

THESIS

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Title of Thesis: Comparison of six monitoring techniques for the
reptiles and amphibians at Chincoteague National
Wildlife Refuge

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ABSTRACT

Title of Thesis: Comparison of six monitoring techniques for the reptiles and
amphibians at Chincoteague National Wildlife Refuge

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Degree and Year: Master of Science 2000

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I compared the cost, time, and sampling effectiveness of aural surveys, basking surveys, time-constrained surveys, cover boards, road surveys, and pitfall trap arrays at Chincoteague National Wildlife Refuge (CHNWR) from May 1998-August 2000. The objectives of my study were to (1) inventory amphibians and reptiles; (2) compare species richness and relative abundance of herpetofauna at CHNWR; (3) compare the cost and effectiveness of sampling techniques; and (4) develop a long-term monitoring protocol for amphibians and reptiles at CHNWR.

I found a significant effect ($p=0.016$) among techniques for the number of species detected. I compared 6 techniques using the Shannon-Weaver species diversity index and found that road surveys had the highest species diversity ($H^i=1.914$). Pitfall trap arrays were the most expensive technique (\$2953) and road surveys were the least expensive (\$485).

For specific information on amphibians I recommend using aural surveys, for turtles basking surveys, and for snakes road surveys. Aural, basking, and road

surveys provide presence or absence data to monitor population trends over time. To develop population estimates, pitfall trap arrays, cover board surveys, and time-constrained searches may be used. I found that road surveys were the most cost effective and yielded the largest number of species. I suggest that road surveys may be the most appropriate technique for long-term monitoring at CHNWR.

COMPARISON OF SIX MONITORING TECHNIQUES FOR THE REPTILES
AND AMPHIBIANS AT CHINCOTEAGUE NATIONAL WILDLIFE REFUGE

by

Holly Allison Toadvine

Thesis submitted to the Faculty of the Graduate School
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DEDICATION

In Loving Memory of my father

Theodore Albert Toadvine Sr.

for inspiring my love for wildlife and their habitats

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LIST OF ABBREVIATIONS

Chincoteague National Wildlife Refuge (CHNWR)

National Park Service (NPS)

United States Fish and Wildlife Service (USFWS)

Moist Soil Management Units (MSMU)

Snout to Vent Length (SVL)

Total Length (TL)

INTRODUCTION

Amphibians and reptiles are important components in many ecosystems providing food resources for a variety of species. Historically, wildlife managers have used their resources to monitor and manage economically important game species such as white-tailed deer and waterfowl, rather than non-game species such as amphibians and reptiles. However, the significance of non-game species was recognized with passage of the Federal Land Policy and Management Act of 1976 that required monitoring and management of non-game species (Jones 1986). Compared with other vertebrate classes such as birds, fish, or mammals, there is little published information on how to monitor amphibian and reptile populations, the cost of these methods, or their relative efficiency.

Amphibians and reptiles are challenging to study in the field because they possess several characteristics that make many species difficult to locate. Some of these characteristics include long periods of time spent underground, lack of vocalizations (except breeding anurans), nocturnal activity, cryptic coloration, low population densities, inaccessible habitats, patchy distributions and seasonal variations in detectability (Pough et al. 1998). Despite these challenges, information about amphibians and reptiles is important to natural resources managers because these animals provide many useful functions in natural ecosystems (Record 1995). Amphibians and reptiles serve as food resources for fish, birds, and mammals and may also provide an indication of the relative health of the ecosystem (Jones 1986, Heyer et al. 1994).

One reason it is important to evaluate the status of non-game species such as amphibians and reptiles is that they are sensitive to changes in water quality and quantity, habitat alterations, pollution, and weather conditions (Hall 1980, Hine 1982). By monitoring the response of amphibian and reptile populations, the overall health of habitats may be assessed, and management actions may be evaluated. For example management actions on Fish and Wildlife Service lands that may affect amphibian and reptile populations include manipulation of habitats, prescribed burns, clearing downed logs, mowing, aerial herbicide spraying, and water level regimes (Record 1995, USFWS 1999).

Another reason information on non-game species is important is that during the past decade researchers have documented worldwide declines in amphibian populations, in some cases to the point of extinction (Wake 1991). Explanations for the declines include habitat destruction, introductions of non-native species, pollution, acid rain, increased UV-B radiation from ozone depletion, and global climate change (Pechmann et al. 1991, Wake 1991, Blaustein 1994, McCoy 1994, Pough et al. 1998). However, Pechmann et al. (1991) suggested that some population declines may be due to natural rather than anthropogenic causes. In addition, reported deformities and malformations of many anuran species have heightened the wildlife managers need for population trend information (Heyer et al. 1994, Welsh and Hodgson 1997). Without standardized techniques to monitor populations it is difficult to determine the cause of population declines. Scientists, resource managers, and the public seek worldwide and nationwide monitoring of non-game species (Welsh and Hodgson 1997).

In response to concerns about worldwide declines, Smith (1995) initiated the investigation of the amphibian populations on the Delmarva Peninsula. The first study of the reptiles and amphibians of Assateague and Chincoteague Islands was conducted by Fowler in 1925 when he recorded two species on Assateague Island, the eastern hognose snake (*Heterodon platirhinos*) and the eastern mud turtle (*Kinosternon subrubrum subrubrum*) (Table 1). Also in 1925, Brady found the Loggerhead sea turtle (*Caretta caretta*) and the northern black racer (*Coluber constrictor constrictor*) on Assateague, and Fowler's toad (*Bufo woodhousii fowleri*) on Assateague and Chincoteague. Since 1925, a total of 7 amphibians and 15 reptiles have been found at Chincoteague National Wildlife Refuge (CHNWR) (Conant et al. 1990). The last study to explicitly focus on the herpetofauna of Assateague Island National Seashore (AINS) was reported by Mitchell et al. (1993).

A variety of techniques are used to sample amphibian and reptile populations. Some of the more commonly used techniques include aural surveys, basking surveys, road surveys, time-constrained searches, pitfall trap arrays, and cover boards (Jones 1986, Heyer et al. 1994).

Aural surveys

Male amphibians vocalize for a variety of reasons including territory defense, to attract females, or to compete with other males for females (Wells 1977, Pough et al. 1998). Sampling of amphibian breeding sites generally involves either visual or aural surveys. Several factors may influence the effectiveness of aural surveys including weather conditions (especially temperature), time of year, time of day,

breeding season, breeding type, and proximity to a breeding site (Wells 1977, USFWS 1999). In many species breeding activity is dependent on climate and availability of breeding sites that affects the length of the calling season (Wells 1977, Cooperrider et al. 1986). Many summer breeders are active for 2 to 3 months. Explosive breeders would typically be found only when seasonal rains fluctuate with the availability of temporary rain pools or other ephemeral habitats (Wells 1977, Heyer et al. 1994, Pough et al. 1998).

Foley and Smith (1996) had volunteers conduct aural surveys to assess the distribution and relative abundance of calling amphibians in Maryland. Data from the aural surveys may be used to generate long-term population trend information from year to year. While aural surveys are effective in detecting the presence of amphibians, they may limit a researcher's ability to quantify the numbers of vocalizing amphibians if there are large numbers in a chorus (Smith 1995).

Basking surveys

Turtles bask to thermoregulate body temperatures and basking behavior has been studied by several researchers (Boyer 1965, Crawford et al. 1983, Schwarzkopf and Brooks 1985, Lovich 1987, Janzen et al. 1992). During basking surveys, turtles may be observed from constructed platforms or anchored logs where basking species are frequently found (Lovich 1988). Turtles tend to bask when environmental conditions may enhance heating, such as a sunny morning in early spring. Observations of basking turtles are most effective during 0800-1300 hr when basking activity is greatest (Schwarzkopf and Brooks 1985, Lovich 1987). Basking behavior

in turtles may enhance the health of adult turtles through removal of parasites, increased growth, increased metabolism, and resting behavior (Schwarzkopf and Brooks 1985, Janzen et al. 1992). However, for juvenile turtles, basking may increase the chances of predation (Janzen et al. 1992).

Turtles are typically found basking on a variety of platforms. Janzen et al. (1992) found yellowbelly sliders (*Chrysemys scripta*) on floating wood and emergent logs. Schwarzkopf and Brooks (1985) found eastern painted turtles (*Chrysemys picta*) basking on floating mats of vegetation and logs. At Mason Neck National Wildlife Refuge in Virginia, Lovich (1988) found *Chrysemys picta* basking on snags, stumps, and trees made abundant by the presence of beaver (*Castor canadensis*).

Road surveys

Road surveys are a popular search method to detect amphibians and reptiles, especially nocturnal species (Campbell and Christman 1979). Road surveys are line transects (the transect being the road) and are used to detect amphibians and reptiles moving through a habitat. The road survey technique was first used to observe nocturnal desert snakes (Klauber 1939). Data from road surveys may be used to estimate species richness and relative abundance and to gather information about breeding sites or other habitats that are frequently used (Cooperrider et al. 1986). Night drives during rainy evenings are typically the most productive (Heyer et al. 1994). Klauber (1939) and Vitt and Ohmart (1978) used spotlights, low headlight beams, and strong flashlights to locate amphibians and reptiles while road riding. Wyman (1990) reported mortality rates of salamander and frog species in New York

by using road surveys.

Animals move throughout their habitat to acquire food and water resources, locate cover, avoid predators, and to mate (deMaynadier 1986, Pough et al. 1998). Movement across forested roads has been documented (deMaynadier 1986). deMaynadier (1986) found that over half of all anuran captures in his study occurred along roadside habitats. Other researchers have also documented the use of roadside habitat by certain amphibian and reptile species. The exotic cane toad (*Bufo marinus*) found in Australia used forested roads as dispersal corridors (Seabrook and Dettmann 1996). Several species of amphibians may be attracted to roadsides bordered by ditches or other potential temporary breeding pools. The presence of the wood frog (*Rana sylvatica*) and other species in pools on logging roads suggests that water filled depressions created by heavy machinery are useful habitats for forest-dwelling amphibians (deMaynadier 1986, Semlitsch 1987, Phelps 1993). Campbell and Christman (1979) used road surveys to compare species richness in Florida and they were able to observe a number of rare species that were not detected using other techniques.

Time-constrained searches

A time-constrained search is a sampling procedure that provides information on the numbers of individuals or species collected per person-hour of effort (Heyer et al. 1994). Area-constrained searches are similar to time-constrained searches in that they provide the same type of information on numbers of individuals or species, but are limited by the area searched instead of by time. The goal of a time-constrained

search is to locate as many species of amphibians or reptiles as possible within a specified period of time (Heyer et al. 1994). Time-constrained searches may include turning over natural cover to locate reptiles and amphibians, dip netting for tadpoles, rolling back moss, walking along streams or wetlands, seeking out rock piles for snakes, or probing aquatic environments for salamanders (Welsh and Hodgson 1997). The specific methods to search an area will depend on the habitat features where the survey is being conducted. Welsh and Hodgson (1997) used time-constrained searches to locate animals in water, on streambeds, and on streamside substrates. Several factors may affect the efficiency and effectiveness of the time-constrained searches such as total search time, experience of observers, topography, weather, season, and time of day (Heyer et al. 1994). Heyer et al. (1994) suggested that time-constrained searches alone may not detect all species present in an area.

Several researchers have suggested that time-constrained searches are effective for anurans that are active in an open understory or for species that are active on the surface after rains (Toft et al. 1982, Bury and Corn 1987, Pough et al. 1998). Foley and Smith (1996) used time-constrained searches by flipping logs, leaf litter and rocks, inspecting stream banks, and watching for surface-active animals. Frazer (1978) and Griffiths (1984) used the time-constrained searches in Britain to locate aquatic newts at night.

Pitfall trap arrays

Pitfall traps are cans or buckets buried in the ground to trap animals (Handley and Kalko 1993, Kalko and Handley 1993, Heyer et al. 1994). Biologists use pitfall

traps to capture a variety of reptiles, amphibians, small mammals, gastropods, and arthropods (Howard and Brock 1961, Campbell and Christman 1979, Gibbons and Semlitsch 1981, Szaro et al. 1988, Handley and Kalko 1993, Bounds 1996). Pitfall traps may be used to generate estimates of species richness, species abundance, species presence, and to detect rare species (Campbell and Christman 1979).

Pitfall arrays may consist of a combination of drift fences and pitfall traps, drift fences with funnel traps, or drift fences with both pitfall and funnel traps (Heyer et al. 1994, Foley and Smith 1996). Imler (1945) used drift fences to capture bull snakes (*Pituophis catenifer sayi*).

Pitfall trap arrays may be linked with drift fences of various lengths, sizes, and configurations depending on the research objectives. Pitfall traps may be 19 L (15gal.) plastic buckets, number 10 coffee cans, or 2 L plastic bottles (Heyer et al. 1994, Bounds 1996). Smaller can traps are more effective in detecting smaller terrestrial species (Campbell and Christman 1979). However, there is an inherent bias in using the small can trap for detecting certain species. Snakes may enter and leave the can traps easily; however, the funnel traps may trap snakes (Campbell and Christman 1979, Gibbons and Semlitsch 1981). Species that are agile climbers (e.g. treefrogs; *Hylidae*) may escape from both the can or bucket traps and are seldom detected using pitfall traps (Cambell and Christman 1979).

Pitfall trap arrays may be left in place for long-term monitoring programs to collect data on species abundance and diversity. Pitfall trap arrays may be used to determine differences between various habitats. Campbell and Christman (1979) suggested using pitfall trap arrays with other sampling methods to gain the maximum

amount of information on the amphibians and reptiles in an area.

Cover board surveys

Cover boards are artificial wooden or aluminum cover objects placed in the habitat to sample reptiles and amphibians (Heyer et al. 1994). Amphibians and reptiles often use ground cover beneath leaf litter, sticks, or logs on the forest floor (Stebbins 1966, Conant 1975, Heyer et al. 1994). Creating artificial cover in a habitat may be a useful technique to sample amphibians and reptiles. By mimicking natural cover that animals frequently use, cover board surveys may be used to encounter some cryptic species of reptiles and amphibians with minimal damage to the environment and with a low risk of injury to those species observed (Grant et al. 1992, Heyer et al. 1994).

Cover boards are usually made of plywood, natural wood, or galvanized roofing tin; however, some researchers have used cardboard, bricks, masonry block, or dinner plates as cover boards (Grant et al. 1992). Grant et al. (1992) used both plywood sheets and roofing tin. The type of cover board material may affect the frequency and total species encountered. Grant et al. (1992) suggested using natural wood for amphibians because of its moisture retention ability and roofing tin for reptiles because of its heat conductivity. Boards that are not in contact with the ground are not as attractive to most salamander species (Heyer et al. 1994).

