1, 2011	73-10	0.00
March 30, 2012	73-10	0.00
April 12, 2012	73-10	1.97
February 21, 2011	Lots of Bats (dry)	9.87
April 1, 2011	Lots of Bats (dry)	13.81
March 30, 2012	Lots of Bats (dry)	1.97
April 12, 2012	Lots of Bats (dry)	0.00
February 21, 2011	Spray Hall	11.84
April 1, 2011	Spray Hall	5.92

$I_\delta=0$ for random distribution

$I_\delta<0$ for homogeneous distribution

$I_\delta>0$ for heterogeneous distribution

Acoustics

All acoustic data collected during harp trapping surveys and attempted mist netting events during 2011 were analyzed using Bat Call Identification 2010 (Bat Call Identification, Inc. Springfield, MO, USA). This program detected the presence of a total of nine bat species at Dunbar Cave State Natural Area (Appendix F).

Bio-acoustic data collected during the early May and August harp trapping events of 2012 were analyzed using Echo Class version 1.0 (ERDC, Army Corp of Engineers, USA). These results were compared with the capture data from the same dates (Appendix F). Echo Class version 1.0 bio-acoustic software detected the presence of 11 species (Appendix F).

Trends in Cave Use (Past and Present)

Mean number of bats observed within Dunbar Cave during cave surveys was not included in the CJS model. Instead these numbers were assessed seasonally, and compared to existing data to investigate trends in abundance over time. A consistent trend of higher population means were observed within the cave during spring and winter compared to summer and autumn. This was found both statistically (Table 10), as well as, observationally (Figure 14). There was no statistically significant difference between mean number of bats observed among spring and winter cave surveys (Table 10). A slightly higher number of bats were recorded within the cave, however, during spring of both years of this study (Table 10). In fact, this is the trend throughout the entire catalog of data from cave surveys at Dunbar Cave (Center of Excellence in Field Biology 2005-2012; Kurz 2011) (Figure 14).

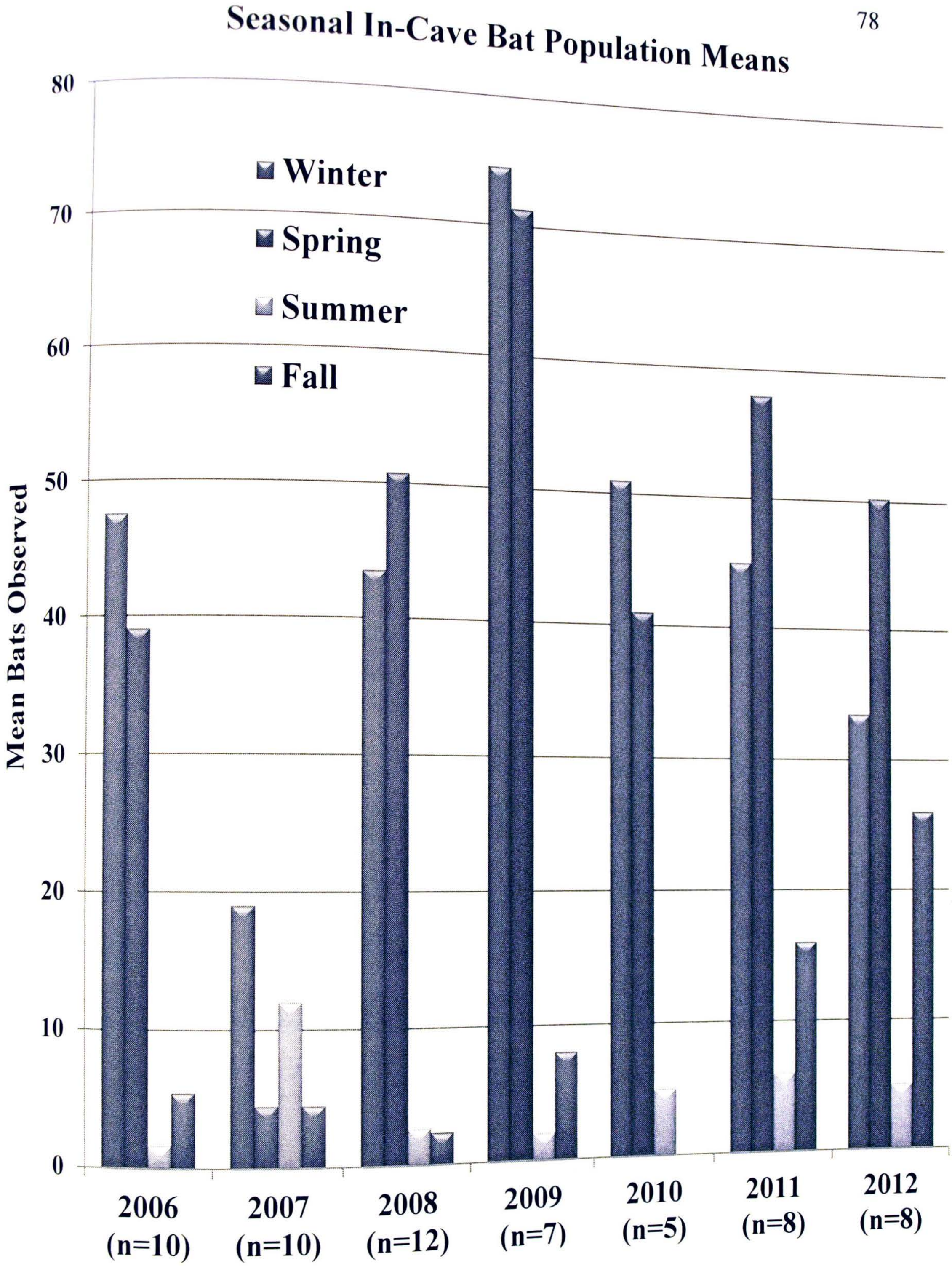


Figure 14. Seasonal mean number of bats observed for all cave surveys in the Center of Excellence for Field Biology data catalog (n=number of cave surveys per year).

The highest number of bats observed within Dunbar Cave was 78 individuals on April 12, 2012 (Table 9). In April of 2009, 86 bats were observed in Dunbar Cave (Center of Excellence for Field Biology 2005-2012). An increase in population means in late March to early May appears to be a consistent trend across the existing data (Center of Excellence for Field Biology 2005-2012).

Yearly mean number of bats observed during cave surveys has remained relatively constant from 2006 to 2012 (Figure 15). In 2007 bat populations declined, due to feral cat predation (Kurz 2011). In 2009 a peak in mean population numbers was observed (Figure 15). In March of 2010, a White Nose Syndrome positive bat was discovered within one of the chambers of Dunbar Cave (Kurz 2011). At this time, the cave was closed by supervising agencies, and research assistants were not allowed to enter until further notice (Kurz 2011). Throughout much of the Eastern United States, it has been commonly observed that upon initial detection of White Nose Syndrome within a cave system, an associated increase in mortality is also observed among the bat population (United States Fish and Wildlife Service 2012). The bat populations of Dunbar Cave did not appear to experience this associated increase in mortality (Kurz 2011).

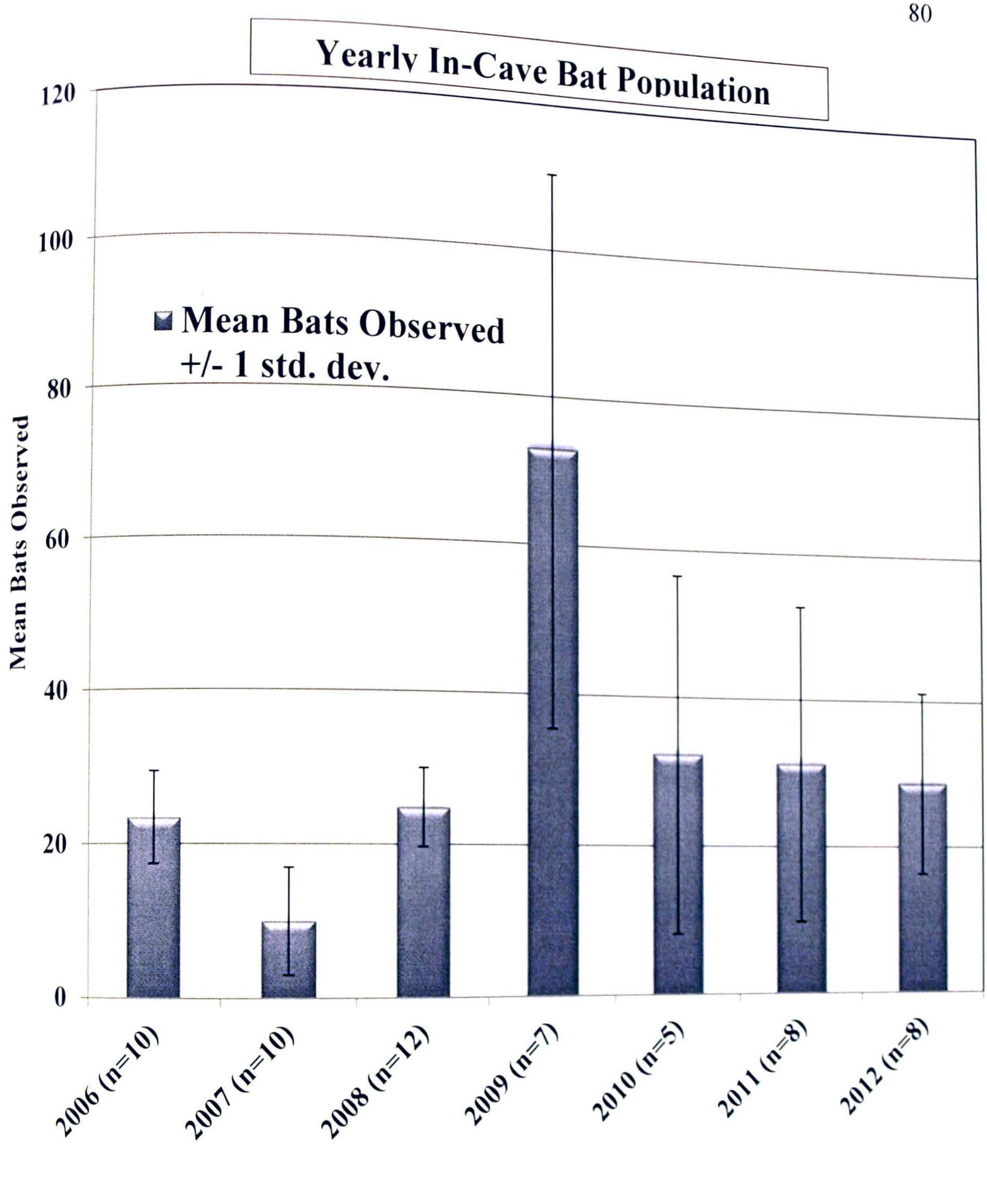


Figure 15. Yearly mean number of bats observed (+/- 1 standard deviation) for all cave surveys in the Center of Excellence for Field Biology data catalog (n=number of cave surveys per year).

CHAPTER IV

DISCUSSION

Conclusions and Future Recommendations

Several conclusions to the original predictions were made based off of literature review and the results of this study.

1. At this time, it cannot be definitively concluded that abundance of the cave-dwelling bat population(s) at Dunbar Cave is increasing.

Historically, it has been notoriously difficult to obtain reliable estimates of abundance on bat populations, and several techniques have been applied in an attempt to do so (McCracken 2003; O'Shea et al. 2004; Thompson 2004). In fact, the general consensus is that abundance estimates may be somewhat erroneous when applied to bat populations (Kunz 2003; McCracken 2003). This is mainly because annual cycles among bat populations can include seasonal long-distance migrations, and different species form colonies of various size, sex, and age compositions at different times of the year (O'Shea et al. 2003). Other invaluable information may be obtained, however, such as, relatively accurate survival estimates (Kunz 2003; McCracken 2003).

The CJS model was a relatively accurate predictor for total abundance (N) at this site. In fact, the actual number of individuals captured during the final month of capture for this study (August 2012), as well as, the actual number of individuals captured during the four years of banding at this site, both fell within the predicted range of N (199.7-597.7). In August 2012, 216 individuals were captured (Table 3), and since 2009, 564 bats have been captured at this site through the use of the harp trap.

Statistical results did not support the prediction that abundance at this site is increasing (Kruskal-Wallis rank sum analysis, $p = 0.4532$, $\alpha = 0.05$, $df = 3$). Despite this, the largest number of individuals since bat research initiated at this site was observed during this study (145 individuals; August 23, 2012; Table 1) (Kurz 2011). Although this capture event alone cannot represent a shift among the entire population, it may be a good indication that the cave-dwelling bat population of Dunbar Cave is thriving.

It is difficult to compare the total abundance estimate from this study to results found in existing literature. This is mainly because there is a general lack in mark-recapture studies that focused primarily on *Perimyotis subflavus*. Nonetheless, this species remains as one of the most common bat species in the Eastern United States, and it is assumed that overall abundance would reflect this (Tuttle 2006). This being said, it is generally accepted among bat biologists that, due to the complexity and lack of understanding of bat movements, most population models will result in an underestimated value of N (McCracken 2003; Tuttle 2006). More intensive and instantaneous sampling is needed to more accurately estimate abundance among bat populations at this site (Krebs 1999; Kunz 2003).

At this time, it cannot be definitively concluded that the bat population at Dunbar Cave is increasing, based solely off of statistical analyses. Despite this, capture events and cave surveys have yielded promising results. The low number of observed recaptured individuals indicates that complete understanding of emigration and immigration at this site is lacking. Therefore, total abundance estimated by the CJS model was most likely an underestimate, and further investigation is needed (Krebs 1999; Gasper & Chytil 2002; O'Shea et al. 2004). In the future, abundance and survival investigations should be separated by age and sex to better fit assumptions, and gain more accurate estimates (Gasper & Chytil 2002).

2. Species diversity has increased among the cave-dwelling bat populations at Dunbar Cave.

Due to the dominating prevalence of one species (*P. subflavus*), diversity indices were low. Kurz (2011) previously documented individuals of *P. subflavus*, *E. fuscus*, and *M. lucifugus* utilizing Dunbar Cave. *Myotis septentrionalis*, however, had not been observed here prior to this study. This species is currently being considered for listing as a threatened species (United States Fish and Wildlife Service 2011). The documentation of *M. lucifugus* during cave surveys, as well as, capture events is also of biological significance at this time. Kurz reported observing only two *M. lucifugus* during the early part of her study in June 2009. This species was not documented again at this Natural Area, until January 2012 (Table 11). All together eight *M. lucifugus* were observed during this study.

Perimyotis subflavus and *Myotis lucifugus* have historically been considered two of the most common and widely distributed bat species of the eastern United States (Fenton & Barclay 1980; Briggler & Prather 2003). Both these species have also been documented as species most severely and commonly affected by White Nose Syndrome (Blehart et al. 2009; Gargas et al. 2009). Increased mortality associated with White Nose Syndrome has not been documented among the cave-dwelling bat population of Dunbar Cave, thus far.

The few individual *M. lucifugus* that were found during this study, however, did not appear to be in the best condition. On January 27, 2012, three little brown bats were found within the Hallway chamber of Dunbar Cave (Appendix H; Figure16). One individual appeared to have a white powdery substance, reminiscent of *Geomyces destructans* on its right forearm (Appendix H, Figure 16). Due to permit restrictions, the bats were not removed from the roost site, and therefore the actual presence of *G. destructans* was not confirmed. In addition, the single *M. lucifugus* captured at the cave entrance scored a WCI of 1, due to extensive wing discoloration (Appendix H, Figure 17).

Although slight, the increase in species diversity among the cave-dwelling bat populations at Dunbar Cave implies that these populations may have begun to recover from prolonged human disturbance. Human activity within Dunbar Cave has only recently been limited. In fact, organized public tours took place for the past three decades, up until late 2009 (Amy Wallace, Personal Communication, August 12, 2011). It is likely that the increase in population numbers and species diversity observed among the cave-dwelling bat population at Dunbar Cave is just the beginning of recovery. Studies involving long-term monitoring of bat populations of the eastern United States are scarce (Kunz 2003; McCracken 2003; O'Shea & Bogan 2003). Many bat species once considered common face severe population declines, and in some cases, potential extinction within the next few decades (Frick et al. 2010). Therefore, it is crucial that these populations continue to be monitored.

3. Several species of cave and forest dwelling bats were detected through the use of acoustic monitoring at Dunbar Cave State Natural Area.

Acoustic data from this study indicate that at least ten additional bat species are utilizing the areas directly adjacent to and surrounding Dunbar Cave. Many of these species are forest-dwelling and are considered relatively common in this area. Nevertheless, presence of *Myotis grisescens* was detected by both EchoClass and BCID software programs. It is suspected that this endangered species once used Dunbar Cave for maternal roosting.

Additional information is needed on the landscape surrounding Dunbar Cave, and how it is being utilized by bat populations. Acoustic monitoring should be expanded at this site, and used in combination with additional capture methods, such as; mist-netting. These methods could confirm the presence of bat species detected by bio-acoustic sampling units. Bio-acoustic data indicates that the majority of additional species detected is foraging above and around Swan Lake. To ensure success with this technique in the future, mist-netting efforts should be more intensive and focus on this area.

4. Site fidelity to this site remains low, and is not well understood at this time.

Upon initial review of the results from this study, two possible conclusions may be drawn, regarding the site-fidelity among the cave-dwelling bat population of Dunbar Cave. Either there is a low rate of site-fidelity among the banded individuals using Dunbar Cave, or only a small portion of the entire population at this site has been banded.

Generally, mark-recapture studies of a variety of bat species have yielded similarly low results of return to the original site of banding (Kunz 2003; McCracken 2003; O'Shea & Bogan, 2003). Cockrum (1969) banded 162,892 free-tailed bats over fifteen years, and reported recapturing a mere two percent of these individuals at the original banding site. An additional 0.3% of banded individuals were captured at other localities (Cockrum 1969). A more recent study focusing on mark-recapture of several cave-dwelling, temperate bat species of the Czech Republic reported results of recapture more comparable to this study (Gaisler & Chetyl 2002). Over the course of forty two years, individuals from eight species were banded during winter cave surveys, and by the final year of study (2002) 22.8% were observed returning (Gaisler & Chetyl 2002). Gaisler and Chetyl (2002) also began netting bats at the same cave in the Czech Republic during the summer of 1991. Over the nine years of continued netting, 13.4% were recaptured (Gaisler &Chetyl 2002).

A similar percentage of return (banded individuals captured to total individuals captured) was observed among the bat population during harp trapping events (8.3%) and cave surveys (9.9%). Although banded individuals observed within the cave were not included in the CJS population model, return rates for both methods were similar to the value predicted by the CJS model of proportion of the population that has been banded (0.095 or 9.5%) (Table 3).

The CJS maximum likelihood model estimated a mean rate of survival of 0.605 +/- 0.500 (0.105-1.105) between months for the sample population captured by harp trap at Dunbar Cave (Table 3). This suggests a low rate of survival (~10% living to four years with some bats living over ten years) among the bat population at this site (Rivers et al. 2006). It is difficult to accurately conclude whether this calculated survival rate is a correct approximation.

Based on rate of recapture of banded individuals, it appears as though a very low proportion of the banded population is returning to Dunbar Cave. There are several potential explanations for this. First of all, a large proportion of the individuals banded throughout the course of this study were juvenile males (Table 8). Although bats are relatively long-lived, in general, survival during the first year of a bat's life is often low, especially for juvenile males (O'Shea et al. 2004). Sendor and Simon (2003) found that age had the strongest effect on survival among colonies of *Pipistrellus pipistrellus,* and concluded that juveniles should be excluded from models, or analyzed separately to ensure more accurate results. In addition, analyses of male skewed bat populations are often affected by what is known as "the transient effect" (Sendor & Simon 2003). In other words, results may be skewed due to fact that male bats naturally display more movement and less site fidelity (Sendor & Simon 2003). Approximately 80.5% of all individuals captured during this study were male (Table 6). Male bats typically disperse farther from natal roost sites, travel farther distances, and move between several local sites during swarming season for optimal mating opportunities (Kerth et al. 2003; McCracken 2003; O'Shea et al. 2004). The low rate of site fidelity observed during summer and autumn could be due to this increased emigration, and difference in dispersion among male bats (McCracken 2003; O'Shea et al. 2004). Therefore, it is very likely that the low recapture rate observed at Dunbar Cave is due to the fact that the banded population is comprised mainly of male bats.

Despite the low number of banded bats observed returning to Dunbar Cave, any degree of site fidelity among bat populations is biologically significant (Rivers et al. 2006). Bats are potentially returning to this site, because specific biological conditions are being met here, such as; preferable roost sites or optimal foraging or reproductive conditions (Krebs & Davies 1997). It may also be possible that the portion of the population that was banded is showing loyalty to Dunbar Cave, as well as, a collection of other local sites (O'Shea et al. 2004). This perhaps explains why only some individuals are observed returning at particular times and some are not.

In order to completely and comprehensively understand bat population dynamics and social structure at this site, more involved methods are needed. It has been found that there is a higher degree of genetic variation among swarming bat assemblages compared to colonies of hibernating bats inhabiting the same sites (Kerth et al. 2003; Parsons et al. 2003; Rivers et al. 2006). In addition, it has been documented that individuals of several cave-dwelling bat species show an extremely high rate of site fidelity among three to four local sites that are usually within close proximity to each other, as well as natal roost sites (Rivers et al. 2006).

At this point, data are lacking, regarding the genetic make-up and local movement patterns of the cave-dwelling bat populations at Dunbar Cave. These factors should be investigated in the future to gain a clearer understanding of bat fidelity to this site. Radio-telemetry methods could be useful to locate nearby maternity roosts, swarming sites and hibernacula. In addition, non-invasive genetic sampling would assist in gaining a clearer insight into the social structure of these populations. Swarming has been noted to peak in mid-September to October and individuals may not return over consecutive nights (Rivers et al. 2006). The majority of harp trapping events undertaken at this site have occurred between the months of May through August. Future swarming studies should attempt to schedule capture events during August, September, October, and November to obtain samples that better represent this time period (Rivers et al. 2006).

5. Statistically, there was not a significant increase of females among the population over time.

Statistical analyses did not support the prediction that an overall increase in female bats would be observed among the population over time. This was difficult to assess, due to the fact that demographic data was not collected during the majority of cave surveys. The increase in female abundance observed during July and August of both years of this study, however, further confirms Kurz's (2011) observation that Dunbar Cave was and continues to be used as a site for swarming (i.e. reproductive behaviors).

Swarming is a reproductive behavior observed among temperate bat species, and is characterized by erratic and irregular flight in and around underground sites, typically used as winter hibernacula (Kerth et al. 2003). Large aggregations of multiple bat species are associated with "swarming" and typically individuals are considered transient during this time (Kerth et al. 2003; Parsons et al. 2003). This further explains why a relatively low number of individuals have been observed returning during months of capture. In fact, the entirety of mark-recapture data used for the calculation of population parameters were collected during late spring, summer, and early fall. This should also be taken into account when interpreting estimates of abundance and site fidelity. More frequent hands-on cave surveys and capture events are needed to explore the sexual composition of the bat population at this site.

6. Individuals of *Perimyotis subflavus* are forming aggregations in at least three of the chambers in Dunbar Cave.

The majority of the cave surveys in which spatial analysis was applied, resulted in a SMI value of $I_\delta>0$ (Table 15). A SMI value greater than zero indicates a heterogeneous distribution among the population, or in other words, implies clumping among the population (Krebs 1999). Three of the ten analyses, however, resulted in a I_δ value of 0.00 (Table 15). Theses specific cave surveys took place on April 1, 2011 and March 30, 2012 within the 73-10 chamber, and on April 12, 2012 within the dry Lots of Bats chamber (Table 15). A I_δ value of zero indicates that the population is randomly distributed, or that no clumping is present (Krebs 1999).

It is difficult to draw definite conclusions regarding the spatial distribution of the bat population within Dunbar Cave, based on these results. It does appear, however, that for the majority of the time, at least during the cave surveys included in the analyses, individual bats are roosting within close proximity of one another.

All individuals included in these analyses were *Perimyotis subflavus*. This species is known to roost singularly (Fujita & Kunz 1984; Sandel et al. 2001). Patterns of spatial distribution, however, imply that individuals may be forming aggregations within specific areas of the cave (Appendix E). In fact, it appears as though specific areas within cave chambers are being used repeatedly (Appendix E). This could imply that there are "prime" or "preferred" roost sites within Dunbar Cave, and that as density increases, individuals form aggregations in these areas. On the other hand, *Perimyotis subflavus* is known to be loyal to winter hibernacula, as well as, specific roost sites within these hibernacula (Briggler & Prather 2003). It is unclear at this point, however, whether the individuals noted in the same areas during separate cave surveys are returning or different individuals.

Perimyotis subflavus is known to use a wide range of temperatures within hibernacula (Hill & Smith 1984). Briggler and Prather (2003) found that this species prefers larger, warmer caves that provide a variety of thermally stable roost sites. This may allow these bats to shift positions throughout the winter. From the same study, Briggler and Prather (2003) found that significantly more individuals of *P. subflavus* were observed in caves that contained wide temperature gradients, but that showed smaller changes in temperature between seasons.

Dunbar Cave is used year round by local bat populations, particularly *P. subflavus*, and numbers within the cave peak in spring (Figure 17). Seasonal cave use at Dunbar Cave is consistent with what would be expected among temperate bat species (Barbour & Davis 1969; Briggler & Prather 2003). First of all, during summer there are few individuals observed roosting in Dunbar Cave (Figure 9; Appendix G). The bat population captured at the cave entrance, during this same season, was comprised almost entirely of males (Table 6). These observations further confirm Kurz's (2011) conclusion that Dunbar Cave serves as a roost site for male bachelor colonies. Investigation of chamber use during this time illustrates that the occupying bat population is typically roosting within the Entrance chamber of Dunbar Cave (Appendix G). It is likely that during summer, individual male bats use Dunbar Cave as a "day-roost", as well as, a place to rest between nightly feeding bouts (Sandel et al. 2001; Kerth 2008).

In autumn individuals of both sexes are observed congregating around the cave entrance, suggesting the onset of swarming (i.e. reproductive behaviors) (Figure 7). At the same time, individuals begin to disperse more evenly throughout cave chambers (Appendix G). During autumn cave surveys several individuals were noted as being completely awake and were often seen flying and chasing each other throughout the cave. This chase behavior is associated with reproduction among Vespertillionid bat species (Kerth et al. 2003; Parsons et al. 2003).

Demographic information, such as, sexual composition of the population was limited during winter cave surveys. The data that was obtained, however, imply that there is a more even sex ratio among the hibernating population (Table 6), which has also been observed in previous studies (Rabinowitz 1981; Sandel et al. 2001). Individuals are also more evenly distributed throughout cave chambers during this time (Appendix G). Numerous individuals were also noted as having visual collections of condensation on body surfaces. Although most bats may begin to collect condensation during short bouts of torpor, *P. subflavus* typically becomes completely covered in large drops of dew during prolonged torpor (Briggler & Prather 2003). This combined with the fact that some of these individuals are documented in the same roost sites during consecutive winter cave surveys, suggests that they may have remained in the same position for a substantial amount of time (Willis et al. 2006; Willis & Brigham 2003). Consequently, this confirms the use of Dunbar Cave as a winter hibernacula (Sandel et al. 2001).

Although statistical analyses did not reveal significant differences between population means within individual cave chambers, at least three chambers appeared to be preferred throughout the year. These included; the Entrance chamber, and both the Wet and Dry Lots of Bats chambers (Figure 11). The Entrance chamber appeared to be used more frequently during summer, and individuals were observed using cave chambers located deeper within the cave system during all other seasons (Appendix G).

Few additional conclusions can be made regarding seasonal cave chamber occupation without more information on confounding biotic and abiotic factors.