Heyer et al. (1994) stated that as long as the temperature is above freezing there are no temperature recommendations for checking boards. Little is known about the disturbance that continual checking of cover objects may have on the

probability of subsequent counts of amphibians or reptiles. Weekly intervals between checks do not appear to inhibit capture success (Heyer et al. 1994).

Standardized cover board designs have only been used in a few studies (Grant et al. 1992). Cover board surveys detected salamanders on Channel Islands in southern California and several salamander species in coastal Georgia (Heyer et al. 1994).

Sampling and Long-term Monitoring

The concern for biodiversity has heightened the need for the establishment of long-term monitoring studies. Standardized methods are needed to assess the status and health of reptile and amphibian populations (Heyer et al. 1994). The development of these standardized methods will help ensure comparability among different time and spatial scales.

An inventory of natural animal populations helps determine the success of many management programs (Bookhout 1994). Long-term studies of non-game species are important in understanding population dynamics and the effects of management activities by resource managers. Managers are often limited by funding, time and personnel. Therefore, cost and sampling effectiveness are important considerations for most resource managers.

OBJECTIVES

The objectives of my study were:

1. To inventory the species of amphibians and reptiles present at CHNWR;

2. To compare the species richness and relative abundance among techniques for the herpetofauna at CHNWR;
3. To compare the cost, time and sampling effectiveness of techniques for sampling amphibian and reptile populations and;
4. To develop a long-term monitoring protocol for amphibians and reptiles at CHNWR.

STUDY AREA

Chincoteague National Wildlife Refuge (CHNWR) is located primarily in Accomack County, Virginia with approximately 167.2 hectares in Worcester County, MD. Most of the 5600 hectare refuge is located on the southern end of Assateague Island, a 59.5 km (37 miles) long Mid-Atlantic coastal barrier island on the east side of the Delmarva Peninsula (Figure 1).

The refuge was established in 1943 under the authority of the Migratory Bird Conservation Act to provide migration and wintering habitat for migratory birds. Since 1943, refuge operations have expanded to include preservation and enhancement of endangered species, protection and enhancement of habitat for migratory and non-migratory species, conservation of indigenous species, and provision of opportunities for wildlife-oriented public use.

In 1965, after severe storms, Assateague Island was designated a National Seashore administered by the National Park Service (NPS) with provisions for the southern end to remain a wildlife refuge under the management of the U.S. Fish and Wildlife Service (FWS). The Maryland Department of Natural Resources (MDDNR) and the National Park Service (NPS) manage the northern portion of Assateague

Island.

Chincoteague National Wildlife Refuge is located on a barrier island. Barrier islands are coastal features composed of sand and other loose sediments transported by waves, currents, storm surges, and winds. Barrier islands protect other features, such as lagoons and salt marshes, from direct ocean waves (USFWS 1993). The soils of barrier islands consist primarily of sand and silt, poorly to moderately drained soil on tidal marshes and other wetlands. The dominant soil types are Assateague fine sand, Fisherman fine sand, Camocca fine sand, and Chincoteague silt loam (USFWS 1993).

Temperatures on the island ranged from a high of 27.5°C to a low 5°C in 1998, a high of 27°C to a low of 4.4°C in 1999 and a high of 33.4°C to a low of 5°C for 2000 (Table 2). Average annual precipitation was 8.6 cm in 1998, 9.4 cm in 1999, and 11.7 cm in 2000. Weather data was collected with a Morris technology systems fire weather station located in the northern portion of the refuge (Figure 1).

The vegetative communities of the refuge consist of the sand dune community; salt marsh/ pinewoods interface community, and pinewoods community (Figure 2). Beach plant species in the sand dune community are exposed to the constantly shifting sand, limited freshwater, high salinity, and extremes in temperature and wind. The most common beach species are American sea rocket (*Cakile edentula*) and sea lavender (*Limonium carolinianum*). The sand dune community occurs beyond the high tide mark. The most characteristic species that establish along the stabilized dunes are American beach grass (*Ammophila breviligulata*), salt marsh cordgrass (*Spartina alterniflora*), seaside goldenrod

(*Solidago sempervirens*), sandbur (*Cenchrus tribuloides*), button bush (*Cephalanthus occidentalis*), carpetweed (*Mollugo verticillata*), and evening primrose (*Oenothera spp.*). Shrubs, evergreens, and deciduous trees are predominant beyond the primary dune system. Composition of the salt marsh and pinewoods community varies according to salt spray, freshwater availability, and frequency of overwash and flooding. Common species in the salt marsh/pinewoods interface are black cherry (*Prunus serotina*), autumn olive (*Elaeagnus umbellata*), common persimmon (*Diospyros virginiana*), Canada serviceberry (*Amelanchier spp.*), northern bayberry (*Myrica spp.*), blackberry (*Rubus allegheniensis*), poison ivy (*Rhus radicans*), false heather (*Hudsonia spp.*) and greenbrier (*Smilax spp.*). Evergreens include wax myrtle (*Myrica cerifera*), red cedar (*Thuja spp.*), and American holly (*Ilex spp.*). The pinewoods community consists of primarily loblolly pine (*Pinus taeda*) (USFWS 1993).

Chincoteague National Wildlife Refuge lies within the Atlantic flyway for waterfowl, providing a resting and feeding habitat for migrating waterfowl, shorebirds, and songbirds. The refuge is known for its abundant, diverse bird habitat. More than 300 bird species use the refuge. The refuge supports wintering snow geese (*Chen caerulescens*), Canada geese (*Branta canadensis*), black ducks (*Anas rubripes*), mallards (*Anas platyrhynchos*), green-winged teal (*Anas crecca*), red-breasted merganser (*Mergus serrator*), ruddy duck (*Oxyura jamaicensis*), tundra swan (*Cygnus columbianus*), and other bird species (USFWS 1993).

Chincoteague National Wildlife Refuge has three federally listed endangered species: the bald eagle (*Haliaeetus leucocephalus*), the peregrine falcon (*Falco*

peregrinus), and the Delmarva Peninsula fox squirrel (*Sciurus niger cinereus*).

Chincoteague also has two federally threatened species: the piping plover (*Charadrius melodus*), and the Atlantic loggerhead sea turtle (*Caretta caretta*), which rarely nests on the refuge (USFWS 1993).

Thirty-one mammal species occur on CHNWR. The most commonly seen mammal species are white-tailed deer (*Odocoileus virginianus*), sika deer (*Cervus nippon*), Chincoteague ponies (*Equus equis*), red fox (*Vulpes vulpes*), raccoon (*Procyon lotor*), eastern cottontail (*Sylvilagus nuttallii*), and Delmarva Peninsula fox squirrel (*Sciurus niger cinereus*) (USFWS 1993).

While Chincoteague National Wildlife Refuge continues to provide important waterfowl habitat, the management emphasis has expanded over the years to address a variety of other non-game wildlife needs as well. The refuge also provides the public with important educational and recreational resources. Public use has grown from an estimated 100,000 visits in 1963 to more than 1.4 million visits in 1999 (USFWS 2000).

METHODS

To estimate reptile and amphibian populations at CHNWR, I compared 6 sampling techniques: aural surveys, basking surveys, road surveys, pitfall trap arrays, time-constrained searches, and cover board surveys. I define “observations” as the number of encounters I had with each species. I was not able to uniquely mark individuals so I was unable to determine if my encounters were with the same or different individuals over time. For each sampling technique, I recorded information

on temperature, wind speed, cloud cover, precipitation, start and ending times. I developed estimates of the costs for purchasing, installing, and sampling for each technique.

Pilot study

I conducted a pilot study to collect preliminary observations, and to assess the feasibility of using various techniques to sample amphibian and reptile populations (Bookhout 1994). During the pilot study in 1998, I tested 5 techniques: aural surveys, road surveys, pitfall trap arrays, cover boards, and area-constrained searches. I conducted the pilot study from July 12-August 4, 1998. I tested these techniques in 3 dominant vegetative communities at CHNWR: pine woods, salt marsh/pinewoods interface, and sand dune (Figure 2 and Table 3). I conducted sampling for 4 weeks in 1998 for the 5 techniques.

In 1998, the aural survey consisted of 6 survey points located at each of the pitfall trap array sites. I conducted aural surveys on July 12 and 22, and August 4, 1998. I conducted aural surveys in each of the 3 habitats by surveying 2 pitfall trap array sites each week for 3 weeks in 1998. Six surveys were conducted in 1998. I listened for vocalizing amphibians at each pitfall array site for 10 minutes each night. I began conducting the surveys 30 minutes after sunset and completed the surveys within 2 hours after sunset. Surveys were conducted on evenings when the weather conditions were most conducive to amphibian vocalizing behavior (Wells 1977). For example, amphibians tend to call when air temperatures are greater than 8°C (USFWS 1999), so I conducted surveys when nighttime air temperatures were above

8 – 12°C. I did not conduct aural surveys when winds were greater than 13-18 mph or during heavy precipitation (USFWS 1999). If the weather became unsuitable, the survey was discontinued.

I recorded the species and an estimate of the number of each species using the Wisconsin Index Value system (USFWS 1999). This system provides an index of 1-3 for each species as well as an estimated abundance count, which is the actual, or estimated number of anurans heard. I used an index of 1 when individual males could be counted and no calls were simultaneous; an index of 2 when calls were distinguishable with some simultaneous calling; and an index of 3 when a full chorus was heard and calls were continuous and simultaneous. Due to the low number of species that I detected with 6 points, I decided to expand my aural surveys to sample more habitats in 1999 and 2000.

The road survey was conducted 4 times a week for 3 weeks from July 13-August 7, 1998 while traveling to study sites to conduct the other techniques. I drove approximately 20 mph while doing road surveys. I conducted road surveys during both daytime and nighttime hours; however, the majority of road surveys were done in the daytime while conducting pitfall trap arrays, area-constrained searches and cover board surveys. Those road surveys conducted at night were done while in route to conduct the aural surveys. I identified any animal encountered to species. I also recorded odometer readings at the starting location, intersections in the roads, landmarks such as gates, and at the points where animals were observed. Weather conditions most suitable for the other techniques were taken into consideration first, since the road survey was typically done in conjunction with another technique to

save time and to determine the feasibility of conducting it with other refuge activities.

I conducted area-constrained searches at each of the pitfall trap array sites during the pilot study. Area-constrained searches are similar to time-constrained searches but are recommended for people with limited herpetological experience (Heyer et al. 1994). I searched 2 sites representing 1 habitat per week for 3 weeks from July 21 – August 4, 1998. I established 2 search areas in the pinewoods and sand dune vegetative communities for a total for 4 search areas. I set up a 15.25 m X 15.25 m quadrant at each of the pitfall trap array sites in these habitats. Each quadrant was approximately 15 m from each pitfall trap array and 30 m from each cover board site. I marked the quadrants with flagged rebar at all 4 corners so I could easily locate the sites. I conducted the area-constrained search by walking through the quadrant starting at the perimeter and working towards the middle. I searched for as many reptiles or amphibians as possible. The amount of time used to search was not fixed so that the entire quadrant could be surveyed. I systematically searched under logs and other suitable substrate using a potato rake, gloves, and a holding container for any amphibians and reptiles found. Captured animals were placed in a Ziploc® bag to measure the snout to vent length (SVL) and total length (TL), and weighed. Each animal was identified to species, held in a container until the end of the search to ensure they weren't sampled again, and released adjacent to the site (Welsh and Hodgson 1997). I returned all substrate material back to its original condition to ensure minimal disturbance to the habitat.

Pitfall trap array sampling began July 12 and occurred weekly for 4 weeks until August 3, 1998 for the pilot study. I selected pitfall trap array sites to represent

each of the three dominant vegetative communities on the refuge. I mapped out the habitats, used a grid to divide the habitats into equal units, and then used a random table of numbers to randomly select areas within each habitat to sample. I tried to select areas that were less likely to be disturbed by refuge visitors. If a site was located within 50 meters of a highly visited area, a new location was chosen using the random numbers table. Each site was located at least 0.5 km apart to achieve independent samples (Hulbert 1984).

I installed 6 pitfall trap arrays during the pilot study; however 2 arrays in the salt marsh were originally placed below the high tide mark and were exposed to periodic flooding. These sites were eliminated, and 2 different sites were selected in the salt marsh/pinewoods interface for 1999-2000. Any trap that was in saturated soil and became flooded was not used in the analysis (Smithberger and Swarth 1993). Therefore, in 1998, the pinewoods and sand dune habitats were sampled using pitfall trap arrays.

Each pitfall trap array was open for 4 consecutive days (96 hrs per week) each week in 1998. I checked each trap array every 24 hrs before noon to minimize stress to animals captured. Each trap was closed in the same order it was opened to ensure the same trapping effort in each habitat. All traps were closed with tight fitting lids when not in use and were maintained throughout the winter months.

Each array was situated North to South. Animals that moved along the substrate surface were redirected into traps placed at the ends and sides of the arrays. Arrays were a modification of the "Y" shaped barriers, with 120° between each leg developed by Jones (1981) that directed the animals into the traps and the drift fences

with funnel traps suggested by Bury and Corn (1987). Each array consisted of four 19 L plastic buckets, 6 funnel traps, and 3 drift fences (Figure 3). Buckets were buried flush with the ground in the center and at the end of the arrays. Drift fences were made with metal flashing 5 m in length, and each array was buried about 15 cm in the ground to prevent animals from tunneling under them. Funnel traps were constructed of aluminum window screen and heavy-duty staples. The funnel traps were shaped into a tube with inward directed funnel shaped openings at both ends. Each funnel was approximately 76 cm in length. Funnel traps were placed on the ground parallel to the array and at the center of the array on both sides (Heyer et al. 1994). Leaf litter was brushed into the opening of each funnel to provide a more natural appearance (Campbell and Christman 1979). Moistened sponges were placed in the funnel traps to prevent desiccation of animals waiting to be released (Daoust 1991, Smithberger and Swarth 1993). The sponges were moistened each time the array was checked.

Animals captured were placed in a Ziploc® plastic bag for taking measurements to reduce stress to the animal. The snout to vent length (SVL) and total length (TL) measurements were taken and the animal was weighed while in the bag. Each animal was then identified visually to species and sex, and then released approximately 2 m from the pitfall array (Foley and Smith 1996). Enough water to moisten the bottom of the buckets was provided every time traps were checked to aid in preventing desiccation (Vogt and Hine 1982). Due to high mortality in both buckets and funnels in 1998, I decided to use shade boards to provide protection for captured animals against desiccation, predation, and drowning (Campbell and Christman 1979, Bounds 1996). I constructed a plywood shade board for each bucket

and funnel trap for use in 1999-2000.

I selected cover board locations during the pilot study adjacent to 4 pitfall trap arrays, representing 2 habitats. I conducted cover board surveys July 16 and 27 and August 3, 1998. However, because I had no capture success, I moved the 4 cover board sites and added 2 sites in 1999 and 2000 to represent habitat used primarily by the red-backed salamander. To allow sufficient time for acclimation, I installed the cover boards in 6 new locations in October 1998 (Figure 4).

I placed plywood cover boards in three 7-board arrays that were parallel to each other and spaced 2.5 cm apart (Heyer et al. 1994). Cover boards were 30.5 cm x 30.5 cm x 1 cm in size. I directed each array North to South. Leaf litter was moved before cover boards were put into place to ensure the boards were flush with the ground. Each array was checked in the same order each week.

The arrays were checked one board at a time, by lifting the board quickly and capturing any amphibians or reptiles underneath (Heyer et al. 1994). The ground surface was gently brushed to the side for any target species not initially observed. I checked each array in less than five minutes. Any animal observed was placed in a Ziploc ® plastic bag for measuring the SVL, TL, and weight. Each animal was then visually inspected to determine the species and sex, and then released approximately 2 m from the cover board array.

Full-scale study

Aural surveys

I conducted aural surveys once a week for 20 weeks during breeding seasons

from February 18 - August 10, 1999 and for 25 weeks from February 16 – August 10, 2000. I established an aural survey route through the refuge from approximately 2.8 km south of the Maryland/Virginia border to the Tom's Cove visitor center (Figure 4). The survey route was 13.4 km long with 25 survey points. Each point was about 0.5 km apart and was marked with PVC pipe. Each point was labeled with an individual number and reflective tape so I could easily locate points at night. When I arrived at each point, I turned the truck engine off and waited for about 1 minute to allow the frog chorus to resume after the possible disturbance of my approach. I listened for vocalizing amphibians for 3 minutes at each survey point on the road shoulder (Foley and Smith 1996). I began conducting the surveys 30 minutes after sunset and completed all surveys within 3 hours after sunset. Surveys were conducted on evenings when the weather conditions were most conducive to amphibian vocalizing behavior (Wells 1977). For example, amphibians call when air temperatures are greater than 8°C, so I conducted surveys when nighttime air temperatures were above 8 – 12°C. I did not conduct the survey when winds were greater than 13-18 mph or if heavy precipitation existed (USFWS 1999). If the weather changed to an unsuitable condition, the survey was discontinued.

I recorded the species, an approximate number heard, and an estimate of the number of each species using the Wisconsin Index Value system (USFWS 1999). This system provides an index of 1-3 for each species as well as an estimated abundance count, which is the actual, or estimated number of anurans heard for that species. I recorded an index of 1 when individual males could be counted and no calls were simultaneous, an index of 2 when calls were distinguishable with some

simultaneous calling, and an index of 3 when I heard a full chorus and the calls were continuous and simultaneous.

Basking surveys

I conducted basking surveys once a week for 23 weeks from March 4, 1999 - August 10, 1999 and for 24 weeks from March 3, 2000 - August 10, 2000. During the 2000 season, I conducted the surveys with the CHNWR biological technicians and in conjunction with the refuges waterfowl surveys.

I conducted basking surveys between 0800-1200hr. While continuously driving along the survey route, I recorded all reptiles basking in the sun. I observed basking species with binoculars at 14 freshwater moist soil management units (MSMU). I recorded species basking on banks, floating vegetation, or downed logs. If a large number of species were observed in one area, I stopped and turned off the vehicle until all observations were recorded. All observations were made without altering the MSMU's. I followed the protocol developed by Patuxent National Wildlife Refuge (B. Woods, U.S. Fish and Wildlife Serv., Laurel, MD, pers. commun.)

Road surveys

I conducted road surveys at least once a week for 22 weeks from March 3, 1999 – August 5, 1999 and for 23 weeks from March 3, 2000 – August 10, 2000. This technique was used to locate amphibian or reptile species crossing the roads. Road surveys were conducted while in route to conduct all other survey techniques at

approximately 20 mph. I did road surveys during both daytime and nighttime hours; however, most road surveys were done in the daytime. Road surveys conducted at night were done while traveling to do the aural surveys and time-constrained searches. I recorded odometer readings at starting locations, intersections in the roads, landmarks such as gates, and at the point where the animals were observed.

Time-constrained searches

I conducted searches monthly at 6 sites for 4 months from April 16, 1999 - July 16, 1999 and for 4 months from April 13, 2000 – July 26, 2000. I established two search areas in each of the dominant vegetative communities for a total for six search areas (Figure 4). I conducted the time-constrained search by walking through an area or habitat for a 10-minute period, systematically searching for as many reptiles or amphibians as possible. I systematically searched under logs and other suitable substrate using a potato rake, gloves, and a holding container for any amphibians and reptiles found. When I captured an animal I placed it in a Ziploc® bag and measured the SVL, TL, and weight. When a large number of individuals were encountered at one site, animals were placed on paint sticks and SVL and TL were marked on the stick for subsequent measurement. Each animal was identified to species and released adjacent to the site at the end of the search to ensure they weren't sampled again (Welsh and Hodgson 1997). I returned all substrate material back to its original condition to minimize disturbance to the habitat.

Pitfall trap arrays

I opened the pitfall trap arrays monthly for 5 months from April 14, 1999 - August 5, 1999 and for 5 months from April 19, 2000 – August 9, 2000. Two pitfall trap arrays were set up in each of the 3 communities, for a total of 6 trap arrays (Figure 4). Each array was a modification of the “Y” shaped barriers, with 120° between each leg developed by Jones (1981) that directed the animals into the traps and the drift fences with funnel traps suggested by Bury and Corn (1987). Each array consisted of four 19 L plastic buckets, 6 funnel traps, and 3 drift fences (Figure 3). Buckets were buried flush with the ground in the center and at the end of the arrays. Each pitfall bucket was equipped with a plastic lid ring to provide an edge that would prevent escape by species with the ability to climb or jump. Drift fences were made with metal flashing 5 m in length, and drift fences were buried about 15 cm in the ground to prevent animals from tunneling under them. Funnel traps were constructed of aluminum window screen and heavy-duty staples. The funnel traps were shaped into a tube with inward directed funnel-shaped openings at both ends. Each funnel was approximately 76 cm in length. Funnel traps were placed on the ground parallel to the array and at the center of the array on both sides (Heyer et al. 1994). Leaf litter was brushed into the opening of each funnel to provide a more natural appearance (Campbell and Christman 1979). Moistened sponges were placed in the funnel traps to prevent desiccation of animals (Daoust 1991, Smithberger and Swarth 1993). The sponges were moistened each time the array was checked.

The pitfall trap arrays were opened for 48 hrs once a month, and were checked every 24 hrs before noon to minimize stress to animals captured. Each trap was

closed in the same order it was opened to ensure the same trapping effort in each habitat. When the traps were not being used they were closed with tight fitting lids and were checked for safety/vandalism throughout the winter months.. Any trap that was in saturated soil and became flooded was not used in the analysis (Smithberger and Swarth 1993).

Animals captured were placed in a Ziploc® plastic bag for taking measurements to reduce stress to the animal. The SVL and TL measurements were taken and the animal was weighed while in the bag. Each animal was identified visually to species and sex, and then released approximately 2 m from the pitfall array (Foley and Smith 1996). Enough water to moisten the bottom of the buckets was provided every time traps were checked to aid in preventing desiccation (Vogt and Hine 1982). I covered each pitfall trap and funnel with a plywood cover. The cover for each bucket was raised approximately 2-4 cm above the bucket and provided protection against desiccation, predation, and drowning (Campbell and Christman 1979).

Cover board surveys

I conducted the cover board survey weekly for 18 weeks from April 7, 1999 - August 12, 1999 and on October 28, 1999 and November 23, 1999. In 2000, I conducted the survey weekly for 23 weeks from March 3, 2000 – August 10, 2000 (Figure 4). The 6 cover board arrays were installed in October 1998 to allow the wood to weather and appear more natural to target species. The cover boards remained in place for the duration of the study.

I placed wood cover boards in arrays of three 7-board arrays that were parallel to each other and spaced 2.5 cm apart (Heyer et al. 1994). Cover boards were 30.5 cm x 30.5 cm x 1 cm in size. I directed each array North to South. Leaf litter was moved before cover boards were put into place to ensure the boards were flush with the ground. Each array was checked in the same order each week.

The arrays were checked one board at a time, by lifting the board quickly and capturing any amphibians or reptiles underneath (Heyer et al. 1994). The ground surface was gently brushed to the side for any target species not initially observed. I checked each array in less than five minutes. Any animals captured were placed in a Ziploc® plastic bag and I recorded SVL, TL, and weight. Each animal was visually identified and released approximately 2 m from the cover board array.

Statistical Analysis

I analyzed my data using SYSTAT® 6.0 for windows, STATISTIX® 2.0 for windows, and Microsoft® Excel 97. I compared the six techniques using analysis of variance (ANOVA). I further analyzed the ANOVA by using the Least significant difference (LSD) comparison of the mean and Tukey's (HSD) statistical tests (Zar 1984).

Cost comparison

I compared the cost for equipment, installation, data collection, and maintenance for each technique. I estimated the total time needed to conduct each survey and compared the effectiveness of each technique in detecting amphibians and

reptiles. I estimated transportation costs at approximately \$0.32 per mile. I estimated labor costs at \$10 per hour.

Comparison of Techniques

The count of species at a particular site, or species richness, is often used to express the concept of species diversity (Pough et al. 1998). A common index used to compare species diversity is the Shannon-Weaver index (Shannon and Weaver 1949). I compared the species diversity for each technique to compare sampling effectiveness. Some techniques are more likely to detect certain taxa. For example, basking surveys are more likely to detect species that bask such as turtles. Therefore, I compared similar techniques that were more likely to detect certain taxonomic groups. Based on the results of previous researchers and species previously found by CHNWR staff, I developed a list of possible species likely to occur on Assateague Island (Table 4). I then divided this list into possible target species for each of the 6 sampling techniques (Table 5). I compared the total species I found using the 6 techniques in 1998-2000 with the potential list to estimate the sampling effectiveness of each of the 6 techniques.

RESULTS

Pilot study

Based on my preliminary results from the pilot study in 1998 (Table 6), I decided to modify some techniques in 1999-2000. I conducted aural surveys at 2 pitfall trap array sites on 3 trap nights in 1998 (n=6). I found 2 species using aural surveys: the green tree frog (*Hyla cinerea*) and the fowler's toad (*Bufo woodhousii*

fowleri). In 1998, using aural surveys, I found 33% of the total possible amphibian species at CHNWR (Tables 5 and 6).

In 1998, I conducted road surveys 13 days and 8 nights. I found 7 individuals representing 5 species: common snapping turtle (*Chelydra serpentina serpentina*), eastern painted turtle, (*Chrysemys picta picta*), eastern mud turtle, (*Kinosternon subrubrum subrubrum*), northern water snake, (*Nerodia sipedon sipedon*), and fowler's toad (*Bufo woodhousii fowleri*). The majority of species I found using road surveys in 1998 were turtles (71%) compared to snakes (14%) and amphibians (14%). I observed 25% of the total possible species (Tables 5 and 6).

I conducted 2 time-constrained searches in 1998 at 4 sites (n=8). I found one species, the fowler's toad (*Bufo woodhousii fowleri*), in the pinewoods habitat. I found 5% of the 20 possible amphibian and reptile species using the time-constrained search (Tables 5 and 6).

I tested pitfall trap arrays for 8 trap nights each in the pinewoods and sand dune habitats (n=16). I found 4 species (2 snakes and 2 amphibians): eastern hognose snake (*Heterodon platirhinos*), northern black racer (*Coluber constrictor constrictor*), fowler's toad (*Bufo woodhousii fowleri*), and southern leopard frog (*Rana sphenoccephala*). I found 20% of the 20 possible amphibian and reptile species using the pitfall trap arrays (Tables 5 and 6). During the pilot study, 3 of the 4 snakes and 14 of 114 fowler's toads were found dead in pitfall or funnel traps. Therefore, in 1999, I added plywood cover boards to reduce mortalities from desiccation and predation (Bounds 1996).

Full-Scale study

For each sampling technique, I recorded information on temperature, wind speed, cloud cover, precipitation, start and ending times (Table 7). The weather conditions for peak observation times included wind speeds less than 7 mph and temperatures ranging from about 9°C to 25°C.

Aural Surveys

I conducted aural surveys 23 nights in 1999 and 23 nights in 2000. I found the same 4 species in 1999 and 2000: fowler's toad (*Bufo woodhousii fowleri*), southern leopard frog (*Rana sphenoccephala*), green tree frog (*Hyla cinerea*), and bullfrog (*Rana catesbeiana*). There were 2 amphibian species I did not detect using aural surveys: New Jersey chorus frog (*Pseudacris triseriata kalmi*) and green frog (*Rana clamitans melanota*). Overall, I detected the fowler's toad (80%) most frequently in 1999, followed by the southern leopard frog (13%), the green tree frog (5%), and the bullfrog (2%) (Figure 5). In 2000, however, I detected the green tree frog (70%) the most frequently, followed by the fowler's toad (16%), the southern leopard frog (13%), and the bullfrog (1%) (Figure 5). Using aural surveys, I found 67% of the possible anuran species in 1999 and 2000 (Table 5 and Figure 5).

I compared the dates (routes) using ANOVA to determine significant differences between 19 routes (of 25 points) in 1999 and 19 routes (of 25 points) in 2000. I found a significant effect ($p = 0.041$) for route in terms of the number of species detected. I found the most amphibians on April 20, May 20 & 27, and June 2, 1999 and May 24 and June 9, 2000 (Table 8). When I compared the differences

between the 1999 and 2000 season, I found no significant difference between years for the number of amphibian species detected. By May 20, 1999 and May 16, 2000 I had detected all of the anuran species. I developed calling phenologies for each species I detected using aural surveys from 1999-2000. The southern leopard frog was found from February–April, the fowler’s toad from March–August, the green tree frog from April–August, and the bullfrog from May-June (Table 9).

I used Spearman’s rank correlation (Zar 1984) to calculate the relative abundance indices for each species found using aural surveys. Species relative abundance ranks (Table 10) were not significantly different between 1999 and 2000 (Spearman’s $P = 0.0716$, $P < 0.001$).