Temperature data along with spatial analyses generally supports the suggestion that *P. subflavus* prefer cave chambers with a low temperature gradient. The Entrance and Hallway chambers of the cave display the highest temperature gradients, and are the only chambers where *Myotis* species were found during this study. *Perimyotis subflavus* is more frequently found in the Entrance chamber during spring and summer. This is most likely, because these bats can better tolerate the higher temperature gradient associated with the Entrance chamber during these warmer periods. The Lots of Bats chambers are preferred by *P. subflavus* throughout the year, most likely, because these chambers have remained undisturbed for several decades and they are located deep within the Dunbar Cave system and do not experience large influxes of water. In addition, the Lots of Bats chambers displayed the most stable temperature. It is unknown how the flow of water through Dunbar Cave may be affecting internal cave temperature and humidity, if at all. A bridge crosses over the River Styx in the specific area where the data logger was placed. This specific location was chosen, because the data logger could be secured to the bridge, and retrieved during successive cave surveys. The water temperatures recorded may only be representative of this specific area. In addition, above ground water is constantly seeping through the walls and ceilings of various chambers. This could also influence water temperature within the River Styx at varying locations throughout the cave.

Seasonal chamber use and overall population movement within Dunbar Cave reveals a more even distribution of bats throughout all cave chambers during the spring season (Appendix G). There are several potential explanations regarding both the increased population means observed within this cave system during the spring season and the observed trend in distribution and increase in overall movement.

First of all, during the last few months of the hibernation season, individual males may alternate between bouts of torpor and arousal in pursuit of final attempts to copulate (Barbour & Davis 1969; Barbour & Davis 1974). Female bats may also begin arousing in anticipation of parturition and emergence for formation of maternity colonies (Willis et al. 2006). Moreover, as external temperature increases, insects once again become available for consumption. Individuals of both sexes begin to arouse from torpor, and will begin to exit the cave in search of food and water (De Jong & Ingemar 1991; Willis & Brigham 2003; Willis et al. 2006). During this time bats have been documented not only moving among cave chambers, but also moving between local sites (Baranauskas 2001; Gaisler & Chytil 2002). It remains unknown, however, whether the observed increase in population between winter and spring at Dunbar Cave is due to the arrival of new individuals to this site, or if, as external temperature increases, individuals slowly move towards the entrance of the cave. These individuals may have been in areas of the cave that were not accessible or visible to researchers during earlier cave surveys.

Additional information is needed to draw more specific conclusions. Roosting habits may vary seasonally, according to sex, reproductive condition, social organization, and food habits (Kunz et al. 1983). Additionally, the type of roost inside the cave, the number of individuals per site, and roost acquaintances/social interactions could each be influenced by individual bats. Other considerations, for individual species-species distribution could be caused by dispersion and abundance of food resources, predation, and energy imposed by body size and the physical environment (Kunz 1983).

External and internal environmental factors, such as fluctuations in temperature, humidity, and air flow, have not been thoroughly explored in this cave system. Therefore, it is impossible to conclude whether differences in population means are simply occurring in response to normal fluctuations in external or internal environmental changes, at this time.

Dunbar Cave was once inhabited by large colonies of *M. grisescens*. This species is known for having very particular and specific roost preferences (Barbour & Davis1969; Ellison et al. 2003). *Perimyotis subflavus* on the other hand are considered to be generalist in almost every aspect of their biology (Briggler & Prather 2003). Therefore, it is not surprising that *P. subflavus* is the most prevalent species at this site. The two *Myotis* species documented at this site, however, are slightly more selective in regard to roost site selection (Owen et al. 2003; Barclay 1982). The presence of *M. lucifugus* and *M. septentrionalis* indicates that Dunbar Cave also possesses characteristics that are appealing to species that are not considered ecological generalists.

Further investigation of biotic and abiotic factors should be explored to better understand this. For example, temperature should be more closely monitored, specifically, at individual roost sites, or areas where aggregations of individuals are observed. The collection of cave temperature data is essential to the monitoring of bat populations within hibernacula and summer roost sites. Any fluctuations detected may reveal possible causes for abundance changes within these populations (O'Shea, et al. 2003).

Additional roost site characteristics should be investigated, as well; including, distance from entrance, relative humidity or percent roost exposure. In addition, differences in seasonal range in body temperature of individuals within the cave could provide better understanding of cave utilization among the local bat population, as well. Spatial heterogeneity among the bat population of Dunbar Cave further suggest population patchiness, social behavior, site fidelity, and mating preferences. All of these may enhance population persistence (Durant, 2000). Spatial distribution analysis should be expanded at this site, and be applied to other areas of the cave.

Conservation and Management Implications

Results from this study indicate that Dunbar Cave is being used year-round by local bat populations. Specifically, this cave serves as a hibernaculum and a swarming site. The presence of *P. subflavus*, *M. lucifugus*, *M. septentrionalis* and *E. fuscus* was confirmed through capture. Additional species, including *M. grisescens*, were detected acoustically in the areas directly surrounding Dunbar Cave. Although the bat population at this site has not increased significantly over the last four years, numbers have remained stable. Moreover, mark-recapture and additional banding data indicate that the individuals utilizing Dunbar Cave vary greatly between years, seasons and, in some cases, within the same month. This variation suggests that more than one population inhabits this cave, and that there is a high level of movement among these populations. In addition, approximately 10% of the banded population has been observed returning to Dunbar Cave. Many of these individuals were originally banded as juveniles. This insinuates that not only are bats depending on Dunbar Cave year after year, but that they may also be recruiting other individuals to this site.

The species captured at Dunbar Cave are, or were at one time, considered common throughout the region (Briggler & Prather, 2003, Caceres & Barclay, 2000, Frick, et al. 2010). Within the last six years, however, these same species have experienced immense population declines, due to White Nose Syndrome (Blehart et al. 2009, Frick, et al. 2010). *Myotis septentrionalis* is currently being considered for listing as a threatened species by United States Fish and Wildlife Service (Center for Biological Diversity, 2011), and the few little brown bats (*M. lucifugus*) found during this study are part of a fragile population. Frick, et al. (2010) predicted that this species (*M. lucifugus*) may suffer regional extinction in some parts of the United States within the next twenty years. It is promising that *M. septentrionalis* was captured during both years of this study. This was the first time this species has been documented at this site. Furthermore, the *M. lucifugus* captured during 2012 were the first individuals of this species observed at Dunbar Cave since it was deemed WNS positive in 2010 (Kurz, 2011).

Historical and physical evidence suggests that Dunbar Cave was once inhabited by large colonies of cave-dwelling bats (Matthews 2011). Due to prolonged human disturbance, however, these populations either abandoned this cave or were destroyed. A study on the effects of commercial cave tours on bats found that there is, in fact, a positive correlation between the intensity of light and sound created by cave tours and bat activity (Mann, et al. 2002). In addition, human disturbance, vandalism, and commercialization of hibernacula, alter cave microclimates, and are principal factors in the decline of certain bat populations (Johnson, et al. 1998). Several bat species depend on caves and other underground sites as areas of refuge during times of vulnerability (O'Shea, et al. 2003). The preservation and conservation of bat roosts, especially caves, has been recognized as the most important issue in bat conservation (Sheffield, et al. 1992). This becomes particularly clear when realizing that many roosts are traditional, and hence used by successive generations of bats over many years (Sheffield, et al. 1992).

As human populations increase, wildlife habitat is used or destroyed. Local bat populations are not only affected by anthropogenic disturbance, but also face complications and potential devastation from White Nose Syndrome. Bat species once considered common may become threatened or extinct within the near future (Frick, et al. 2010). Studies on cave-dwelling bat populations indicate that various species display differing levels of sensitivity to the combined damaging effects of climate change and anthropogenic disturbance (Mann, et al. 2002, Scheel, et al. 1996). The cave-dwelling bat population of Dunbar Cave appears to be recovering from prolonged human disturbance, perhaps due to the elimination of cave tours (Mann, et al. 2002). In general, bats are long-lived and are not highly fecund (Jones, et al. 2009). Therefore, severely decimated bat populations may take several years, if not decades, to recover. It is imperative that monitoring of these populations is continued. Banding is especially important at this site and will assist in documenting consecutive generations of bats at Dunbar Cave. Bat populations should be continually and consistently monitored at this site in order to gain insight into future conservation efforts needed.

LITERATURE REVIEWED

Baranauskas, K. 2001. Hibernation of Barbastelle (*Barbastella barbastellus*) in Dedkinë bunkers in Vilnius (Lithuania): A possible bat population response to climate change. *Acta Zoologica Lituanica*.**11**, 15-19.

Barbour, R. W. & Davis, W. H. 1969. *Bats of America*. Lexington, KY: The University Press of Kentucky.

Barbour, R.W. and Davis, W.H. 1974. *Mammals of Kentucky*. Lexington, KY: The University Press of Kentucky.

Barclay, R.M.R. 1982. Night roosting behavior of the little brown bat, *Myotis lucifugus*. *Journal of Mammalogy*. **63(3)**, 464-474.

Blehart, D.S., Hicks, A.C., Behr, M. Meteyer, C.U., Berlowski-Zier, B.M., Buckles, E.L., Coleman, J.T.H., Darling, S.R., Gargas, A., Niver, R., Okoniewski, J.C., Rudd, R.J., Stone, W.B. 2009. Bat white-nose syndrome: An emerging fungal pathogen? *Science*. **323**, 227.

Boyles, J.G., Cryan, P.M., McCracken, G.F., Kunz, T.H. 2011. Economic importance of bats in agriculture. *Science*. **332,** 41-42.

Boyles, J.G. & Willis, C. K.R. 2009. Could localized warm areas inside cold caves reduce mortality of hibernating bats affected by white-nose syndrome? *Frontiers in Ecology and the Environment*. **8**, 92-98.

Briggler, J.T. & Prather, J.W. 2003. Seasonal use and selection of caves by the Eastern Pipistrelle bat (*Pipistrellus subflavus*). *American Midland Naturalist*. **149 (2)**, 406-412.

Brigham, R.M. 2007. Bats in forests: What we know and what we need to learn. In: *Bats in forests: Conservation and management.* (Ed. By M.J. Lacki, J.P. Hayes, A. Kurta), pp. 1-15. Baltimore, MD: Johns Hopkins University Press.

Buckland, S.T. & Elston, D. A. 1993. Empirical models for the spatial distribution of wildlife. *Journal of Applied Ecology*. **30**, 478-495.

Caceres, M.C., Barclay, M.R. 2000. *Myotis septentrionalis. Mammalian Species.* **634**. 1-4.

Center for Biological Diversity. 2011. Two U.S. bat species one step closer to endangered species act protection [Press release]. Retrieved from: http://www.biologicaldiversity.org/news/press_releases/2011/white-nose-syndrome-06-28-2011.html.

Center of Excellence for Field Biology. 2005-2012.[Dunbar Cave Bat Survey]. Unpublished raw data.

Cockrum, E.L. 1969. Migration in the guano bat. *Tadarida brasilliensis. Miscellaneous Publications.* The University of Kansas Museum of Natural History, **51**, 303-336.

Cockrum, E.L. 1955. Reproduction in North American bats. *Transactions of the Kansas Academy of Science.* **58 (4)**, 487-511.

Cope, J.B. & Humphrey, S.R. 1977. Spring and autumn swarming behavior in the Indiana bat, *Myotis sodalis*. *Journal of Mammalogy*. **58(1)**, 93-95.

Cryan, P.M., Meteyer, C.U., Boyles, J.G., Blehert, D.S. 2010. Wing pathology of white-nose syndrome in bats suggests life-threatening disruption of physiology. *BMC Biology*. **8**, 135.

Davis, W.H. 1959. Disproportionate sex ratios in hibernating bats. *Journal of Mammalogy*. **40**, 16-19.

De Jong, J. & Ingemar, A. 1991. Factors affecting the distribution pattern of bats in Uppland, Central Sweden. *Holarctic Ecology*. **14**, 92-96.

Durant, S. 2000. Dispersal patterns, social organization, and population viability. In: *Behaviour and Conservation*. (Ed. By Gosling, L.M. & Sutherland, W.J.). pp.172-197. Cambridge, UK: Cambridge University Press.

Ellison, L.E., O'Shea, T.J., Bogan M.A., Everette A.L., and Schneider D.M.. 2003. Existing data on colonies of bats in the United States: summary and analysis of the U.S. Geological Survey's Bat Population Database. In: O'Shea, T.J., and M.A. Bogan (eds.). Monitoring trends in bat populations of the United States and territories: problems and prospects. *Information and Technology Report* **2003-0003**. : U.S. Geological Survey. 127-237.

Fenton, B.M. & Barclay, M.R. 1980. *Myotis lucifugus*. *Mammalian Species*. **142**, 1-8.

Fenton, M.B. 2003. Science and conservation of bats: Where to next? *Wildlife Society Bulletin*. **31 (1)**, 6-15.

Fenton, M.B. 1988. Detecting, recording, and analyzing vocalizations of bats. In: *Ecological and Behavioral Methods for the Study of Bats* (Ed. By T.H. Kunz), pp. 91-104. Washington, D.C.: Smithsonian Institution Press.

Findley, J.S. 1993. *Bats: A community perspective.* Cambridge, UK: Cambridge University Press.

Flaguer, C., Torre, I., Arrizabalaga, A. 2007. Comparison of sampling methods for inventory of bat communities. *Journal of Mammalogy.* **88**, 2, 524-533.

Forsyth, A. 1999. *Mammals of North America: Temperate and arctic regions.* Buffalo, NY: Firefly Books (U.S.) Inc.

Francis, C.M. 1989. A comparison of mist nets and two designs of harp traps for capturing bats. *Journal of Mammalogy.* **70(4)**, 865-870.

Frick, W.F., Pollock, J.F., Hicks, A.C., Langwig, K.E., Reynolds, D.S., Turner, G.G., Butchkoski, C.M., Kunz, T.H. 2010. An emerging disease causes regional population collapse of a common North American bat species. *Science.* **329**, 679.

Fujita, M.S. & Kunz T.H. 1984. *Pipistrellus subflavus. Mammalian Species.* **228**, 1-6.