Basking surveys

I conducted 21 basking surveys in 1999 and 23 in 2000. I found 6 turtle species: eastern painted turtle (*Chrysemys picta picta*), red-bellied turtle (*Pseudemys rubriventris*), common snapping turtle (*Chelydra serpentina serpentina*), diamondback terrapin (*Malaclemys terrapin terrapin*), eastern mud turtle (*Kinosternon subrubrum subrubrum*), and eastern box turtle (*Terrapene carolina carolina*). I saw 6 turtles that I could not identify to species because the animals moved before I could make a positive identification. In 1999-2000, I found the eastern painted turtle (n=143) and the red-bellied turtle (n=68) most frequently (Figure 6). Of the 7 species of freshwater/brackish turtle species known to occur on the refuge, I found 6 (86%) using the basking technique; however, only 5 (71%) turtle species were detected each year. A total of 251 observations of 6 freshwater/brackish

turtles were made from 1999-2000 using basking surveys (Figure 7 and Table 11). The only species not detected using the basking technique was the spotted turtle (*Clemmys guttata*).

I analyzed the data by comparing the 14 moist soil management units (MSMU's) per year by the number of observations made and the number of species found using basking surveys. Analysis of variance showed a highly significant site effect on the number of observations ($p=0.0095$) and species ($p=0.0002$). I observed the highest number of observations and species in the MSMU's "Bnorth" and "Bsouth" (Figure 8). I observed significantly fewer turtles in 1999 ($n=85$) than in 2000 ($n=161$) ($p=0.026$) (Figure 7). I compared the monthly observations using basking surveys (Figures 9 and 10) and found that surveys made during June-July yielded the highest total number of species (mean = 3.75, SD = 1.25, .95 % CI = 1.7-5.8) (Table 12).

Road Surveys

I conducted road surveys 36 days and 19 nights in 1999, and 25 days and 15 nights in 2000. I found no differences in the number of observations or the number of species for 1999 and 2000 so I combined the data. Overall, I found 10 species: Fowler's toad (*Bufo woodhousii fowleri*) ($n=13$), northern black racer (*Coluber constrictor constrictor*) ($n=3$), eastern hognose snake (*Heterodon platirhinos*) ($n=2$), northern water snake (*Nerodia sipedon sipedon*) ($n=1$), black rat snake (*Elaphe obseleta obseleta*) ($n=6$), red-bellied turtle (*Pseudemys rubriventris*) ($n=1$), diamondback terrapin (*Malaclemys terrapin terrapin*) ($n=14$), eastern painted turtle

(*Chrysemys picta picta*) (n=13), eastern mud turtle (*Kinosternon subrubrum subrubrum*) (n=3), and common snapping turtle (*Chelydra serpentina serpentina*) (n=14) (Figure 11).

I compared the total number of species and observations found by month for 1999 and 2000 using road surveys. In 1999, I detected the highest number of species and the highest number of observations in July (Figures 12 and 13). However, in 2000 I detected the highest number of species and observations in May and June. The common snapping turtle and eastern painted turtle were detected most frequently in 1999. The eastern painted turtle and diamondback terrapin were detected most frequently in 2000 (Figure 14).

I found no significant difference between 1999 and 2000 when comparing the road survey routes using ANOVA. The data was compared for 9 routes in both 1999 and 2000. I found no significant effects for route or year for the number of species detected or the number of observations.

I calculated relative abundance indices for each species from 9 road survey routes in 1999 and 2000 using Spearman's rank correlation (Zar 1984). Species relative abundance ranks (Table 13) were not significantly different between 1999 and 2000 (Spearman's $P = 0.0769$, $P < 0.001$).

Time-constrained searches

I did 5 time-constrained searches in 1999 and 6 in 2000. I made 104 observations of 5 species: green tree frog (*Hyla cinerea*), bullfrog (*Rana catesbeiana*), southern leopard frog (*Rana sphenoccephala*), Fowler's toad (*Bufo*

woodhousii fowleri), and common snapping turtle (*Chelydra serpentina serpentina*) (Figure 15). The southern leopard frog was detected the most frequently in 1999 (n=9); however, the bullfrog was detected the most frequently in 2000 (n=39) (Figure 16). I was unable to identify 1 individual in 1999 and 14 individuals in 2000 due to escapes. I found 25% of the possible species present using the time-constrained search.

Using analysis of variance, LSD, and TUKEY, I found a significant effect between 1999 and 2000 in terms of the number of observations made ($p=0.045$). I found no significant difference ($P=0.1250$) between the numbers of species detected between habitats using the time-constrained search.

Pitfall trap arrays

The salt marsh, sand dune, and pinewoods habitat pitfall trap arrays were open for 16 trap nights in 1999 and 10 trap nights in 2000. I made 73 observations of 6 species: green tree frog (*Hyla cinerea*), Fowler's toad (*Bufo woodhousii fowleri*), southern leopard frog (*Rana sphenoccephala*), northern black racer (*Coluber constrictor constrictor*), eastern hognose snake (*Heterodon platyrhinos*), and eastern mud turtle (*Kinosternon subrubrum subrubrum*) (Table 11). Of the 6 total species I detected using pitfall trap arrays, I found 4 (20%) in 1999 and 5 (25%) in 2000. I found fowler's toad most frequently (73%) (Figure 17).

In 1999, I detected 3 amphibians and 1 reptile species in the pinewoods habitat, 1 amphibian and 1 reptile species in the salt marsh, and 1 reptile species in the sand dune habitat. In 2000; however, I detected 1 amphibian and 2 reptile species

in the salt marsh habitat, 1 amphibian and 1 reptile in the sand dune habitat, and 2 amphibian species in the pinewoods habitat (Figure 18). One pitfall trap array in the pinewoods habitat had periodic flooding because of its location in a low depression. One of the 4 buckets in the array remained flooded for the entire 2000 trapping season.

The species richness between habitats appeared to be about the same with the total species found in each habitat ranging from 1-4 species (Figure 18). Each of 3 habitats was compared statistically in an analysis of variance table between years for significance. A comparison of the three habitats with ANOVA showed no significant difference between the numbers of species detected between habitats. However, I compared the number of observations made in the 3 habitats and found a significant difference ($p = 0.0198$).

I compared the difference between 1999 and 2000 number of observations (Figure 19) made with the pitfall trap arrays using a paired t-test and found a significant difference ($p = 0.0252$, 95% CI: 0.7003-8.633, SD=1.8021, mean=4.6667) between years. I found no significant difference ($p = 0.6576$, 95% CI: -0.6385-0.9719, SD=0.3658, mean=0.1667) for the number of species detected when I compared 1999 and 2000 with a paired t-test.

I captured 4 small mammal species in pitfall trap arrays: shrew (*Sorex spp.*); white-footed mouse (*Peromyscus leucopus*); mole (*Talpidae spp.*); meadow jumping mouse (*Zapus hudsonius*); and 2 bird species: titmouse (*Parus spp.*) and tree swallow (*Tachycineta bicolor*). I released all live captures adjacent to the pitfall trap array. From 1998-2000, I had mortalities in pitfall traps including 8 shrews, 1 titmouse, 3

white-footed mice, and 1 mole.

Cover board surveys

I found 2 species, Fowler's toad (*Bufo woodhousii fowleri*) and the red-backed salamander (*Plethodon cinereus*), using cover boards and captured 5 individuals (Figure 20). The 6 cover board locations (sow pond, white hills, wills hole, PW5, gravel pile, and maintenance) were in just the pine woods habitat. I used cover boards mainly to sample the red-backed salamander. I believed the salamander was not likely to be found in either the sand dune or salt marsh habitats. Due to the low numbers of species detected in 1999-2000, I did not conduct statistical analysis for cover boards.

Cost comparison

I compared the costs for equipment, installation, data collection, maintenance and overall total costs for each of the 6 sampling techniques (Table 14). The pitfall trap arrays were the most expensive overall (\$2953) and road surveys were the least expensive (\$485).

The materials I used for the aural survey included PVC pipe and reflective tape to mark points along the route, headlamp, insect suit, data sheets, recorder, and batteries. I also purchased anuran call tapes for volunteers to use who helped with the survey. Initial cost for materials to conduct the survey was approximately \$193. It took approximately 8 hours to cut, label, and place the PVC pipe along the route. Labor to install the survey markers was approximately \$80. It took me about 3 hours/

night (23 surveys per season). To collect data for one season required approximately \$690 in labor costs and \$15 in transportation costs. There were no costs associated with maintenance of the PVC pipe, because extra pipe was purchased in the beginning and the pipe stays in place from year to year. Total estimated costs for aural surveys for 1 season were \$978.

The material I used for the pitfall trap arrays was donated by the Maryland Department of Natural Resources; however, I estimated the costs for this equipment. The materials used to construct each array included aluminum flashing, window screen, cable ties, rebar, shovels, post hole diggers, stakes, 19 L buckets with lids, shade lids, and heavy duty staples. Total costs for materials per array were approximately \$219 (\$1314 for 6 arrays). It cost approximately \$579 for supplies and labor to install the pitfall trap arrays. Additional materials to check the pitfall trap arrays (scales, thermometers, Ziploc® bags, and data sheets) cost approximately \$90. These materials were also used for other sampling techniques. The time to conduct one pitfall trap array in the 3 habitats was approximately 3-4 hours, depending on the number of animals captured. To conduct 6 pitfall trap arrays (approximately 13 trap nights per season), labor costs were approximately \$520. I had to maintain (rebuild) the pitfall trap arrays after every major storm event, heavy winds, and between seasons. The pitfalls located in the sand dune habitat were rebuilt each season due to storm damage. I conducted approximately 35 hours of maintenance (\$350 labor/\$100 supplies) on the pitfall trap arrays between 1999 and 2000. Total estimated costs for 6 pitfall trap arrays for 1 season were \$2953.

I used plywood for cover boards. The cover boards cost approximately \$350

for lumber and materials to paint the boards. The time needed to construct the arrays included time to paint, cut, and install each array. The 6 arrays took approximately 1 week to construct and install (\$300 labor). Cover board surveys took approximately 3 hrs to conduct. To conduct 6 cover board surveys (20 surveys per season), labor costs were approximately \$600 (\$15 transportation). I maintained the cover board arrays (\$20 supplies/labor) by replacing windblown or damaged boards. Total estimated costs to conduct cover board surveys for 1 season were \$1285.

Once I determined the routes for my other sampling techniques, the road survey did not require any additional time for set-up or maintenance. No materials were needed to install the road survey. The road survey took 1-3 hours to complete (\$470 labor/ \$15 transportation) depending on the other surveys I was doing. Therefore, the total estimated costs for road surveys for 1 season were \$485.

For time-constrained searches I used paint sticks, scales, Ziploc® bags, thermometer, insect suits, headlamps, batteries, and data sheets. The supplies I used cost approximately \$183.50. The time-constrained search required minimal set-up costs (\$60 labor). I estimated approximately \$600 labor /\$15 transportation to collect data for 1 season at 6 sites (approximately 20 surveys per season). There was no cost associated with maintaining the time-constrained search areas. The total estimated costs for time-constrained searches for 1 season were \$858.50.

The supplies needed for basking surveys were binoculars, data sheets, and field guides. The approximate cost of these supplies was \$225. The basking survey required minimal time for set-up (\$60 labor). To collect data using basking surveys for 1 season (approximately 22 surveys per season), labor was approximately \$660

and transportation \$15. There was no cost associated with maintaining the basking survey route. Total estimated costs for basking surveys for 1 season were \$960.

Comparison of techniques

I detected the highest total number of species ($n=10$) using the road survey, followed by pitfall trap arrays ($n=6$) basking surveys ($n=6$), time-constrained searches ($n=5$), aural surveys ($n=4$), and cover board surveys ($n=2$) (Table 15). I detected the same number of amphibian species using the aural survey ($n=4$) and the time-constrained searches ($n=4$) for both 1999 and 2000. Using pitfall trap arrays I detected 3 amphibian species and the road survey 1 amphibian species. Interestingly, using road surveys I found the same number of turtle species as I did using basking surveys. I observed 3 snake species while conducting road surveys compared with 2 snake species using pitfall trap arrays.

In comparing the Shannon-Weaver indices for each technique, I found that the road survey had the highest species diversity ($H^i = 1.914$) followed by the time-constrained searches ($H^i = 1.443$), basking survey ($H^i = 1.037$), cover boards ($H^i = 0.566$), and pitfall trap arrays ($H^i = 0.517$) (Table 16). I was not able to calculate a species diversity index for the aural surveys because I did not record the number of individuals in each survey; instead I used the Wisconsin Index.

Two pitfall trap arrays and two time-constrained search areas were located in each of the three habitats (Table 3). I detected 3 different species using pitfall trap arrays that I did not find using time-constrained searches: the northern black racer (*Coluber constrictor constrictor*), the eastern hognose snake (*Heterodon platyrhinos*),

and the eastern mud turtle (*Kinosternon subrubrum subrubrum*). I detected 2 species using time-constrained searches that were not found using pitfall trap arrays: bullfrog (*Rana catesbeiana*) and common snapping turtle (*Chelydra serpentina serpentina*) (Table 11). Because of the similarity in design of the pitfall trap arrays and time-constrained searches, I statistically compared the effects between techniques and years for the number of species detected and the number of observations made. I did not find a significant difference ($p=0.7152$) between techniques or years ($p=0.2817$) for the number of species when I compared pitfall trap arrays and time-constrained searches. In comparing the average number of observations made using pitfall trap arrays to those made using time-constrained searches, I found no significant difference ($p= 0.0615$) between 1999 and 2000.

DISCUSSION

As visitation and local development increases, the need to monitor the adverse impacts on CHNWR's herpetological communities increases. Much of the scientific community is aware of the need for standardized sampling methods that are critical for comparisons of data gathered at different times, places, and from different people (Heyer et al. 1994). My study shows that road surveys can be used to obtain more cost effective and efficient results than the other 5 techniques. Some techniques will need to be modified in different situations. I urge investigators to take the results that I have found and make modifications when it is truly necessary. No survey method alone will produce a complete list of the amphibians and reptiles in an area (Foley

and Smith 1996).

Pilot Study

I detected only 33% of the total possible amphibian species using aural surveys in 1998. This could have been due to the time of year aural surveys were begun in 1998. By starting in July, I had missed the breeding seasons for both the southern leopard frog and the bullfrog.

In 1998, I used the area-constrained search (Heyer et al. 1994) to familiarize myself with the species and the technique before attempting the time-constrained searches for 1999-2000. Campbell and Christman (1979) suggest that experienced field herpetologists should conduct time-constrained searches. Using area-constrained searches in the pilot study allowed me the time to gain valuable experience in species identification and survey techniques before beginning the full- scale study in 1999. I only captured one species during the pilot study. Some possible reasons for low capture success in 1998 could have been my inexperience with the technique, the length of the sampling period (July-August), the drought-like conditions in 1998, or location of the search areas. Due to low capture success, the area-constrained sites used in 1998 were not used in 1999 or 2000. I had much higher capture success in 1999-2000, possibly due to the search areas being located closer to possible breeding sites.