Gaisler, J. & Chytil, J. 2002. Mark-recapture results and changes in bat abundance at the cave of Na Turoldu, Czech Republic. *Folia Zoologica.* **51**, 1-10.

Gargas, A., Trest, M.T., Christensen, M., Volk, T.J., Blehart, D.S. 2009. *Geomyces destructans* sp. nov. associated with bat white-nose syndrome. *Mycotaxon.* **108.** 147-154.

Glover, A. M. and Altringham, J.D. 2008. Cave selection and use by swarming bat species. *Biological Conservation.* **141**, 1493-1504.

Graham, G. L. 1994. *Bats of the world*. Racine, WI: Western Publishing Company, Inc.

Hargrove, J.W. and Borland, C.H. 1994. Pooled population parameter estimates from mark-recapture data. *Biometrics.* **50**, 1129-1141.

Harvey, M.J., Altenbach, J.S., Best, T.L. 1999. *Bats of the United States*. Little Rock, AR: Arkansas Game and Fish Commission.

Hester, S.G. & Grenier, M.B. 2005. A conservation plan for bats in Wyoming. Wyoming Game and Fish Department, Nongame Program, Lander, WY.

Hester, L. & Myers, P. 2001. "Vespertilionidae". Animal Diversity Web. Accessed July 09, 2011, Retrieved from: http://141.213.176.11/site/accounts/information/Vespertilionidae.html.

Hickey, M.B.C., Fenton, M.B., MacDonald, K.C., Souliere, C. 2001. Trace elements in the fur of bats (Chiroptera:Vespertilionidae) from Ontario and Quebec, Canada. *Bulletin of Environmental Contamination and Toxicology.* **66**, 699-706.

Hill, J.E. & Smith J.D. 1984. *Bats: A Natural History.* Austin, TX: University of Texas Press.

Jones, G., Jacobs, D.S., Kunz, T.H., Willig, M.R., Racey, P.A. 2009. Carpe noctum: The importance of bats as bioindicators. *Endangered Species Research.* **8**, 93-115.

Johnson, S.A., Brack Jr., V., Rolley, R.E. 1998. Overwinter weight loss of Indiana bats (*Myotis sodalis*). *American Midland Naturalist.* **139 (2)**, 255-261.

Kerth, G. 2008. Causes and Consequences of Sociality in Bats. *BioScience.* **58 (8)**, 737-746.

Kerth, G., Kiefer, A., Trappmann, C., Weishaar. 2003. High gene diversity at swarming sites suggests hot spots for gene flow in the endangered Bechstein's bat. *Conservation Genetics.* **4**, 491-499.

Krebs, C.J. 1999. *Ecological Methodology(2nd Edition).* Menlo Park, CA: Addison-Welsey Educational Publishers.

Krebs, J.R. & Davies, N.B. (Eds.) 1997. *Behavioural Ecology: An Evolutionary Approach (4th edition).* Osney Mead, Oxford: Blackwell Science, Inc.

Kuenzi, A.J. and Morrison, M.L. 1998. Detection of bats by mist-nets and ultrasonic sensors. *Wildlife Society Bulletin.* **26 (2)**, 307-311.

Kunz, T.H. 2003. Censusing bats: challenges, solutions, and sampling biases. In: O'Shea, T.J., and M.A. Bogan (eds.). Monitoring trends in bat populations of the United States and territories: problems and prospects. Information and Technology Report **2003-0003**. : U.S. Geological Survey. 9-19.

Kunz, T.H., August, P.V., Burnett, C.D. 1983. Harem social organization in cave roosting *Artibeus jamaicensus* (Chiroptera: Phyllostomidae). *Biotropica.* **15(2)**, 133-138.

Kunz, T.H. & Kurta, A. 1988. Capture methods and holding devices. In: *Ecological and Behavioral Methods for the Study of Bats* (Ed. By T.H. Kunz), pp. 1-30. Washington, D.C.: Smithsonian Institution Press.

Kurta, A. & Baker, R.H. 1990. *Eptesicus fuscus. Mammalian Species.* **356**, 1-10.

Kurz, M.E. 2011. **Species richness, population densities, reproductive behaviors, and sex ratios of the cave-dwelling bats at Dunbar Cave, Clarksville, Montgomery County, Tennessee (Master's Thesis). Austin Peay State University, Clarksville, TN.**

Lamb, J.W. & Wyckoff, G.R. (Eds.) 2010. Cooperative white-nose syndrome monitoring and surveillance plan for Tennessee.

Matthews, L.E. 2005. Dunbar Cave; The showplace of the South. Huntsville, AL; National Speleological Society.

Matthews, L.E. 2011. Dunbar Cave; The showplace of the South. (2^{nd} ed.). Huntsville, AL; National Speleological Society.

Mann, S.L., Steidl, R.J., Dalton, V.M. 2002. Effects of cave tours on breeding *Myotis velifer*. *The Journal of Wildlife Management*. **66**, 618-624.

Mulder, C.P.H., Bazeley-White, E., Dimitrakopoulos, P.G., Hector, A., Scherer-Lorenzen, M., Schmid, B. 2004. Species evenness and productivity in experimental plant communities. *OIKOS.* **107**, 50-63.

McCracken, G.F. 2003. Estimates of population size in summer colonies of Brazilian free-tailed bats (*Tadarida brasiliensis*). In: O'Shea, T.J., and M.A. Bogan (eds.).

Monitoring trends in bat populations of the United States and territories: problems and prospects. *Information and Technology Report* **2003-0003**. : U.S. Geological Survey. 21-30.

National Oceanic and Atmospheric Administration. 2012, December 6. Past Monthly Climate Report. Retrieved from: http://www.nws.noaa.gov/climate/index.php?wfo=ohx

O'Farrell, M. J. and Gannon, W.L. 1999. A comparison of acoustic versus capture techniques for the inventory of bats. *Journal of Mammalogy*. **80**, 24-30.

O'Shea, T.J., and M.A. Bogan (Eds.). 2003. Monitoring trends in bat populations of the United States and territories: problems and prospects. *Information and Technology Report* 2003-003: U.S. Geological Survey.

O'Shea, T.J., Bogan, M.A., Ellison, L.E. 2003. Monitoring trends in bat populations of the United States and territories: status of the science and recommendations for the future. *Wildlife Society Bulletin*, **31**, 16-29.

O'Shea, T.J., Ellison, L.E., Stanley, T.R. 2004. Survival estimation in bats: Historical overview, critical appraisal, and suggestions for new approaches. In: *Sampling Rare and Elusive Species: Concepts, Designs, and Techniques for Estimating Population Parameters*. (Ed. By W.L. Thompson). pp. 297-336. Washington, D.C.: Island Press.

Owen, S.F., Menzel, M. A., Ford, M.W. Chapman, B.R. Miller, K.V., Edwards, J.W., Wood, P.B. 2003. Home-range size and habitat used by the Northern Myotis (*Myotis septentrionalis*). *American Midland Naturalist.* **150**. 352-359.

Parsons, K., Jones, G., Davidson-Watts, I., Greenaway, F. 2003. Swarming of bats at underground sites in Britain-implications for conservation. *Biological Conservation.* **111**, 63-70.

Pettigrew, J.D. Dreher, B. Hopkins, C.S., McCall, M.J., Brown, M. 1988. Peak density and distribution of ganglion cells in the retinae of Microchiropteran bats: Implications for visual acuity. *Brain, Behavior, and Evolution.* **32**, 39-56.

Rabinowitz, A. 1981. Thermal preference of the Eastern Pipistrelle bat (*Pipistrellus subflavus*) during hibernation. *Journal of the Tennessee Academy of Science.* **56(4),** 113-114.

Reichard, J.D. & Kunz, T.H. 2009. White-nose syndrome inflicts lasting injuries to the wings of little brown myotis (*Myotis lucifugus). Acta Chiropterologica.* **11(2),** 457-464.

Rivers, N.M., Butlin, R.K., Altringham, J.D. 2006. Autumn swarming behavior of Natterer's bats in the UK: Population size, catchment area and dispersal. *Biological Conservation.* **127**, 215-226.

Sandel, J.K., Benatar, G.R., Burke, K.M., Walker, C.W., Lacher Jr., T.E., Honeycutt, R. 2001. Use and selection of winter hibernacula by the Eastern Pipistrelle (*Pipistrellus subflavus*) in Texas. *Journal of Mammalogy.* **82,** 173-178.

Scheel, D. Vincent, T.L.S. Cameron, G.N. 1996. Global warming and the species richness of bats in Texas. *Conservation Biology* **10**, 452-464.

Sheffield, S.R. 1992. Guidelines for the protection of bat roosts. *Journal of Mammalogy* . **73(3)**, 707.

Shump, Jr., K.A. & Shump, A.U. 1982. *Lasiurus borealis. Mammalian Species.* **183**. 1-6.

Simek, J.F., Douglas, J.C., Wallace, A. 2007. Ancient cave art at Dunbar Cave State Natural Area. *The Tennessee Conservastionist.* **September/October**, 24-26.

Sendor, T. & Simon, M. 2003. Population dynamics of the pipistrelle bat: effects of sex, age, and winter weather on seasonal survival. *Journal of Animal Ecology.* **72**. 308-320.

Tennessee Department of Environment and Conservation. 2011, June 20. *Dunbar Cave Class One Scenic-Recreational State Natural Area.* Retrieved from: http://www.tn.gov/environment/na/natareas/dunbar/

Thomas, D.W. Dorais, M. Bergeron, J. 1990. Winter energy budgets and cost of arousal for hibernating little brown bats, *Myotis lucifugus*. *Journal of Mammalogy*. **71**:475-479.

Thompson, W.L. (Ed.) 2004. *Sampling rare or elusive species: Concepts, designs, and techniques, for estimating population parameters.* Washington, DC: Island Press.

Tuttle, M.D. 2006. *America's Neighborhood Bats: Understanding and Learning to Live in Harmony with Them.* Austin, TX: University of Texas Press.

United States Fish and Wildlife Service. 2011, June. *Listing a species as threatened or endangered.* Retrieved from: http://www.fws.gov/endangered/esa-library/pdf/listing.pdf

United States Fish and Wildlife Service. 2012, January 17. *North American bat death toll exceeds 5.5 million from White Nose Syndrome*. Retrieved from: http://www.fws.gov/whitenosesyndrome/

United States Fish and Wildlife Service (b). 2012, June 25. *National White-Nose Syndrome decontamination protocol-Version 06.25.2012.* Retrieved from: http://whitenosesyndrome.org/sites/default/files/resource/national_wns_revise_final_6.25.12.pdf.

United States Geological Survey. 2012, May 30. *White-Nose Syndrome (WNS).* Retrieved from: http://www.nwhc.usgs.gov/disease_information/white-nose_syndrome/.

Veith, M., Beer, N., Kiefer, A., Johannesen, J., Seitz, A. 2004. The role of swarming sites for maintaining gene flow in the brown long-eared bat (*Plecotus auritus*). *Heredity*. **2004**, 1-8.

Weller, T.J. & Lee, D.C. 2007. Mist net effort required to inventory a forest bat species assemblage. *Journal of Wildlife Management.* **71**: 251-257.

Willis, C.K.R., Brigham, R.M. 2003. Defining torpor in free-ranging bats: Experimental evaluation of external temperature-sensitive radiotransmitters and the concept of active temperature. *Journal of Comparative Physiology B.* **173**, 379-389.

Willis, C.K.R., Brigham, R.M. 2004. Roost switching, roost sharing, and social cohesion: Forest- dwelling big brown bats, *Eptesicus fuscus*, conform to the fission-fusion model. *Animal Behaviour.* **68**, 495-505.

Willis, C.K.R., Brigham, R.M., Geiser, F. 2006. Deep, prolonged torpor by pregnant, free-ranging bats. *Naturwissenschaften.* **93**, 80-83.

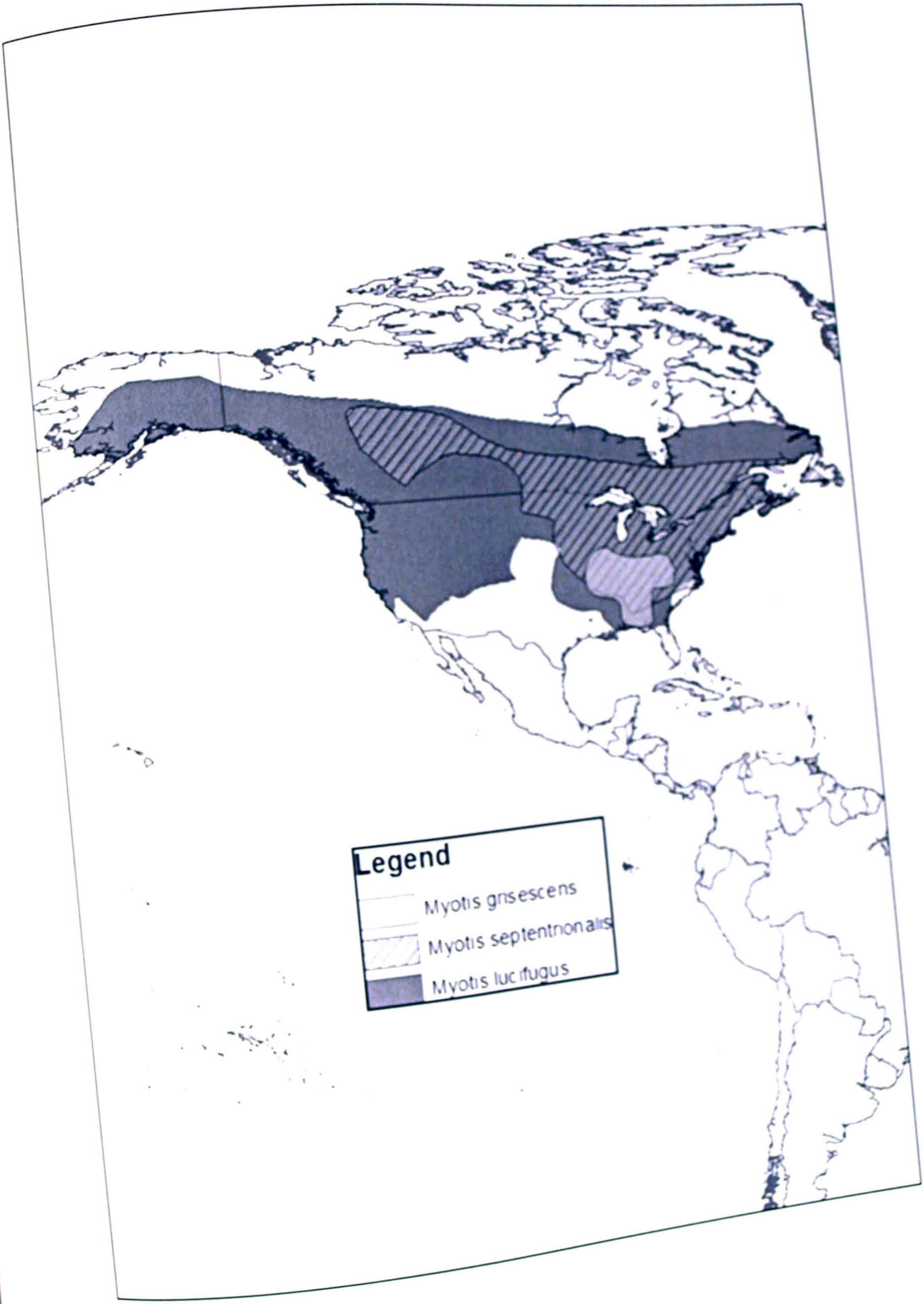
Weller, T.J. and Lee, D.C. 2007. Mist net effort required to inventory a forest bat species assemblage. *The Journal of Wildlife Management.* **71**, 251-257.