Due to the relatively high mortalities I had using pitfall trap arrays in the pilot study, I decided to use plywood boards to provide shade and protection to both pitfall and funnel traps in 1999. My results support the findings of other researchers who

recommend using shade boards to reduce mortalities with pitfall traps (Heyer et al. 1994, Bounds 1996). I found that the pitfall trap arrays had much lower mortalities after the shade boards were installed.

Full-Scale Study

Aural Surveys

Conant et al. (1990) reported 6 anuran species on Assateague Island, and I found 4 of those 6 species using the aural survey. One species I did not find was the New Jersey chorus frog (*Pseudacris triseriata kalmi*), which was last observed in the early 1970's by Lee (1972). Lee (1972) found this species in only one location on CHNWR, near the lighthouse. One survey point on my route was adjacent to the lighthouse, yet I did not find the species in 1999 or 2000. Neither Gibbons and Coker (1978) or Mitchell et al. (1993) reported this species at CHNWR. In addition, staff at CHNWR have not encountered this species on the refuge in recent years (I. Ailes, U.S. Fish and Wildlife Serv., Chincoteague, VA, pers. commun.). Possibly, the New Jersey chorus frog population may be extirpated.

I also did not hear the green frog (*Rana clamitans melanota*) using the aural survey. Harris (1975) and Conant et al. (1990) reported the green frog at CHNWR. Green frogs occupy primarily permanent bodies of freshwater (Conant and Collins 1991, Mitchell and Anderson 1994). Several periods of drought-like conditions may have eliminated the habitat for this species and it may no longer be present at CHNWR. I found that most permanent freshwater ponds at CHNWR were completely dry by mid-July in 1999. In addition, the spring peeper (*Pseudacris*

crucifer) and the northern cricket frog (*Acris crepitans*) both occur on the nearby mainland; however, the eggs of both species cannot tolerate even low levels of salinity and therefore it is likely that neither species can reproduce on the island (Mitchell and Anderson 1994).

I developed a calling phenology for each of the anuran species I found during the aural surveys from 1999-2000. My results were similar to the breeding seasons reported for each species on the island by Mitchell and Anderson (1994).

Recently, managers and administrators have placed a high priority on monitoring amphibian populations. For example, in 1999 funding for aural surveys became available to all National Wildlife Refuges in the Northeast region of the U.S. Fish and Wildlife Service. Approximately \$640 was provided for start-up costs and an additional \$320 per year was available for the aural surveys at each refuge. The funding for this survey should be available to CHNWR until 2009; however, funding priorities could change under different political administrations.

Basking surveys

The basking survey was the most effective technique to detect turtle species (not including sea turtles). Mitchell and Anderson (1994) suggested the spotted turtle, the eastern box turtle, and the eastern mud turtle are all species that do not generally bask; however, they did find the spotted turtle basking in late winter or early spring. Lee (1972) and Mitchell and Anderson (1994) both found the spotted turtle (*Clemmys guttata*). Although I observed the spotted turtle while conducting other refuge activities, I did not find it using either of the 6 techniques during my research. I did

find the eastern box turtle and eastern mud turtle during basking surveys, but it may have been due to chance. I observed these species on the banks of the MSMU's and not on basking logs. I may have observed these turtles while they were moving through the habitat. I observed two species most frequently, the eastern painted turtle (*Chrysemys picta picta*) and the common snapping turtle (*Chelydra serpentina serpentina*). Kofron and Schreiber (1987) also observed these species most frequently using basking surveys. I recommend conducting the basking survey January - December to detect all possible species.

During the drought-like conditions of 1999, most MSMU's were completely dry by mid-July. All MSMU's (except North wash flats) retained water throughout the 2000 season. Total average precipitation for 1999 was approximately 1 inch less than that of the average rainfall for only 8 months in 2000. I observed a significantly higher number of turtle species in 2000 than in 1999, which could have been due to the increased rainfall in 2000 and drier conditions of 1999. North wash flats remained dry for 1999-2000 because it is managed as a satellite nesting area for the piping plover, and because it is pumped dry before each nesting season.

The dry conditions of the MSMU's in 1999 seemed to attract the diamondback terrapin. All of the terrapins I observed during basking surveys in 1999 were seen in the "South wash flats" MSMU. However, in 2000, South wash flats retained water from heavy precipitation and all of the terrapins I observed were seen on the roadside adjacent to South wash flats laying eggs, not in the MSMU. I found 80% of all terrapin observations during May and June. Conant et al. (1990) also found diamondback terrapins emerging from the marshes of the bayside of CHNWR to lay

eggs in May through June.

Kofron and Schreiber (1987) suggested that common snapping turtles and adult eastern painted turtles selected man-made areas for cover rather than natural areas because of the differences in bottom substrate. They also found that eastern painted turtles had periods of movement between the two types of areas. This may indicate the man-made freshwater impoundments of CHNWR are important to turtle species that bask. Future research could compare basking turtle species, habitat selection, and the types of substrate at CHNWR.

Lovich (1988) secured a 22 X 600 cm log in a deep channel where turtles were frequently seen. I did not use artificial basking sites in my study. The placement of logs in basking areas could be one way to standardize the results obtained from year to year. Placement of such logs could also be a way for resource managers to provide basking sites in the ditches along the MSMU's to provide basking opportunities throughout the summer months when most MSMU's are dry.

Previous researchers have found that turtles generally select basking sites away from shore and near deep water (Boyer 1965, Moll and Legler 1971, Bury et al. 1979). I found most species were located near MSMU shores, except for the diamondback terrapin which was generally observed searching for nesting sites. I did not collect data on the depth of the water in areas where turtles were basking, so I cannot determine if most were near deep water.

Road surveys

Campbell and Christman (1979) suggested that road surveys may be used to

develop an estimate of the species richness of a site. For example, if the road survey is done near a breeding site, it can provide a reasonable estimate of the animals moving to the site. In my study, the transects were roads throughout CHNWR. If the roads were divided into smaller sections the data may have been more comparable by site for species richness and relative abundance (T. Robinson, Virginia Tech, Blacksburg, VA, pers commun.). I suggest that future studies divide the road transect into several sections of equal length (perhaps 500 m) to compare results among sites and years.

Heyer et al. (1994) suggested the most productive night driving occurs when the same stretch of road is sampled several times in the same evening because animals are almost always moving across roadways. This approach assumes that individual animals are observed only once during a night. However, this assumption may not be valid and some individuals may be counted more than once which would bias the results. One alternative would be to conduct multiple road surveys in a single night, but use just the highest value or peak value for each species on an individual transect suggested as by Rosen and Lowe (1995) and Bounds (1996). If I had surveyed the same sections of the roadways repeatedly in a single night, I could have developed an index of peak values for each species and compared my results among sites and years. I also conducted road surveys primarily during the day, which may have resulted in a larger number of reptile species ($n=9$) than amphibian species ($n=1$). The roads I traveled were made mostly of dirt and gravel, which made it difficult to see amphibians, especially at night. By conducting the road survey as I traveled to other surveys, I may have biased my results. Of the 5 other techniques, 3

were conducted during the day and 2 at night. The best time to conduct the survey to detect amphibians would be at night after a rain (Heyer et al. 1994). If the objective is to observe reptile species, I would conduct the survey during the day. The results of my study show that conducting the road survey during the day can detect the same number of turtle species as the basking survey and more snake species than any other technique.

Time-constrained searches

I found one species, Fowler's toad, using the time-constrained search method in the salt-marsh habitat. Fowler's toads are able to tolerate low levels of salinity (Mitchell and Anderson 1994). The salinity content in the soil and tidal fluctuations may be a deterrent for most reptile and amphibian species in the salt-marsh habitat (Hagstrom 1981, Beebee 1985, Mitchell and Anderson 1994). The animals I found were located on the ground above the high tide mark.

My success in finding species using time-constrained searches may have been affected by weather conditions in 1999 and 2000. For example, the pine woods habitat experienced major drying effects of the drought-like conditions on CHNWR in the summer of 1999. By mid-July, most MSMU's and ephemeral pools located on the refuge were dry. All permanent breeding pools remained as only shallow depressions with a minimal amount of water. Drying conditions may have reduced metamorphisms of amphibian species.

In addition, the sand dune habitat was altered by storms in the winter of 1999-2000. The change in physical appearance of one of the sand dune sites may have

affected the species in 2000. However, barrier islands such as Assateague Island, are subject to constant environmental changes and effective monitoring techniques should allow for temporal and spatial variations.

I found more species in the pinewood and sand dune habitats in 2000 than in 1999. I believe my higher capture success in 2000 for time-constrained searches may have been due to more favorable weather conditions (higher precipitation).

I found time-constrained searches to be effective for locating individuals and made the second highest number of observations using time-constrained searches. Of my 6 techniques, only basking surveys had a higher number of observations. Using time-constrained searches, I captured amphibian and turtle species.

The most abundant species detected by Foley and Smith (1996) using time-constrained searches in Maryland were American toad (*Bufo americanus*), red-backed salamander (*Plethodon cinereus*), and the spring peeper (*Pseudacris crucifer*). In comparison to pitfall trap arrays, Foley and Smith (1996) did not find a significant difference in the species diversity indices between 5 time-constrained searches and 3 days of pitfall trap array sampling. Welsh and Hodgson (1997) found multiple species of snakes, lizards, and terrestrial salamanders using time-constrained searches.

Heyer et al. (1994) suggested that using time-constrained searches alone was not effective for detecting all species in an area. Habitat disturbance and differences in observers abilities were suggested as reasons the time-constrained searches may not be effective in detecting all possible species. My results tend to support Heyer et al. (1994). If I had just used time-constrained searches I would not have found as

many species as I did by using the 5 other techniques. For example, I would not have observed several snake and turtle species. I believe other techniques such as road surveys, aural surveys, and basking surveys would yield similar results as time-constrained searches and may be more effective in terms of sampling time and species detection.

Pitfall trap arrays

I found a significant difference in the number of captures I had among habitats. I captured more animals in the pinewoods habitat, followed by the sand dune habitat, and then the salt marsh habitat. Campbell and Christman (1979) found approximately the same number of captures between sandhill and scrub habitats dominated by pine trees. I also found a significant difference between 1999 and 2000. I believe my higher capture success in 2000 may have been due to increased rainfall in 2000 and drought-like conditions in 1999. I found that pitfall trap arrays were effective for capturing anuran and snake species. As suggested by Bounds (1996), I recommend using shade boards for pitfall and funnel traps to reduce mortalities of species captured in pitfall trap arrays. I also found that using lid rings for pitfall trap buckets as suggested by Vogt and Hine (1982) was effective in capturing the green tree frog. Bennett et al. (1970) reported that the eastern mud turtle, because of its size and land use, was more likely to be captured using the pitfall trap arrays than other larger turtle species. I also captured the eastern mud turtle using pitfall trap arrays in the salt marsh habitat.

Vogt and Hine (1982) recommended that the best times for trapping frogs,

toads and snakes was late April-early May, and early June for turtles. At CHNWR I found snakes and toads from late April-early August, frogs from mid April-late July, and turtles in mid July. I found that annual weather conditions will affect trapping success for amphibians and reptile species.

Cover board surveys

Mitchell et al. (1993) suggested that cover boards were an effective technique to assess the distribution, relative abundance, and habitat use by snakes on Assateague Island. However, I did not find that cover boards were effective in detecting species. After conducting the pilot study in 1998, when I had no captures using cover boards, I decided to target one species, the red-backed salamander (*Plethodon cinereus*). To target this species, I relocated cover board sites from the 3 major habitats in 1998 to 6 locations within just the pine woods habitat in 1999-2000. I decided to focus my efforts in the pinewoods habitat because the red-backed salamander occupies forested habitats, including pine forests on barrier islands (Mitchell and Anderson 1994). Conant et al. (1990) found several salamanders under rotting logs on Chincoteague. However, despite my efforts to sample the habitat for the red-backed salamander, I only captured 2 salamanders using cover boards from 1999-2000. I also captured Fowler's toads using cover boards. Given the time needed to use this technique and the low capture success, I do not recommend using cover boards.

Assateague Island is subject to constant overwash from storm activity. Also, prescribed burning is used as a management tool for the endangered Delmarva

Peninsula fox squirrel. Overwash and prescribed burning may make the forested habitat of CNHWR unsuitable for the red-backed salamander (Mitchell and Anderson 1994).

Some studies have indicated that amphibians and reptiles make no distinction between new cover boards and those aged over a few years (Grant et al. 1992). My cover boards were new in 1998 and aged throughout my project for 3 years. Due to very low capture success in all 3 years, I was unable to determine if age of the cover boards affected capture success.

One factor that may have affected my capture success for cover boards was the type of material I used and the design of my cover board arrays. Grant et al. (1992) used sheets of plywood shipboard and galvanized roofing tin measuring 0.66 m X 1.33 m in arrays of 60-250 boards to sample for salamanders. Tietje and Vreeland (1997) used plywood cover boards 1.3 X 61.0 X 61.0-cm. Heyer et al. (1994) suggested using larger cover boards to detect the greatest number of individuals and species. Heyer et al. (1994) did note that using larger cover boards might limit a researchers ability to check the boards or to have larger sample sizes. I chose to use smaller arrays to sample six areas and to estimate the ability of a single refuge biologist to conduct the technique. The small array size may have affected the number of species and individuals I captured. Due to my observations and those of other refuge staff, I believe the red-backed salamander population at CHNWR is very small. For example, the red-backed salamander has been observed at just 1 location at CHNWR for the past several years (I.Ailes, biologist, CHNWR, pers. commun). Mitchell and Anderson (1994) did not locate this species for Assateague Island.

Vogt and Hine (1982) recommend sampling for fossorial salamanders from late April-early May. I found the red-backed salamander from late fall (October)-early spring (May). If cover boards are used, I suggest conducting the cover board surveys once a month from September – December and March - May.

Cost comparison

Costs are generally an important factor in any research or monitoring program. I compared the total costs for the 6 sampling techniques and found road surveys were the most cost effective, followed by time-constrained searches, basking surveys, and aural surveys. I found that pitfall trap arrays and cover boards were the least cost effective techniques. Bounds (1996) compared the cost of pitfall traps and visual transects and also found that pitfall traps were more costly, yet yielded similar results as transects for lizards. Other researchers have suggested that pitfall traps are more costly and time-consuming than other techniques (Bury and Raphael 1993, Jones 1986, Corn and Bury 1990, Morrison et al. 1995, Record 1995). My results support these findings.

I found that cover board and pitfall trap array techniques were the most expensive, time-consuming, and labor intensive of the 6 techniques I used. I suggest that resource managers first determine the objectives of their research and monitoring programs and then select a sampling technique that will be effective in meeting their objectives within their time and funding constraints. I found that road surveys, aural surveys, and basking surveys were effective in detecting several amphibian and reptile species at minimal costs.

Comparison of techniques

I compared the cost, time, and sampling effectiveness of each of the six techniques. Short operation time, low costs for materials, data collection and maintenance, and effectiveness in species detection are ideal qualities for all monitoring programs, where time, money, and personnel are limited. My results suggest that the road survey may be the technique most beneficial to CHNWR. I found the road survey was the most effective method for anuran, snake, and turtle species detection. Like Jaeger et al. (1982), I did not find that road surveys were effective for the red backed salamander (*Plethodon cinereus*). The road survey was very cost effective, required minimal time and personnel, and could be done in conjunction with other refuge activities. My findings are similar to those of Campbell and Christman (1979), who determined the road survey was more productive than quadrat sampling, time-constrained searches, or pitfall trapping. Additional techniques would enhance the abilities of CHNWR to monitor long-term population trends. If resources are available, I suggest that aural surveys be used to monitor amphibians and that basking surveys be used for turtles.

The results of my study and previous studies seem to indicate a loss of species diversity on CHNWR (Hranitz et al. 1986). Despite using 6 sampling techniques, I did not find 4 species during my study that previously were found at CHNWR: New Jersey chorus frog (*Pseudacris triseriata kalmi*), green frog (*Rana clamitans melanota*), northern fence lizard (*Sceloporus undulatus hyacinthinus*), and northern brown snake (*Storeria dekayi dekayi*). The New Jersey chorus frog was last seen by

Lee (1972), the green frog by Conant et al. (1990), the northern fence lizard by Lee (1972), and the northern brown snake by Mitchell and Anderson (1994). The last rough green snake seen was on the north end of Assateague Island by Mitchell and Anderson (1994). I found one rough green snake (*Opheodrys aestivus*) during my study and it was dead. The amphibian and reptile species present on Assateague Island may experience local extirpation, but may be able to recolonize from mainland sources (Mitchell and Anderson 1994). The populations of amphibians and reptiles will be affected by management activities, human disturbance, weather, and the physical processes of a barrier island. To monitor population trends for amphibians and reptiles at Chincoteague National Wildlife Refuge, I recommend using 3 of the 6 sampling techniques: road surveys, aural surveys, and basking surveys.

Summary of techniques

Aural surveys

Aural surveys may be used to obtain presence or absence information and to develop relative abundance indices for vocalizing amphibians (males). While knowledge of species calls is required, these skills may be learned using call tapes and minimal training with experienced staff. Monetary assistance was available to refuges in the Northeast region of U.S. Fish and Wildlife Service in 1999 and 2000. Even if monetary assistance was not available, aural surveys were still a cost effective technique with an average seasonal cost of \$978.

Basking surveys

The basking survey is effective for detecting turtle species. No installation is needed, however a mowing program should be implemented to assure that visibility of basking areas is not affected by changes in vegetation. Basking surveys may be conducted with waterfowl surveys. The average seasonal cost was \$960.

Road surveys

The road survey was the most cost effective technique. Minimal personnel were required to conduct the survey and it can be used to detect a high number of species. The survey can be conducted by day or night depending on the species of interest. There is no maintenance or installation required. In addition, refuge staff may be trained to collect amphibians or reptiles (especially snakes) found dead on the road for study skins or subsequent identification. The average seasonal cost was \$485.

Time-constrained searches

Time-constrained searches require knowledge of the species in the area and are affected by the abilities of the observers. However, they can provide more detailed information on individual species. The average seasonal cost was \$858.50

Pitfall trap arrays

Pitfall traps arrays may be used to detect a large number of mostly traveling species. However, pitfall trap arrays were the most labor intensive and most

expensive technique. If not run properly pitfall trap arrays can result in a large number of mortalities. The average seasonal cost was \$2953.

Cover board surveys

The cover boards were not effective in determining species presence. I found that cover boards were labor intensive and yielded few results. I was not able to make recommendations on the type of material or size of array due to the low capture success in my study. If cover boards are used, I suggest conducting a pilot study using different materials and arrays covering a larger area. The average seasonal cost was \$1285.

Management recommendations for long-term monitoring of amphibians and reptiles at CHNWR

I recommend using a combination of sampling techniques to sample the herpetofauna at CHNWR. I developed 6 protocols for this study (Appendix 1). If managers want to capture individual animals to develop population estimates then pitfall trap arrays, cover board surveys, or time-constrained searches may be used in combination with mark and recapture techniques. Using mark and recapture techniques will increase the amount of time and labor costs for each technique. Because I observed more Fowler's toads than any other species, I believe that Fowler's toad would be the most effective species for developing population estimates using a mark and recapture data. For estimates of relative abundance using indices, I recommend aural surveys, basking surveys, or road surveys. I suggested

the advantages and disadvantages for each of the 6 sampling techniques (Appendix 2).

Individual Capture Techniques

Pitfall trap arrays

- I do not recommend using pitfall trap arrays at CHNWR because they are not cost effective and are labor intensive;
- If pitfall trap arrays are used, I recommend using the sites in the pinewoods habitat to maximize the number of animal observations;
- At least 3 pitfall trap arrays should be placed in the habitat; however, a lower number of sites may not yield sufficient captures for population estimates;
- The traps should be opened for at least 24 hrs once a month from April-July; additional trap nights would increase trap success.

Cover board surveys

- I do not recommend using cover boards at CHNWR because they were not cost effective and did not yield a large number of species;
- If cover boards are used, I recommend using a variety of sizes of plywood sheets or galvanized roofing in a pilot study to detect a higher number of species;
- At least 10 cover board sites should be established in the pinewoods habitat;
- The cover boards should be checked once a month from September –

December and March – May.

Time-constrained searches

- I do not recommend using time-constrained searches at CHNWR because they are time-consuming and I did not detect a high number of species using the technique;
- If the time-constrained search is used I recommend using the same sites I used in the sand dune and pinewoods habitat;
- Each site should be sampled for 10 minutes once a month from April – July.

Estimates of Relative Abundance

Aural surveys

- I recommend using 13 of the established survey points rather than the 25 points I used;
- Financial assistance makes this survey cost effective and supports the Region 5 initiative for amphibian and monitoring programs;
- I recommend that the survey be conducted once a month from March-May.

Basking surveys

- I recommend that basking surveys should be conducted in conjunction with the refuge's waterfowl survey route of the 14 MSMU's at CHNWR;
- The survey can provide useful trend information on turtle populations;
- I recommend conducting the surveys once a month from May – July at

each MSMU's "Apool", "Bnorth", and "Bsouth".

Road surveys

- I recommend using road surveys because they were the most cost effective technique for information on population trends;
- The survey should be conducted during the day and at night;
- It may be conducted in conjunction with the aural survey and with the refuge's waterfowl survey route;
- It should be used in conjunction with other sampling techniques if resources are available.

For specific information on green tree frogs, southern leopard frogs, Fowler's toads, or bullfrogs I recommend aural surveys. For information on freshwater turtles that commonly bask such as eastern painted turtles and red-bellied turtles I recommend using basking surveys. To obtain information on freshwater turtles that don't commonly bask, and on snakes I recommend using road surveys. I do not recommend the use of the pitfall trap arrays, cover boards, or time-constrained searches for CHNWR (Table 17). I found these 3 techniques to be labor intensive, time-consuming, expensive, and not effective in species detection.

These recommendations are based on the results of my study and are intended to be guidelines for natural resource managers. I recommend that resource managers first establish statistical criteria for their monitoring goals and then evaluate any monitoring program after 5 years of data collection to see if the criteria were met.

Appendix 1

Protocols

Aural Survey

When

- Surveys will begin in early spring when air temperatures are above 8°C and amphibians have initiated calling activity
- Surveys will begin one half-hour after sunset and will end before midnight.

How

- Data will be collected at 13 predetermined points located along the service road of the refuge. Points are placed approx. ½ mile apart.
- Data will be collected using a 3-minute observation period at each sample point. Upon arrival, a silent pause of at least one-minute before the sample period will allow the frogs to return to normal calling activity.
- Calls will be identified to species, and the intensity of each call is done by using the Wisconsin Index Value.

Data Collection

Before you leave be sure to get data sheets

At the first site record date, observer, start time, and initial weather data

At each site:

- Any change in weather conditions
- Any additional information

- Species heard and index

When finished record:

- Final weather information
- Stop time

This technique is used to identify which species are present and what number of each species is present in the population.

Time-constrained search

When

- Survey for a 30-minute period of time without confining the search to any given area
- Time-constrained surveys will be conducted once a week

How

- We will systematically look under logs and other suitable substrate to find any target species.
- Surveyor will look for 10 minutes at each survey site, using appropriate tools such as potato rake, bar for lifting rocks, etc.
- Captured animals are identified and released after the 10 minute period is finished
- Animals found are identified to species, measured, weighed, and released.
- Additionally, care is given to returning the substrate back to its original condition to minimize habitat disturbance.

Data Collection

At the first site:

- Record data, observer, start time, and initial weather data
- Systematically survey area for 10 minutes

When finished record stop time

This technique can be used to spot any of the possible target species.

Road Survey

When

- Road surveys are conducted to and from each survey site approximately four times per week.
- Optimal conditions are during a fine mist or the day after a rain

How

- Done by driving the roads of the refuge approximately 20 mph to and from each survey site while traveling to do other surveys
- When an amphibian or reptile is spotted, stop the vehicle and try to identify it. If you are unable to identify it get out to take a closer look (especially good idea for frogs/toads).
- Take note of the odometer reading and any landmarks for easy relocation on a map.

Data Collection

Before you leave be sure to

- Get data sheets

- Record date, observer, start time, starting odometer, and initial weather data

At each site record:

- Number of each species present on the road, or visible from the road
- Odometer reading
- Any additional information

When finished record:

- Final weather information
- Stop time
- Odometer reading

This technique can be used to encounter any of the target species moving among habitats. This will also provide an estimate of road kills. It is most effective for turtles, snakes, frogs, or toads.

Turtle basking survey

When

- Warm/sunny days
- Start no earlier than 8am and no later than 12noon
- Survey all MSMU's once a week

How

- Done by driving road to and around (where possible) all accessible MSMU's

- When a turtle is spotted, stop the vehicle and try to identify it. Use the vehicle as a blind. If you are unable to identify any turtle, identify all others in the area, then slowly move closer. If you get too close, the turtle may drop into the water.
- Make sure you check all logs, stumps, and shorelines for basking turtles
- Record any unidentified as well as identified turtles

Data collection

Before you leave be sure to:

- Get data sheets
- Record date, observer, start time, and initial weather data

At each site record:

- Number of turtles of each species present
- Approximate water level (or if possible record actual level)
- Any additional information

When finished record:

- Final weather information
- Stop time

Pitfall trap arrays

When

- The pitfall trap arrays will be opened for 72 hrs each week, and will be checked every 24 hrs before noon to minimize stress to animals captured.

- Each trap will be closed in the same order it was opened to ensure the same trapping effort in each habitat.

How

- There should be *at least* two (preferably three) pitfall array traps set up in each of the dominant vegetative communities. Each array consists of four 19L plastic buckets and six funnel traps. Each array is designed to direct any amphibian or reptile traffic down the sides of the flashing into one of the four pitfall buckets or funnel traps. Buckets will be buried flush with the ground and with the end of the array. The array is made with metal flashing cut to be approximately 5 m in length, and each array is buried about 15 cm in the ground to prevent any animals from tunneling under them. The array can also be made of simple sediment control fencing.
- Animals captured will be identified to species, measured, weighed, marked for recapture, and released approximately 2 m from the pitfall trap array.
- Enough water will be placed in the buckets to moisten the bottom every time they are checked to aid in preventing desiccation (Vogt and Hine 1982).
- Wooden cover can be constructed for each pitfall bucket and each funnel trap. This will also aid in preventing desiccation and predation.
- Funnel traps are constructed of window screen and shaped into a tube with inward directed funnel shaped openings at both ends.
- Funnel traps are placed on the ground parallel to the array at the center of the array on both sides (Heyer 1994). As snakes or amphibians move along the

side of the array they will move into the funnel trap and not be able to back out.

- Sponges are placed in the funnels to prevent desiccation and are rewet every time the array is checked.

Data Collection

- When you start:
- Record date, observer, start time, and initial weather data
- Record the number of each species present
- Caught in Bucket or Funnel
- Weight, SVL, sex (if possible), Adult/Juv, TL, etc.(as indicated on the data sheet
- Stop time

Cover board survey

When

- Best if done after a rain or during a light mist or drizzle.
- Best if boards are left in place for months prior to first survey
- Boards can be put into place and left indefinitely

How

- Wood cover boards are cut into 1 ft X 1 ft sections and placed in 7 x 3 arrays.
This can be increased according to management/research needs elsewhere.
Also, large sections of sheet metal or large pieces of plywood can be installed.
- Preferable to have at least 3 sites per habitat.

- It is important to know the life history of the species you are targeting with this survey. For example sampling for the red-backed salamander (*Plethodon cinereus*), on the dunes would not be appropriate.
- Leaf litter is to be moved aside for boards to be put flush with the ground.

Data Collection

At the first site:

- Record date, observer, start time, and initial weather data
- Check each board individually for any target species
- Lightly finger through the top of the soil to the moist areas below most likely to have species present.
- Any species captured should be measured, weighed, identified to species, and released about 2 m from the site.

When finished:

- Replace boards gently into same position.
- Record final weather information
- Stop time

Creating artificial cover in a habitat can be used to sample amphibians and reptiles by mimicking natural cover that the animals frequently use. This design is similar to what is recommended by Heyer et al. (1994). The boards will remain in place for the entire research period. This technique can be used to detect any species that may use cover such as snakes, frogs, or toads.