Wojciechowski, M.S., Jefimow, M., Tegowska, E. 2007. Environmental conditions, rather than season, determine torpor use and temperature selection in large mouse-eared bats (*Myotis myotis*). *Comparative Biochemisty and Physiology.* **147**, 828-840.

APPENDICES

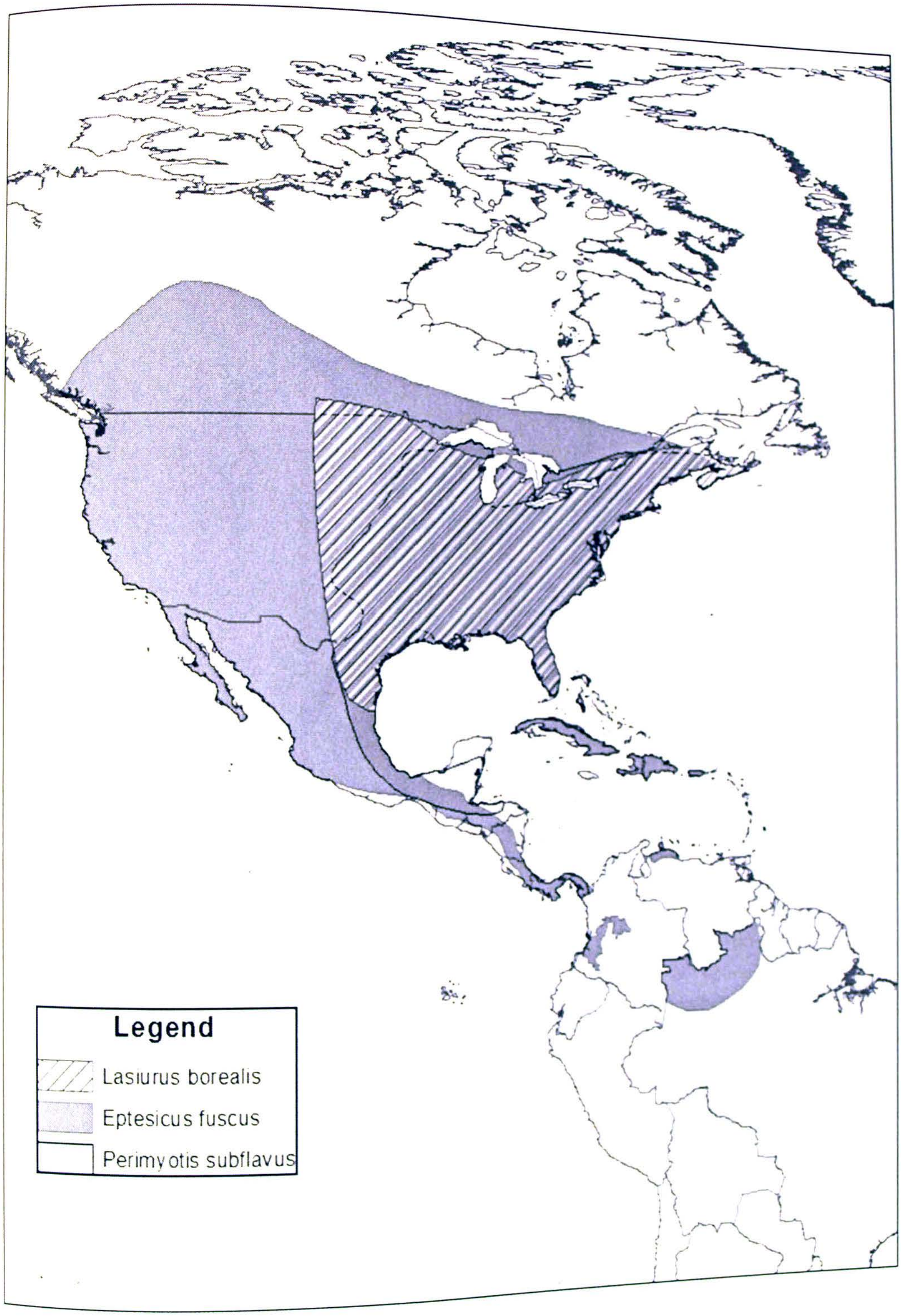
APPENDIX A

Geographic Distribution of Myotis Bat Species at Dunbar Cave State Natural Area



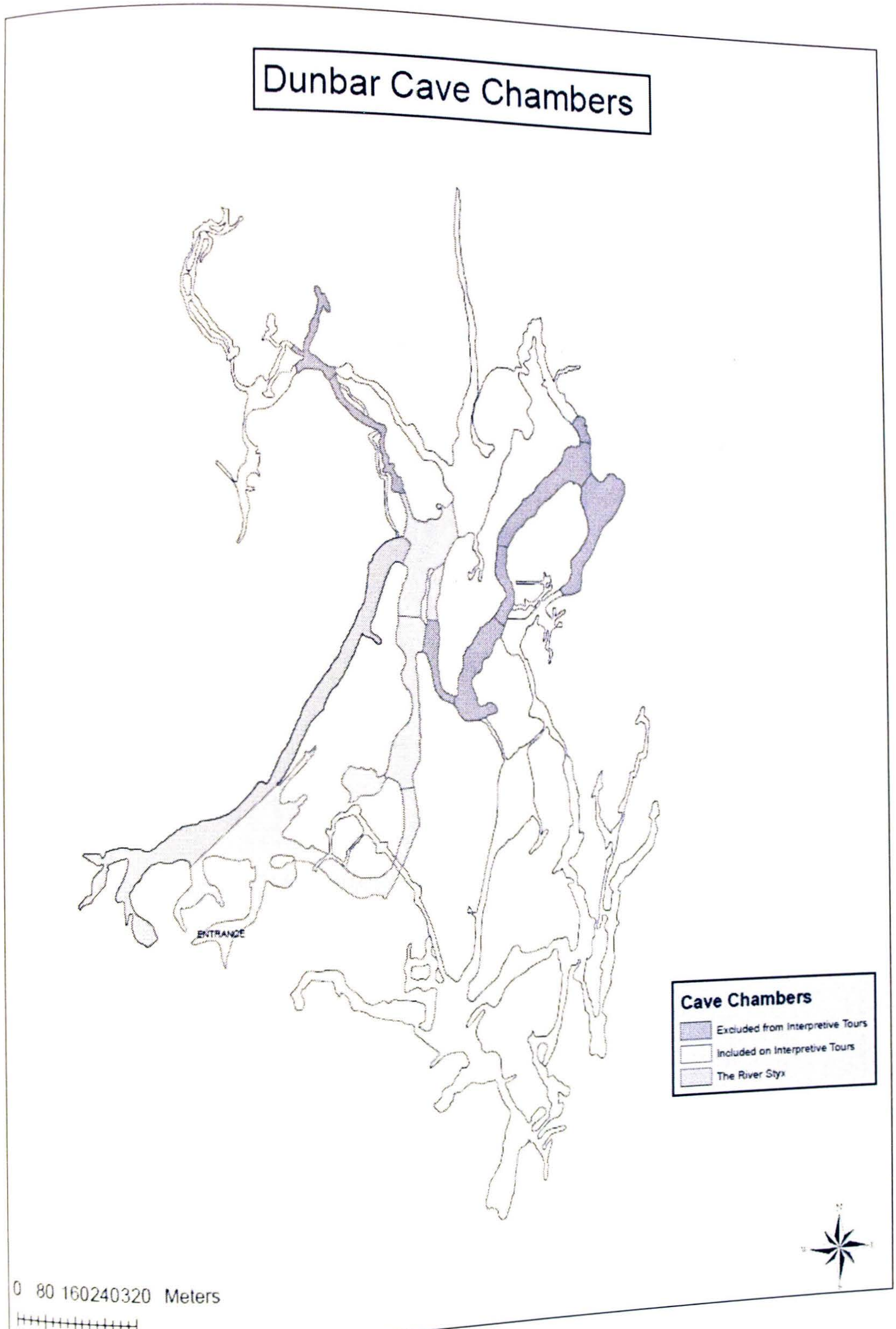
APPENDIX B

Geographic Distribution of Non-Myotis Bat Species at Dunbar Cave State Natural Area