Appendix 2

Review of sampling techniques

Aural Survey

Advantages

- Monetary assistance available for Region 5 refuges
- Volunteers can be trained on calls when only a few species
- Data can be taken by one observer
- Can be done in conjunction with other survey routes (i.e. Breeding Bird Survey, turtle basking survey, waterfowl survey)
- Requires little to no maintenance; only of markers that fall down
- Effective for detecting most of the amphibian species present
- Minimal costs to conduct by refuge personnel

Disadvantages

- Observers must have full hearing ability and experience when a large number of calls may be heard
- Cannot be used to detect reptile species
- Cannot be used to detect females or males of species that do not call (Fellers 1979, Heyer et al. 1994)
- Actual number of males cannot be determined in large choruses
- Must conduct over the entire season to hear all species; explosive breeders may be missed

Basking survey

Advantages

- Is effective for detecting a large number of turtle species
- Only one observer required

- Can be done in conjunction with other refuge activities (such as waterfowl surveys)
- Minimal costs
- No time required for set up
- No maintenance

Disadvantages

- Knowledge needed to identify basking species visually
- Vehicle may disturb basking turtles at some sites

Time-constrained search

Advantages

- Minimal cost for materials
- Researchers may detect a large number of individuals and species
- Best for sampling species that are rare or less likely to be caught in traps

Disadvantages

- Cannot give insight into the total number of species present
- Survey is easier in open habitat rather than dense habitat
- Differences in observer abilities will bias results
- Habitat disturbance (but may be minimized by restoring debris to original locations)

Road survey

Advantages

- Effective for detecting highly mobile species

- Provides an estimate of species richness and relative abundance
- Can be done in conjunction with other daily refuge activities
- Effective for detecting a large number species
- Requires only one observer
- Minimal costs

Disadvantages

- Not effective for animals with small home ranges
- A road is needed so this may not be appropriate for wilderness areas or areas without roads
- Small species are harder to see on different road surfaces
- Identification skills are needed by observer

Pitfall trap arrays

Advantages

- Effective for detecting a large number of individuals and species
- May be left in place over time
- Useful for long-term studies
- May use volunteers to install and maintain traps

Disadvantages

- Time consuming
- Labor intensive
- Expensive
- Difficult to install in rocky areas, areas with a lot of roots, or hard substrate

- Requires the most amount of maintenance (dune habitat at CHNWR)
- Disturbs the habitat
- Capture mostly traveling animals
- If not run properly can result in a large number of mortalities associated with captures
- Hard to detect species that can climb easily
- Predation

Cover board survey

Advantages

- Limited disturbance to habitat
- Minimal time required
- Limited training needed
- Easy to construct

Disadvantages

- Detects few individuals and species
- Type of material may affect species detected
- Depends on factors such as availability of cover (natural), weather, habitat needs
- May be difficult to re-locate in fast growing vegetation
- May be disturbed by ponies, feral pigs, ground squirrels, wind, or falling branches (Tietje and Vreeland 1997)

TABLES

Table 1: Species recorded on Assateague and Chincoteague Islands (Conant et al. 1990).

Species	Author	Date	Location
Fowler's toad; <i>Bufo woodhousii fowleri</i>	Brady	1925	Chincoteague & Assateague
Green tree frog; <i>Hyla cinerea</i>	Lee	1972	Assateague
Bullfrog; <i>Rana catesbeiana</i>	Mitchell and Pague	1986	Assateague
New Jersey chorus frog; <i>Pseudacris triseriata kalmi</i>	Lee	1973	Assateague
Green frog; <i>Rana clamitans melanota</i>	Ernst & Gotte	1984	Assateague
Southern leopard frog; <i>Rana sphenoccephala</i>	Lee	1972	Assateague
Red-back salamander; <i>Plethodon cinereus</i>	Bridwell	1938	Chincoteague
Northern fence lizard; <i>Sceloporus undulatus hyacinthinus</i>	Lee	1972	Assateague
Northern black racer; <i>Coluber constrictor constrictor</i>	Brady	1925	Assateague
Southern ring neck snake; <i>Diadophis punctatus edwardsii</i>	Bridwell	1938	Chincoteague
Black rat snake; <i>Elaphe obseleta obseleta</i>	Gibbons & Coker	1978	Assateague
Eastern hognose snake; <i>Heterodon platirhinos</i>	Fowler	1925	Chincoteague & Assateague
Rough green snake; <i>Opheodrys aestivus</i>	Lee	1972	Assateague
Northern brown snake; <i>Storeria dekayi dekayi</i>	Conant & Collins	1991	Chincoteague & Assateague
Northern water snake; <i>Nerodia sipedon sipedon</i>	Lee	1972	Chincoteague & Assateague
Spotted turtle; <i>Clemmys guttata</i>	Lee	1972	Assateague
Eastern box turtle; <i>Terrapene carolina carolina</i>	Lee	1973	Assateague
Eastern mud turtle; <i>Kinosternon subrubrum subrubrum</i>	Fowler	1925	Chincoteague & Assateague
Diamondback terrapin; <i>Malaclemys terrapin terrapin</i>	Lee	1972	Chincoteague & Assateague
Eastern painted turtle; <i>Chrysemys picta picta</i>	Lee	1972	Assateague
Red-bellied turtle; <i>Pseudemys rubriventris</i>	Lee	1972	Assateague
Loggerhead sea turtle; <i>Caretta caretta</i>	Brady	1925	Assateague
Atlantic green turtle; <i>Chelonia mydas mydas</i>	Schwartz	1960	Chincoteague
Common snapping turtle; <i>Chelydra serpentina serpentina</i>	Lee	1972	Assateague & Chincoteague
Atlantic ridley turtle; <i>Lepidochelys Kempi</i>	VIMS	1980	Seen offshore
Leatherback sea turtle; <i>Dermochelys coriacea</i>	VIMS	1980	Seen offshore

Table 2. Weather data recorded for 1998-2000 at Chincoteague National Wildlife Refuge

Weather Data (in Fahrenheit)									
MONTH	Maximum temperature			Minimum temperature			Total precipitation (inches)		
	1998	1999	2000	1998	1999	2000	1998	1999	2000
Jan	65.8	70.3	62.8	22.6	15.8	14	7.35	3.49	4.29
Feb	58.6	62.6	72.1	25	23.7	23.4	5.62	2.59	1.94
Mar	83.7	72.5	80.2	21	21.2	30.9	4.31	7.46	5.56
April	77.4	82.4	74.5	38.1	36.9	36.9	2.51	3	4.39
May	89.6	90.9	88.3	43.9	47.3	47.7	5.27	1.87	3.05
June	91.6	96.6	91.8	54.3	54.9	54	3.93	2.04	2.07
July	97.5	99.1	91.9	63.3	63	61	1.5	3.28	7.02
Aug	93	94.5	92.1	62.1	61.3	55.4	1.49	3.44	8.54
Sept	90.3	83.3	0	57.9	58.4	0	2.16	7.3	0
Oct	85.8	77.9	0	43.9	43.9	0	1.29	6.44	0
Nov	68.7	73.9	0	32.7	31.3	0	1	0.74	0
Dec	76.6	66.4	0	23	22.3	0	4.66	2.21	0
Average	81.55	80.87	81.71	40.65	40.00	40.41	3.42	3.66	4.61

Table 3. Habitat associations by sampling technique and site name at Chincoteague National Wildlife Refuge 1998-2000.

	Pitfall trap arrays	Time-constrained search	Cover board survey	Aural, Basking, and Road surveys
Site Name	Habitat Type	Habitat Type	Habitat Type	
SD3	Sand Dune/ Pine woods interface			
SD4	Sand Dune/ Pine woods interface			
White Hills	Pine Woods			
PW5	Pine Woods			
Ragged point	Salt Marsh/ Pine Woods interface			
Sow Pond	Salt Marsh/ Pine Woods interface			
Weather station		Pine Woods/breeding site		
Turnaround		Pine Woods/breeding site		
Cdike		Sand dune interface/MSMU borrow ditch		
SBCT		Sand dune interface/breeding site		
Pony Corral		Salt Marsh/ Pine Woods interface		
Boat dock		Salt Marsh/ Pine Woods interface		
Wills Hole			Pine Woods	
Sow Pond			Salt Marsh/ Pine Woods interface	
White Hills			Pine Woods	
PW6			Pine Woods	
Gravel Pile			Pine Woods	
Maintenance			Pine Woods	
Mapped route				Transversed all habitat types

Table 4. List of possible species likely to occur at Chincoteague National Wildlife Refuge.

Species	Scientific Name
Anurans	
Fowler's toad	<i>Bufo woodhousiii fowleri</i>
Green tree frog	<i>Hyla cinerea</i>
Bullfrog	<i>Rana catesbeiana</i>
New Jersey chorus frog	<i>Pseudacris triseriata kalmi</i>
Green frog	<i>Rana clamitans melanota</i>
Southern leopard frog	<i>Rana sphenoccephala</i>
Salamanders	
Red-back salamander	<i>Plethodon cinereus</i>
Snakes	
Northern black racer	<i>Coluber constrictor constrictor</i>
Black rat snake	<i>Elaphe obseleta obseleta</i>
Eastern hognose snake	<i>Heterodon platirrhinos</i>
Rough green snake	<i>Opheodrys aestivus</i>
Northern brown snake	<i>Storeria dekayi dekayi</i>
Northern water snake	<i>Nerodia sipedon sipedon</i>
Turtles	
Spotted turtle	<i>Clemmys guttata</i>
Eastern box turtle	<i>Terrapene carolina carolina</i>
Eastern mud turtle	<i>Kinosternon subrubrum subrubrum</i>
Diamondback terrapin	<i>Malaclemys terrapin terrapin</i>
Eastern painted turtle	<i>Chrysemys picta picta</i>
Red-bellied turtle	<i>Pseudemys rubriventris</i>
Common snapping turtle	<i>Chelydra serpentina serpentina</i>
Sea Turtles	
Loggerhead sea turtle	<i>Caretta caretta</i>
Atlantic green turtle	<i>Chelonia mydas mydas</i>
Atlantic ridley turtle	<i>Lepidochelys kemp</i>
Leatherback sea turtle	<i>Dermochelys coriacea</i>
Totals	
Frogs and toads	6
Salamanders	1
Snakes	6
Turtles	11

Table 5. Possible species that could be detected using different sampling techniques at Chincoteague National Wildlife Refuge 1998-2000.

Techniques	Frogs & Toads	Salamanders	Snakes	Turtles (not including sea turtles)	Total possible species
Aural survey	1 toad, 5 frogs	None	None	none	6
Basking survey	None	None	None	7 turtles	7
Time-constrained search	1 toad, 5 frogs	1 salamander	6 snakes	7 turtles	20
Road survey	1 toad, 5 frogs	None	6 snakes	7 turtles	20
Pitfall trap array	1 toad, 5 frogs	None	6 snakes	7 turtles	20
Cover board survey	1 toad	1 salamander	None	none	2
TOTAL	6	1	6	7	20

Table 6. Total number of observations made per species by technique at Chincoteague National Wildlife Refuge during the pilot study in 1998. (PF= pitfall trap array, TCS= time-constrained search, RS= road survey, AS= aural survey, CS= cover board survey, BS= basking survey)

Common name	Scientific name	PF	TCS	RS	AS	CS	BS
Green tree frog	<i>Hyla cinerea</i>	100	1	1	detected		
Fowler's toad	<i>Bufo woodhousii fowleri</i>	2			detected		
Southern leopard frog	<i>Rana sphenoccephala</i>	3				No species recorded	Not conducted
Northern black racer	<i>Coluber constrictor constrictor</i>	2					
Eastern hognose snake	<i>Heterodon platyrhinos</i>			1			
Northern water snake	<i>Nerodia sipedon sipedon</i>			1			
Eastern painted turtle	<i>Chrysemys picta picta</i>			1			
Eastern mud turtle	<i>Kinosternon subrubrum subrubrum</i>			3			
Common snapping turtle	<i>Chelydra serpentina serpentina</i>			7			
TOTAL		107	1				

Table 7. Weather conditions recorded at peak observation times for Chincoteague National Wildlife Refuge 1999-2000.

Technique	Time of Day	Temperature Range	Wind Speed	Cloud Cover	Precipitation (% of sample days w/rain present the last 24hrs)
Aural surveys	After sunset	12.2°C - 19.4°C	4-7	20%	17%
Basking surveys	Morning	8.8°C -27.9°C	0-1	5%	39%
Time-constrained searches	After sunset	20.5°C -25°C	0-1	30%	100%
Road surveys	Day and Night	18°C -32.2°C	4-7	30%	28%
Pitfall trap arrays	Afternoon				
Cover board surveys	Afternoon	9.8°C -16.9°C	4-7	50%	50%

Table 8. Total number of species detected at each point by month and year on the aural survey route at Chincoteague National Wildlife Refuge 1999-2000.

Point #	# species detected by month												Total # species detected by year		
	2000														
	1999												1999	2000	
Feb	Mar	April	May	Jun	Jul	Aug	Feb	Mar	April	May	Jun	Jul	Aug		
1			1	2	2	1	1		1	2	2	2	1	3	2
2	1		1	2	2		1	1	1	1	2	1	1	3	3
3	1	1	1	2	2	1		1	1	1	2	1	1	3	3
4	1	1	1	2		2		1	1	1	2	1		3	3
5	1	1	2	2	2		1	1	1		2	1		3	3
6	1	1	2	2		1		1	1		2	1		3	2
7	1	1	2	3	2	1	1	1	2	2	2	2	1	4	3
8	1		2	2	1	1	1	1	1	2	2	1	1	3	3
9			1	1					1		2	1	1	2	1
10			1	1	1	1					2			2	2
11	1		2	1	1	2		1	2		2	1		3	3
12	1	1	1	2	1	1	1	1	1		2			3	3
13			1	2	2			1	1		2		1	2	3
14			2	2	1	1		1	1	1	2	1	2	3	3
15	1		2	3	2		2	1	2	1	2	2	1	4	4
16	1	1	2	3	2	2	2	1	1	1	2	2	1	4	4
17	1	1	2	2	1	1	1		1	1	2	2		3	3
18	1	1	3	2	2	2		2	2	1	2	2		3	3
19	1		2	2	2	1	2	1	2	2	2	2		3	3
20			2	2		1	1	1	2	2	2	2	1	3	3
21	1		2	1	2			1	1	1	2			3	3
22			2	2		1		1	1	1	2			3	3
23			1	2	2	1		1	1	1	2			3	3
24		1	1	2	3	1		1	1	2	3	2	1	3	4
25			1	1		1					2			1	2

Table 9. Calling phenologies of the amphibian species at Chincoteague National Wildlife Refuge indicated by the intervals species were heard on aural survey routes for 1999-2000.

February	March	April	May	June	July	August
↔	SOUTHERN LEOPARD FROG ↔	↔				
	↔	FOWLER'S TOAD				↔
		↔	GREEN TREE FROG			↔
			↔ BULLFROG	↔		

Table 10. Spearman rank correlation relative abundance indices based on aural survey routes at Chincoteague National Wildlife Refuge 1999-2000. (GTF=green tree frog; *Hyla cinerea*, SLF= southern leopard frog; *Rana sphenoccephala*, FT= Fowler's toad; *Bufo woodhousii fowleri*, and BF= bullfrog; *Rana catesbeiana*).