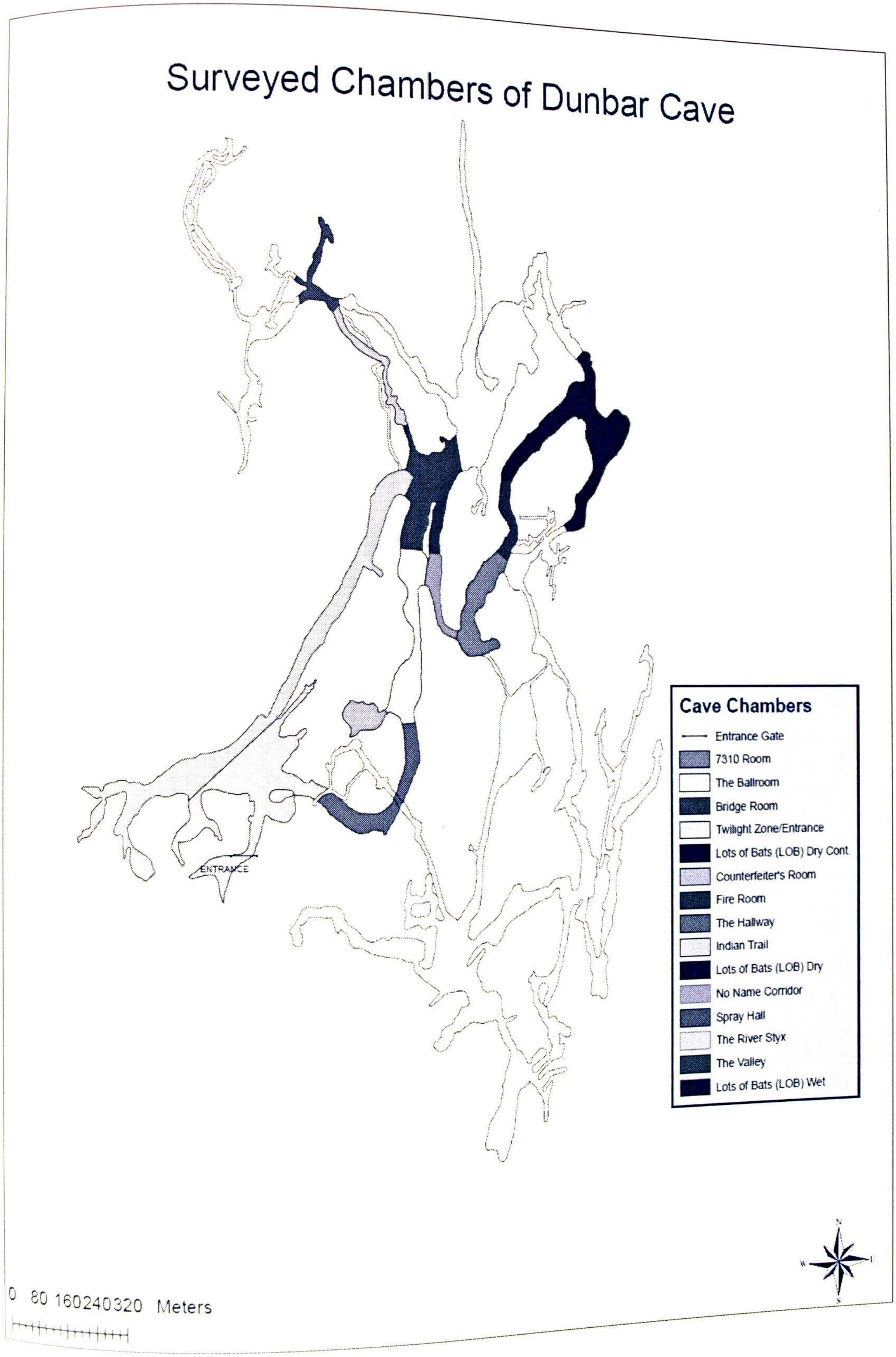
APPENDIX C

Chambers of Dunbar Cave Included on Bat Surveys and Interpretive Cave Tours



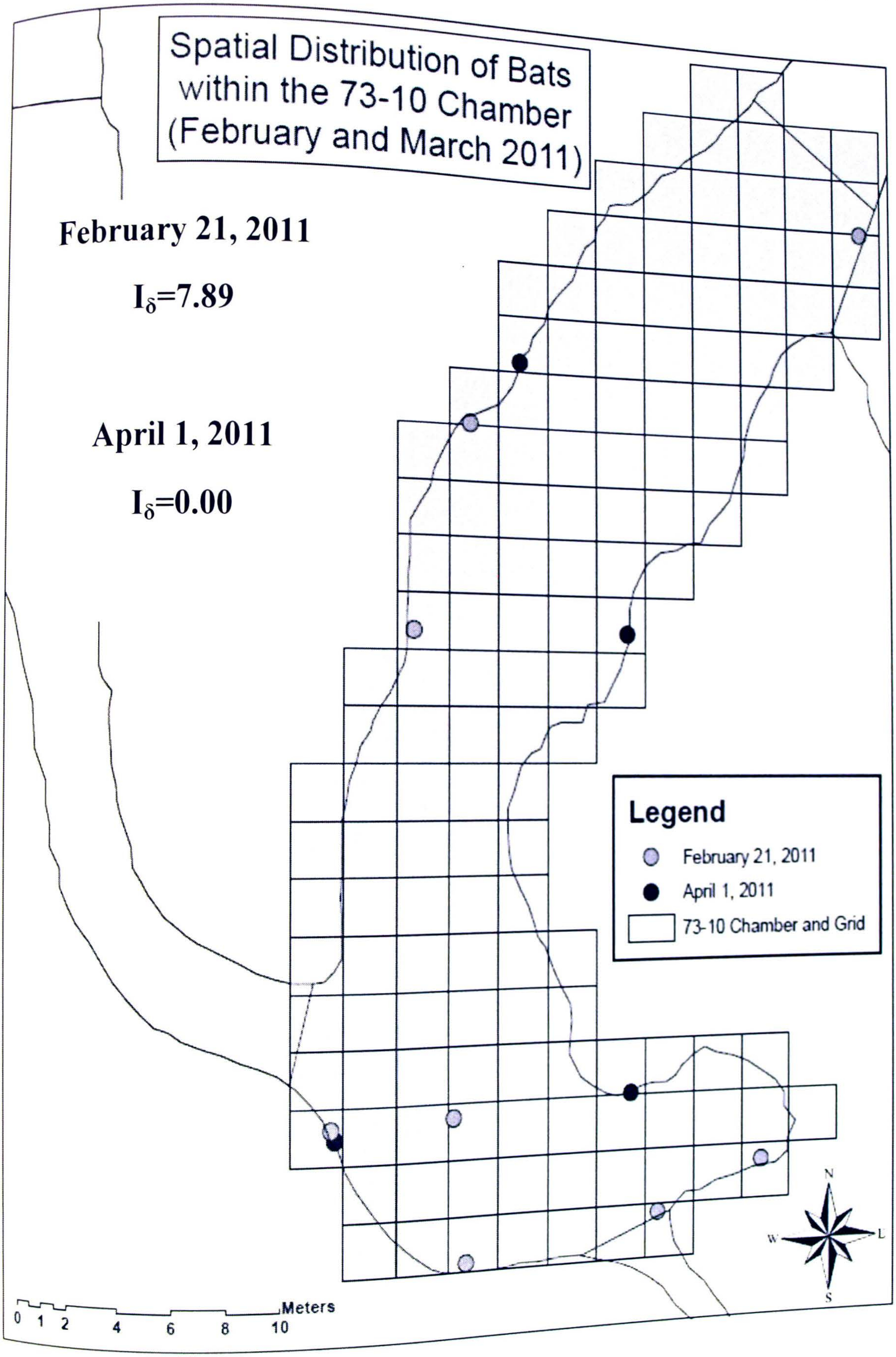
APPENDIX D

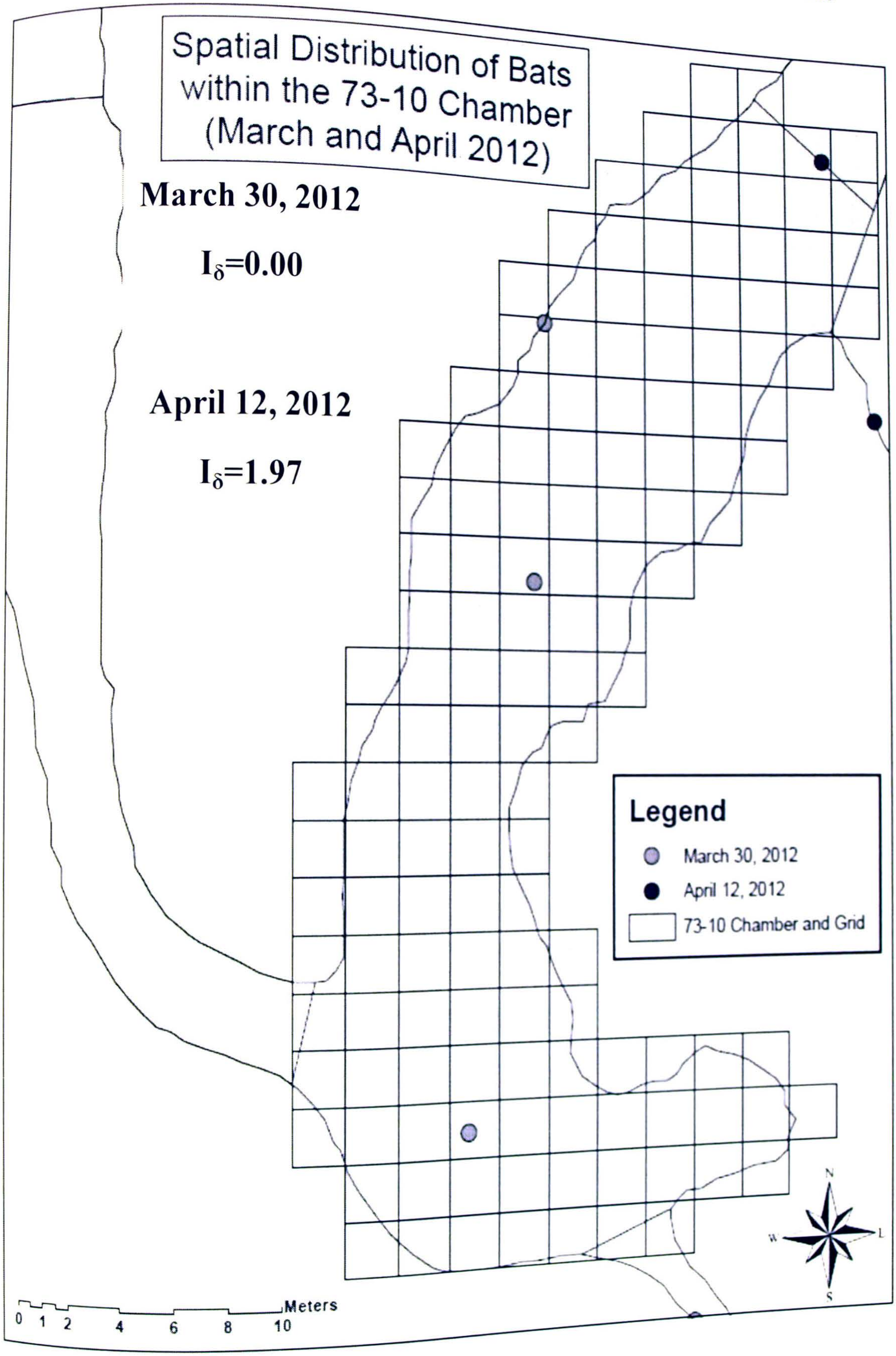
Example Base Map of Dunbar Cave Used for GIS Mapping of Bat Surveys

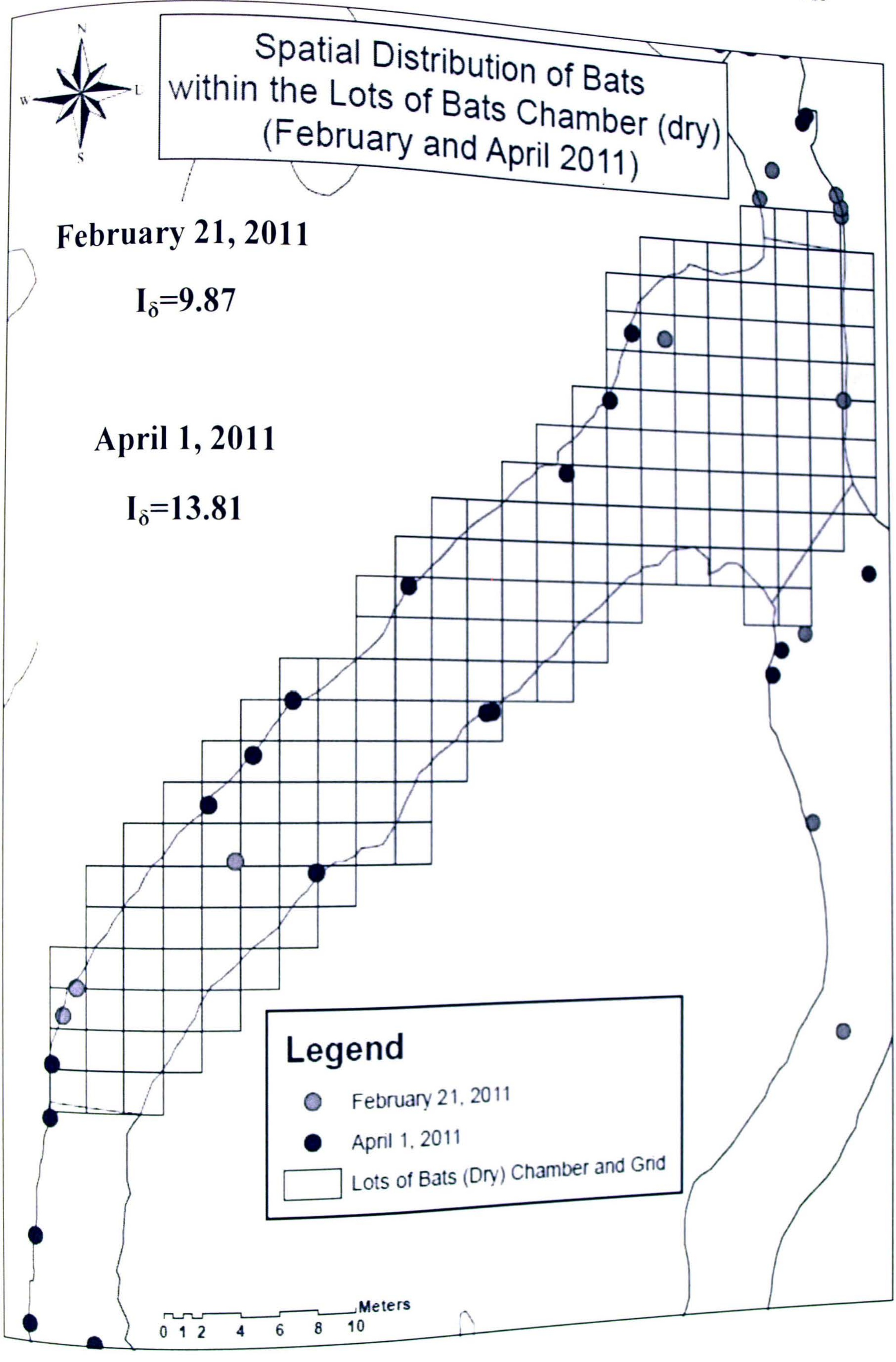


APPENDIX E

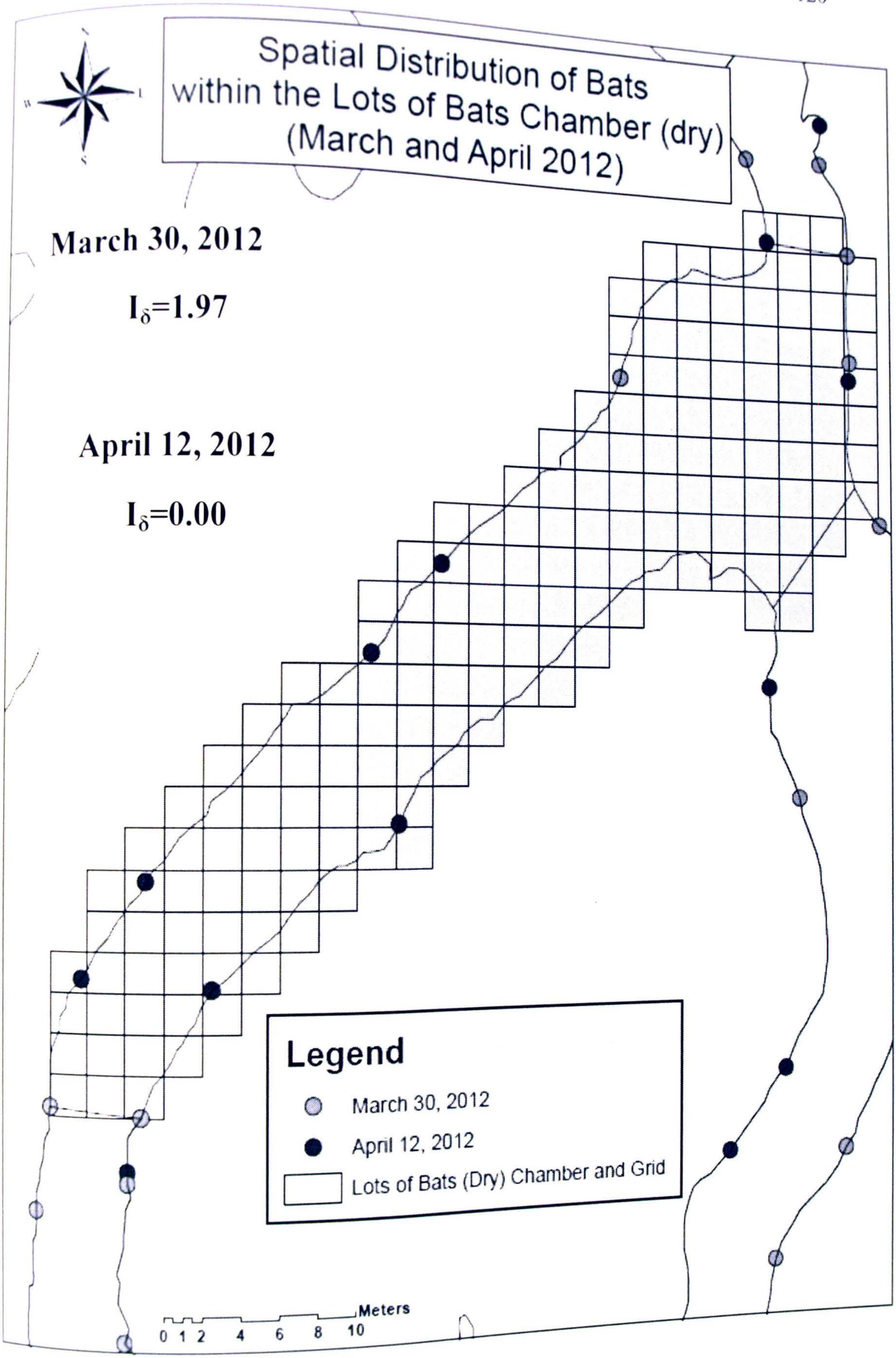
Maps of Cave Chambers and Spatial Distribution Grids with Standardized Morisita Index of Dispersion (Iδ) Values for Investigation of Spatial Distribution of the Bat Population within Dunbar Cave

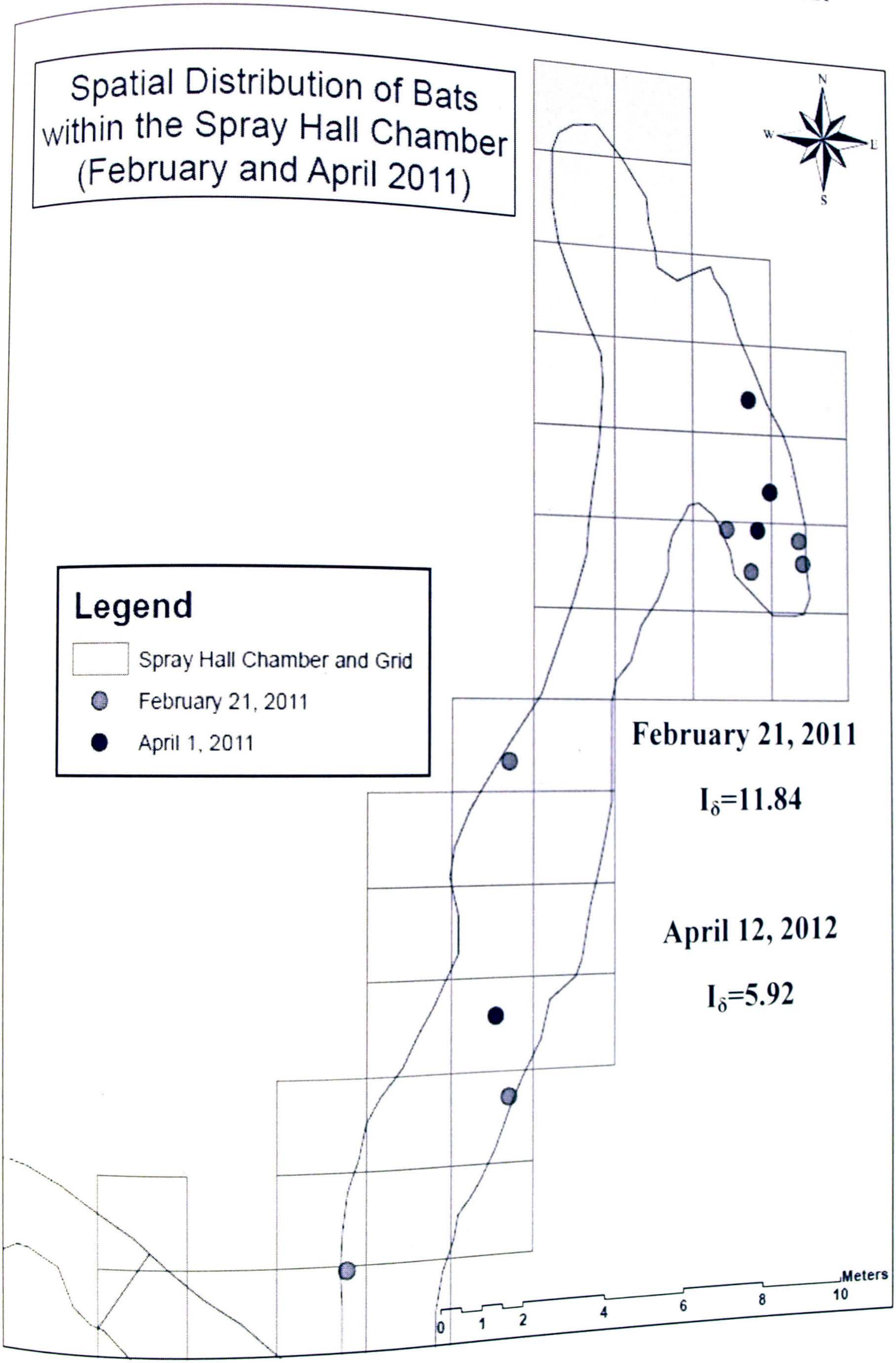

Spatial Distribution of Bats within the 73-10 Chamber (February and March 2011)
February 21, 2011
I_δ=7.89
April 1, 2011
I_δ=0.00
Legend
February 21, 2011
April 1, 2011
73-10 Chamber and Grid
N
W
L
S
0 1 2 4 6 8 10 Meters


Spatial Distribution of Bats within the 73-10 Chamber (March and April 2012)
March 30, 2012
Iδ=0.00
April 12, 2012
Iδ=1.97
Legend
March 30, 2012
April 12, 2012
73-10 Chamber and Grid
0 1 2 4 6 8 10
Meters
N
S
W
L



Spatial Distribution of Bats
within the Lots of Bats Chamber (dry)
(February and April 2011)
N
S
W
February 21, 2011
Iδ=9.87
April 1, 2011
Iδ=13.81
Legend
February 21, 2011
April 1, 2011
Lots of Bats (Dry) Chamber and Grid
Meters
0 1 2 4 6 8 10


Spatial Distribution of Bats
within the Lots of Bats Chamber (dry)
(March and April 2012)
March 30, 2012
I_δ=1.97
April 12, 2012
I_δ=0.00
Legend
March 30, 2012
April 12, 2012
Lots of Bats (Dry) Chamber and Grid
Meters
0 1 2 4 6 8 10



Spatial Distribution of Bats within the Spray Hall Chamber (February and April 2011)
N
W
E
S
Legend
Spray Hall Chamber and Grid
February 21, 2011
April 1, 2011
February 21, 2011
Iδ=11.84
April 12, 2012
Iδ=5.92
Meters
0
1
2
4
6
8
10

APPENDIX F

Acoustic data from 2011 and 2012

Acoustic Data Collected During Capture Events 2011								
Date	**Type**	**Location**	**EPFU**	**LANO**	**LABO**	**LACI**	**MYGR**	**MYLU**
5/31/2011	Harp	Amphitheater	91	21	11	0	0	13
5/31/2011	Harp	Over Lake	88	11	174	0	0	9
6/28/2011	Harp	Amphitheater	24	4	24	0	0	2
7/27/2011	Harp	Concession Stand	51	0	9	0	0	1
7/27/2011	Harp	Over Lake	89	5	87	2	0	12
8/8/2011	Harp	Amphitheater	104	1	21	0	14	1
8/8/2011	Harp	Over Lake	67	15	36	0	13	8
6/29/2011	Mist	Trail above cave	3	0	1	0	0	0
7/20/2011	Mist	Forest above cave	1	0	0	0	0	0
7/28/2011	Mist	Between lake and road	0	2	4	1	0	0
8/9/2011	Mist	on trail near stairs to cave	6	3	20	0	0	5

Acoustic Data Collected During Capture Events 2011								
Date	**Type**	**Location**	**NYHU**	**MYSO**	**PESU**	**UNKN**	**MYOTIS**	**Total**
5/31/2011	Harp	Amphitheater	92	0	39	6	18	275
5/31/2011	Harp	Over Lake	8	0	47	4	13	342
6/28/2011	Harp	Amphitheater	88	0	351	4	4	498
7/27/2011	Harp	Concession Stand	20	0	718	3	41	822
7/27/2011	Harp	Over Lake	91	0	1169	22	158	1550
8/8/2011	Harp	Amphitheater	1	1	875	3	17	1030
8/8/2011	Harp	Over Lake	31	1	747	13	24	931
6/29/2011	Mist	Trail above cave	0	0	5	5	0	14
7/20/2011	Mist	Forest above cave	1	0	0	0	0	2
7/28/2011	Mist	Between lake and road	4	0	63	0	0	74
8/9/2011	Mist	on trail near stairs to cave	25	0	207	2	14	274

All acoustic data from 2011was analyzed using Bat Call Identification software from Bat Call Identification, Inc. Springfield, MO, USA.

Acoustic Files Recorded During two Harp Trapping Events at DCSNA-2012

DATE	ANABAT # 80665 (SWAN LAKE)		ANABAT # 80685 (CONCESSION STAND/AMPHITHEATER)	
	Prominent Bat Species	**# Files Detected**	**Prominent Bat Species**	**# Files Detected**
May 18, 2012	*Eptesicus fuscus*	23	*Eptesicus fuscus*	10
	*Lasionycteris noctivagans**	12	*Lasionycteris noctivagans**	2
	Lasiurus borealis	147	*Lasiurus borealis*	129
	Lasiurus cinereus(2)	25	*Lasiurus cinereus*	17
	Myotis grisescens	13	*Myotis grisescens*	4
	Myotis septentrionalis	2	*Myotis sodalis**	1
	*Myotis sodalis**	1	*Nycticeius humeralis**	2
	*Nycticeius humeralis**	6	*Perimyotis subflavus*	14
	Perimyotis subflavus	30	Unknown	72
	Unknown	94	**Total Call Files**	251
	Total Call Files	353		
	Prominent Bat Species	**# Files Detected**	**Prominent Bat Species**	**# Files Detected**
August 8, 2012	*Eptesicus fuscus*	53	*Eptesicus fuscus*	27
	Lasionycteris noctivagans	2	*Lasionycteris noctivagans**	4
	Lasiurus borealis	725	*Lasiurus borealis*	598
	Lasiurus cinereus	8	*Lasiurus cinereus*(2)	1
	Myotis austroriparius	1	*Myotis austroriparius*	
	Myotis grisescens	232	*Myotis grisescens*	636
			Myotis leibii	
	Myotis lucifugus	1	*Myotis lucifugus*	1
	Myotis septentrionalis	1	*Myotis septentrionalis*	1
	Myotis sodalis	1	*Myotis sodalis*	
	Nycticeius humeralis	9	*Nycticeius humeralis**	10
	Perimyotis subflavus	272	*Perimyotis subflavus*	265
	Unknown	161	Unknown	138
	Total Call Files	1413	**Total Call Files**	1681

Species listed were detected by Echoclass Acoustic ID Program as the Prominent Bat Species per file

Each species was assigned a quantitative output indicating % probability of presence ranging from 0-3

3 = highest probability (99%)

2 = 95%

1= 90%

0 = no presence

All species listed were assigned a 3 unless otherwise indicated

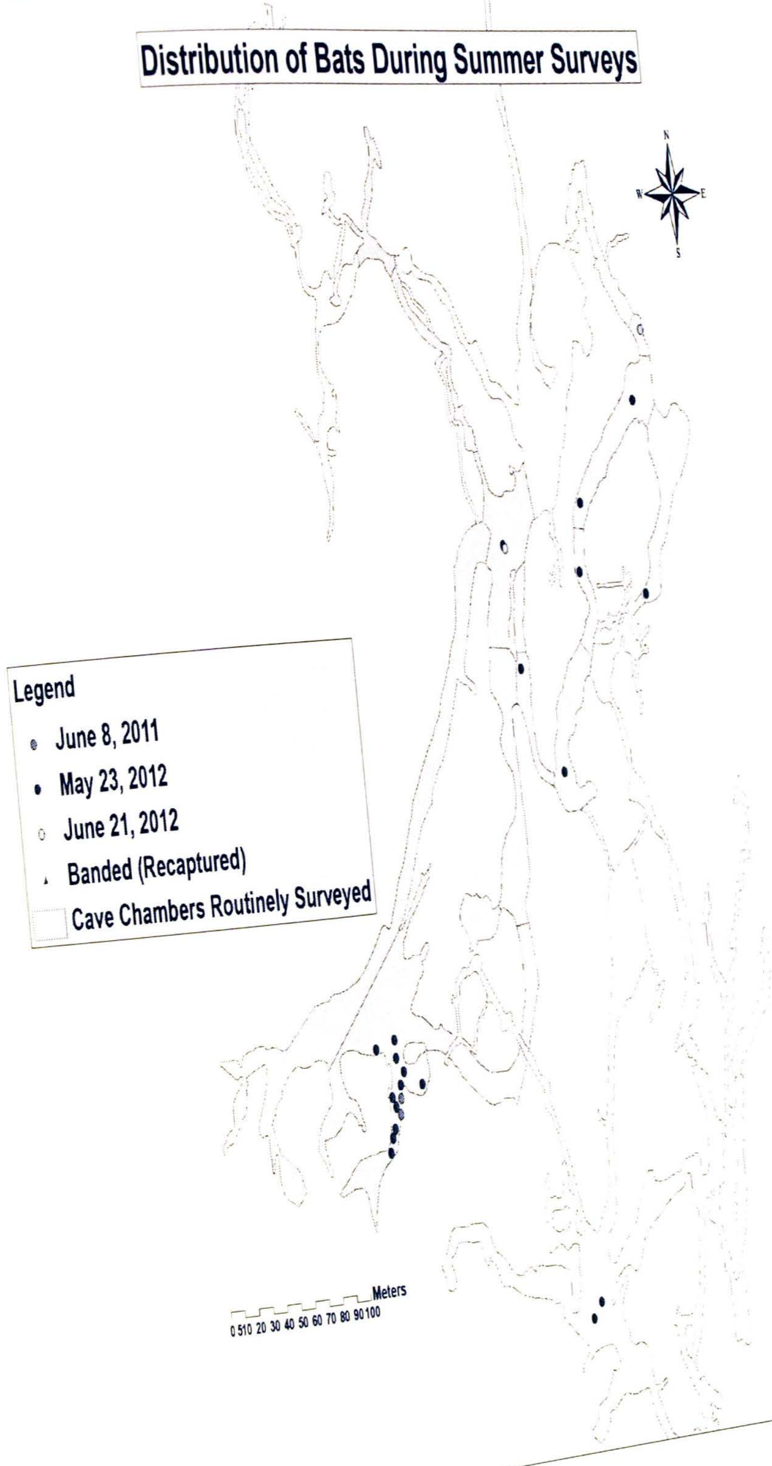
*= no presence

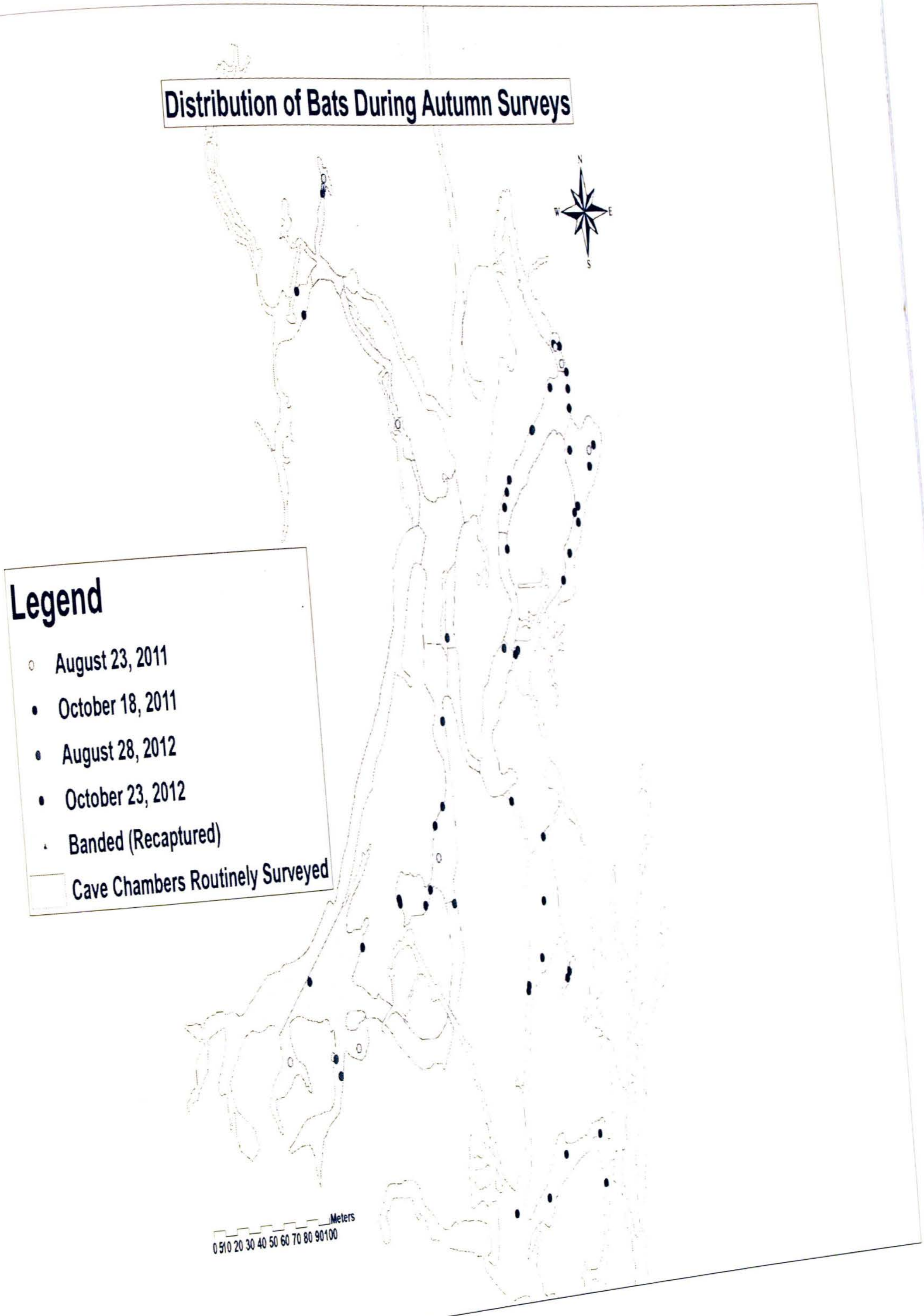
Both Anabat® Units were equipped with Anabat® green high mount microphones, and set up at a 45° angle pointing out over the area of interest. Due to the close proximity of the units overlap in recording may have occurred.

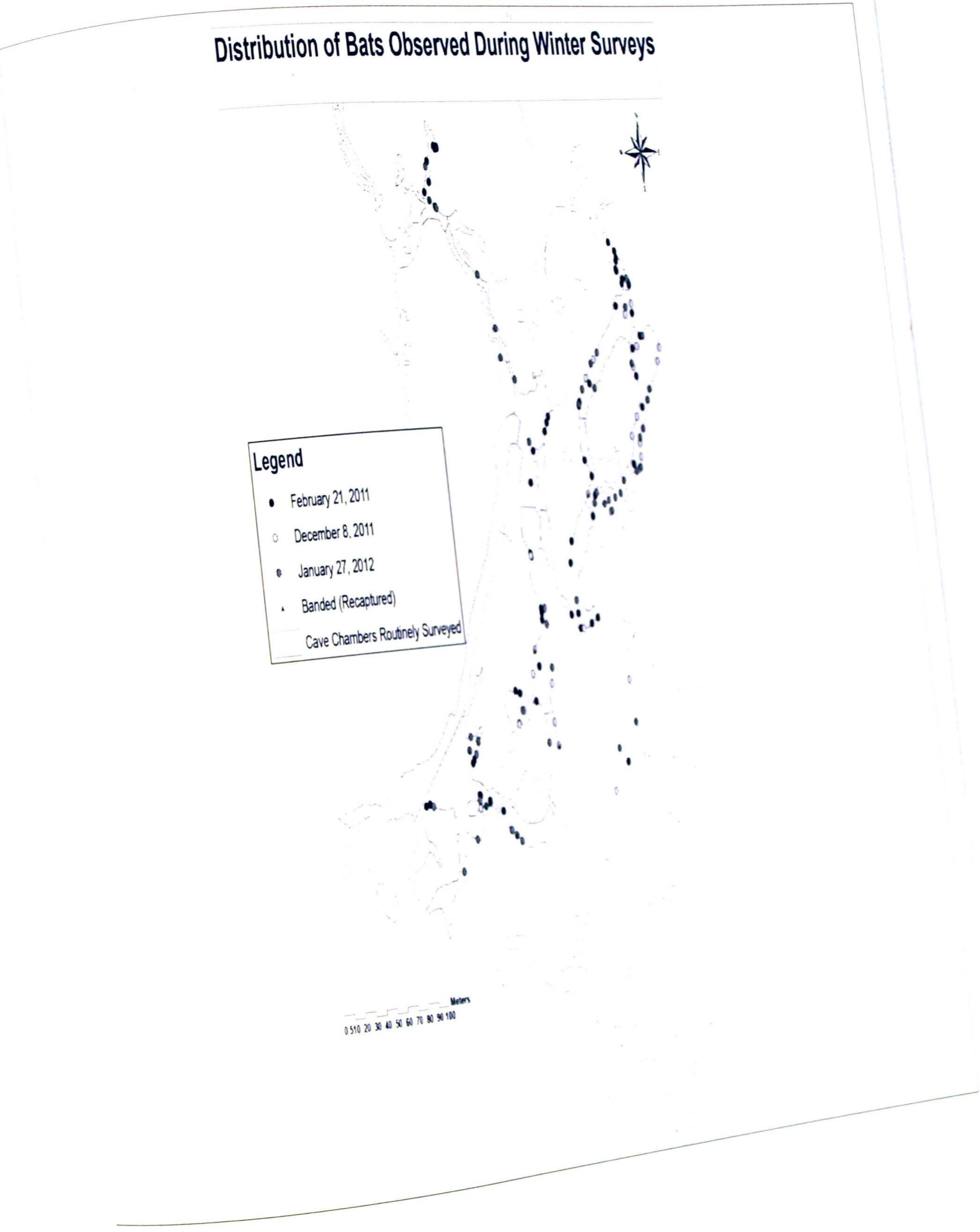
APPENDIX G

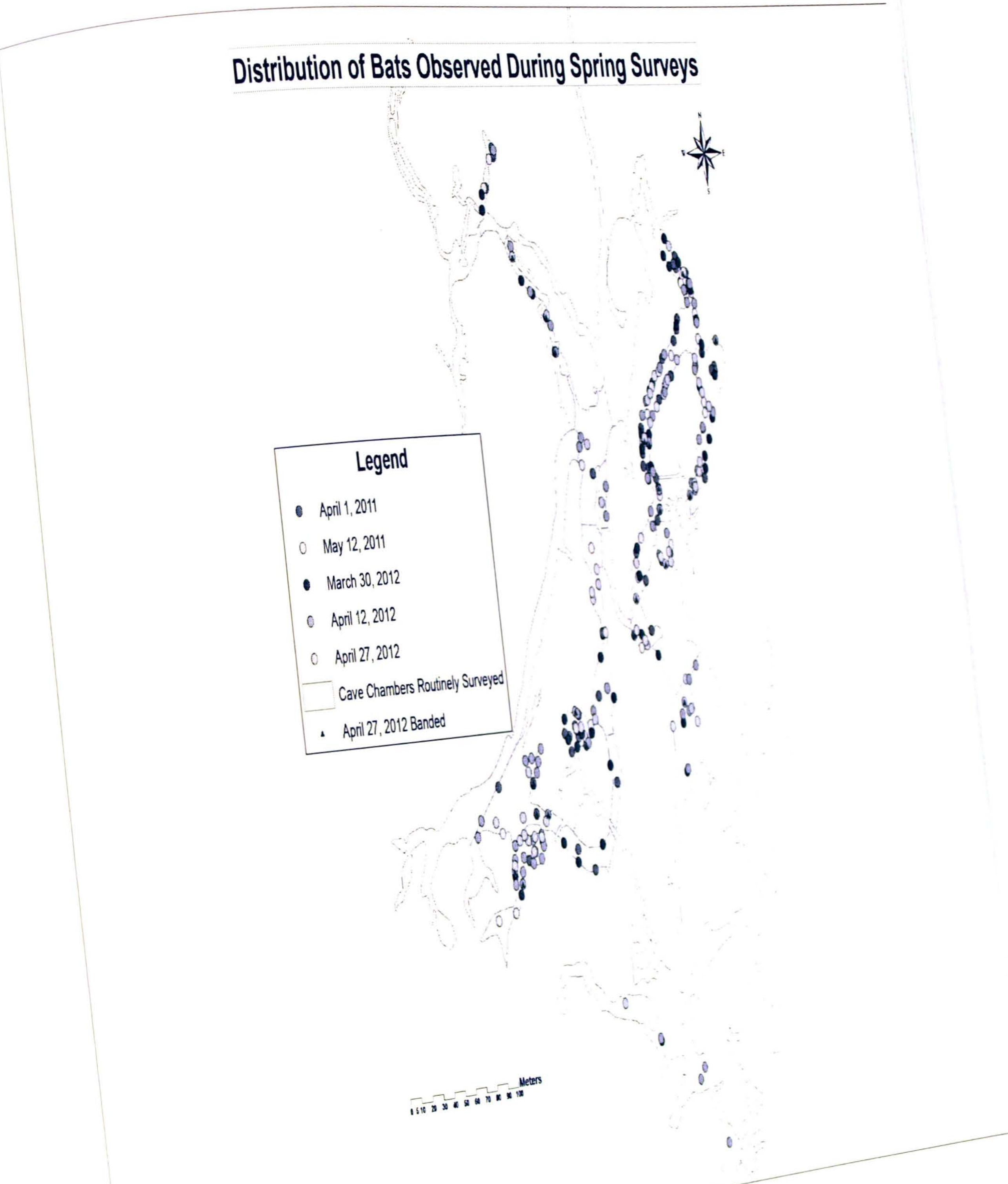
Seasonal Cave Chamber Selection within Dunbar Cave

(2011-2012)


Distribution of Bats During Summer Surveys
N
W
E
S
Legend
June 8, 2011
May 23, 2012
June 21, 2012
Banded (Recaptured)
Cave Chambers Routinely Surveyed
Meters
0 5 10 20 30 40 50 60 70 80 90 100


Distribution of Bats During Autumn Surveys
Legend
August 23, 2011
October 18, 2011
August 28, 2012
October 23, 2012
Banded (Recaptured)
Cave Chambers Routinely Surveyed
N
S
E
W
Meters
0 5 10 20 30 40 50 60 70 80 90 100


Distribution of Bats Observed During Winter Surveys
Legend
February 21, 2011
December 8, 2011
January 27, 2012
Banded (Recaptured)
Cave Chambers Routinely Surveyed
Meters
0 5 10 20 30 40 50 60 70 80 90 100


Distribution of Bats Observed During Spring Surveys
Legend
April 1, 2011
May 12, 2011
March 30, 2012
April 12, 2012
April 27, 2012
Cave Chambers Routinely Surveyed
April 27, 2012 Banded
Meters

APPENDIX H

Photographs of Myotis lucifugus

Figure 16. Three *Myotis lucifugus* found within the Hallway chamber of Dunbar Cave. Note the white powdery substance, reminiscant of *Geomyces destructans*.

Figure 17. An individual *Myotis lucifugus* caught during harp trapping. Note extensive wing discoloration, associated with *Geomyces destructans*.

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Signature Date