Species	Rank ^a	Index ^b
GTF	2	0.0607
SLF	3	0.0140
FT	1	0.0986
BF	4	0.0012

^aBased on 19 aural surveys; Index = $\frac{\# \text{ routes species was detected} \times \# \text{ survey points species was detected}}{\text{total \# routes} \times \text{total \# survey points}}$

EX. $\frac{\text{number of routes FT was found in 1999+2000} \times \text{number of points FT was found in 1999+2000}}{(23 \text{ in } 1999 + 23 \text{ in } 2000) \times 46} = \frac{25 \text{ survey points} \times 46 \text{ routes from 1999 and 2000}}{46 \times 1150}$

$$= \frac{32 \times 163}{46 \times 1150}$$

$$= 0.0986$$

^b Spearman rank correlation $\phi = 0.0716$, $P < 0.001$

Table 11. Total number of observations made per species by technique at Chincoteague National Wildlife Refuge 1999-2000. (PF= pitfall trap arrays, TCS= time-constrained searches, RS= road surveys, AS= aural surveys, CS= cover board surveys, BS= basking surveys).

Common name	Scientific name	PF	TCS	RS	AS	CS	BS
Green tree frog	<i>Hyla cinerea</i>	1	22				
Fowler's toad	<i>Bufo woodhousii</i>	53	11	8	individuals not counted	6	
Southern leopard frog	<i>Rana sphenocephala</i>	11	21				
Bullfrog	<i>Rana catesbeiana</i>		40				
Red-backed salamander	<i>Plethodon cinereus</i>					2	
Northern black racer	<i>Coluber constrictor constrictor</i>	5		3			
Eastern hognose snake	<i>Heterodon platyrhinos</i>	2		2			
Northern water snake	<i>Nerodia sipedon sipedon</i>						
Black rat snake	<i>Elaphe obseleta obseleta</i>			6			
Red-bellied turtle	<i>Pseudemys rubriventris</i>			1			68
Diamondback terrapin	<i>Malaclemys terrapin terrapin</i>			14			11
Eastern painted turtle	<i>Chrysemys picta picta</i>			16			143
Eastern box turtle	<i>Terrapene carolina carolina</i>			1			1
Eastern mud turtle	<i>Kinosternon subrubrum subrubrum</i>	1		3			2
Common snapping turtle	<i>Chelydra serpentina serpentina</i>		1	12			20
UNID	Unidentified		9				6
TOTAL		73	104	66		8	251

Table 12: Comparison of monthly observations using basking surveys at Chincoteague National Wildlife Refuge for 1999-2000.

Interval	Total # species observed	95% CI	MEAN	SD
March-April	(3,4)-(3,3)	2.5-4.0	3.25	0.50
April- May	(3,3)-(3,3)	3.0-3.0	3.0	0.00
May- June	(3,3)-(4,4)	2.6-4.4	3.5	0.58
June-July	(4,4)-(2,5)	1.7-5.8	3.75	1.25
July-August	(2,5)-(1,1)	-0.76-5.3	2.25	1.89

Table 13. Spearman rank correlation ratios and ranks for road surveys at Chincoteague National Wildlife Refuge from 1999-2000.

Route	Ratio ^a	Rank ^b
4/28/99	0.053	4.5
4/29/99	0.053	4.5
4/30/99	0.053	4.5
5/7/99	0.053	4.5
5/28/99	0.368	1
6/2/99	0.158	2.5
6/30/99	0.158	2.5
7/15/99	0.053	4.5
7/22/99	0.053	4.5
4/25/00	0.048	4.5
4/26/99	0.143	2.5
4/27/00	0.095	6.5
5/6/00	0.095	6.5
5/15/00	0.048	4.5
6/23/00	0.095	6.5
6/30/00	0.095	6.5
7/11/00	0.143	2.5
7/13/00	0.238	1

^aBased on 9 survey routes; Ratio= $\frac{\text{total \# observations per route}}{\text{total \# observation per year}}$

^bSpearman Rank Correlation $\varphi = 0.0769$, $P < 0.001$

Table 14: Comparison of cost, sampling time, and maintenance for 6 techniques used to sample reptiles and amphibians at Chincoteague National Wildlife Refuge from 1998-2000.

Technique	Materials	Labor to install	Labor to collect data	Maintenance	Total costs
Aural surveys	\$193	\$80	\$705	No maintenance	\$978
Road surveys	\$0	\$0	\$485	No maintenance	\$485
Time-constrained searches	\$183.50	\$60	\$615	No maintenance	\$858.50
Pitfall trap arrays	\$1314	\$669	\$520	\$450	\$2953
Basking surveys	\$225	\$60	\$675	No maintenance	\$960
Cover board surveys	\$350	\$300	\$615	\$20	\$1285

Table 15. Total number of species detected using 6 techniques at Chincoteague National Wildlife Refuge 1998-2000.

Technique	Frogs/Toads	Snakes	Salamanders	Turtles	Total
Pitfall trap arrays	3	2	0	1	6
Aural surveys	4	0	0	0	4
Basking surveys	0	0	0	6	6
Road surveys	1	3	0	6	10
Cover board surveys	1	0	1	0	2
Time-constrained searches	4	0	0	1	5

Table 16: Diversity indices calculated using the Shannon-Weaver diversity index, $H^i = \sum (P_i \ln(P_i))$, where P_i = the total number of individuals observed comparing 6 sampling techniques at Chincoteague National Wildlife Refuge from 1998-2000.

Sampling Technique	H^i
Road surveys	1.914
Time-constrained searches	1.443
Basking surveys	1.037
Cover board surveys	0.566
Pitfall trap arrays	0.517
Aural surveys	undetermined

Table 17. Amphibian and reptile species detected using each technique at Chincoteague National Wildlife Refuge 1999-2000. PF= pitfall trap arrays, CS= cover board surveys, AS= aural surveys, RS= road surveys, BS= basking surveys, TCS= time-constrained searches, and Other= observations not associated with the research project.

Species	PF	CS	AS	RS	TCS	Other
Family Bufonidae						
Fowler's toad, <i>Bufo woodhousii fowleri</i>	X	X	X	X	X	X
Family Hylidae						
Green tree frog, <i>Hyla cinerea</i>	X		X		X	X
Family Ranidae						
Southern leopard frog, <i>Rana sphencephala</i>	X		X		X	X
Bullfrog, <i>Rana catesbeiana</i>			X		X	
Family Plethodontidae						
Red-backed salamander, <i>Plethodon cinereus</i>		X				X
Family Colubridae						
Northern black racer, <i>Diadophis punctatus punctatus</i>	X			X		X
Eastern hognose snake, <i>Heterodon platirhinos</i>	X			X		X
Northern water snake, <i>Nerodia sipedon sipedon</i>				X		
Black rat snake, <i>Elaphe obsoleta obsoleta</i>				X		X
Rough green snake, <i>Opheodrys aestivus</i>				X		X
Family Chelydridae						
Common snapping turtle, <i>Chelydra serpentina serpentina</i>				X	X	X
Family Emydidae						
Eastern painted turtle, <i>Chrysemys picta picta</i>				X		X
Spotted turtle, <i>Clemmys guttata</i>						X
Red-bellied turtle, <i>Pseudemys rubriventris</i>				X		X
Diamondback terrapin, <i>Malaclemys terrapin terrapin</i>				X		X
Eastern box turtle, <i>Terrapene carolina carolina</i>						X
Family Kinosternidae						
Eastern mud turtle, <i>Kinosternum subrubrum subrubrum</i>	X			X		X

Figures

Figure 1: Location of Chincoteague National Wildlife Refuge (CHNWR).

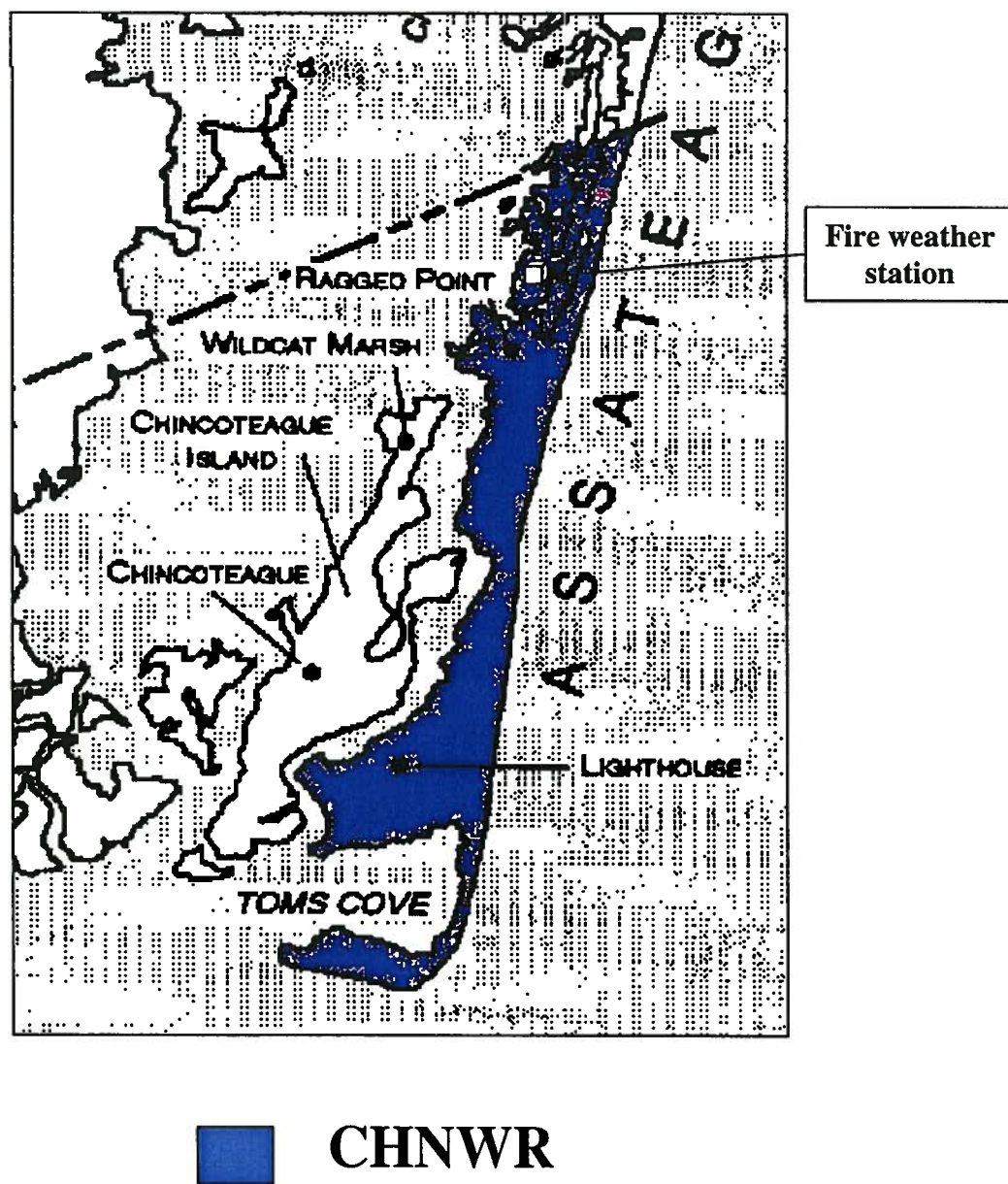
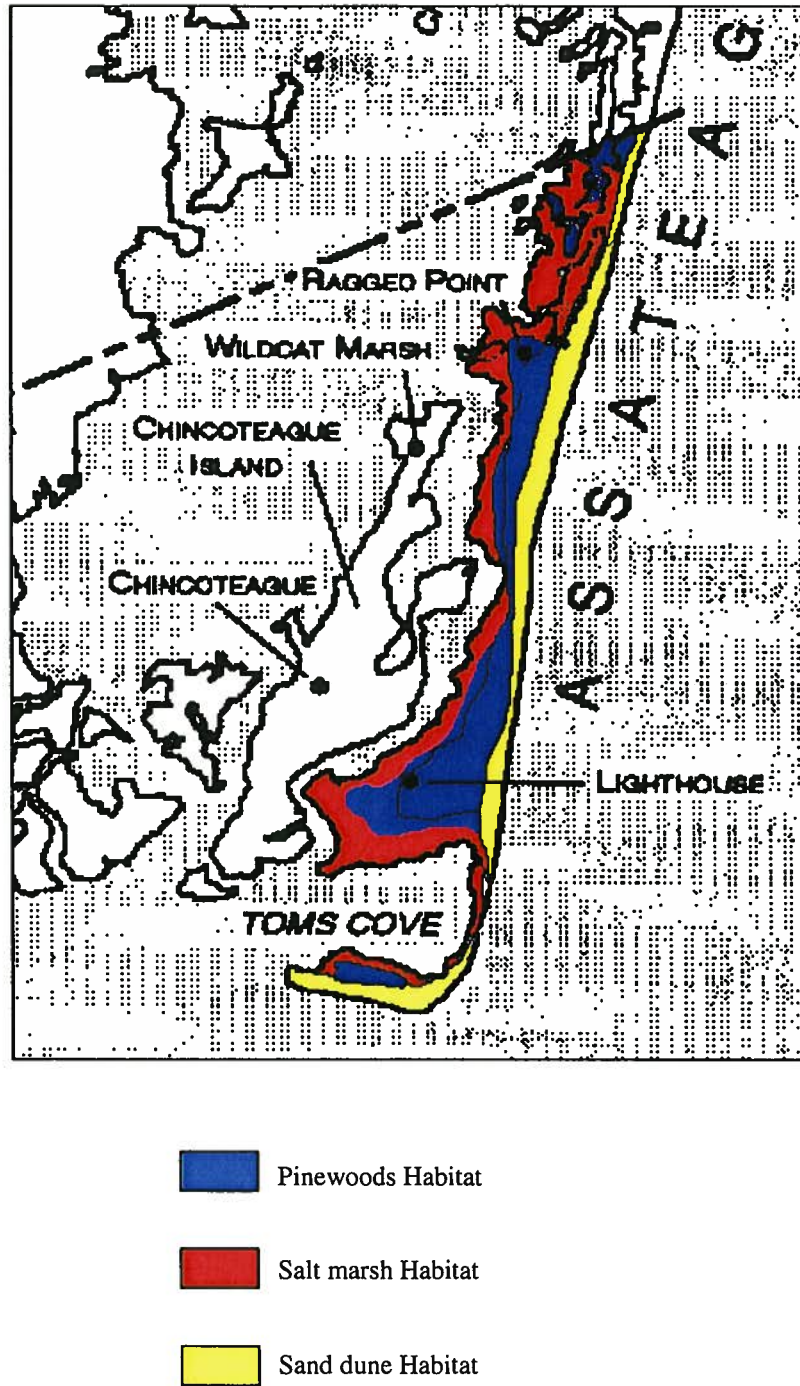


Figure 2: Three dominant vegetative communities at Chincoteague National Wildlife Refuge, 1998-2000.



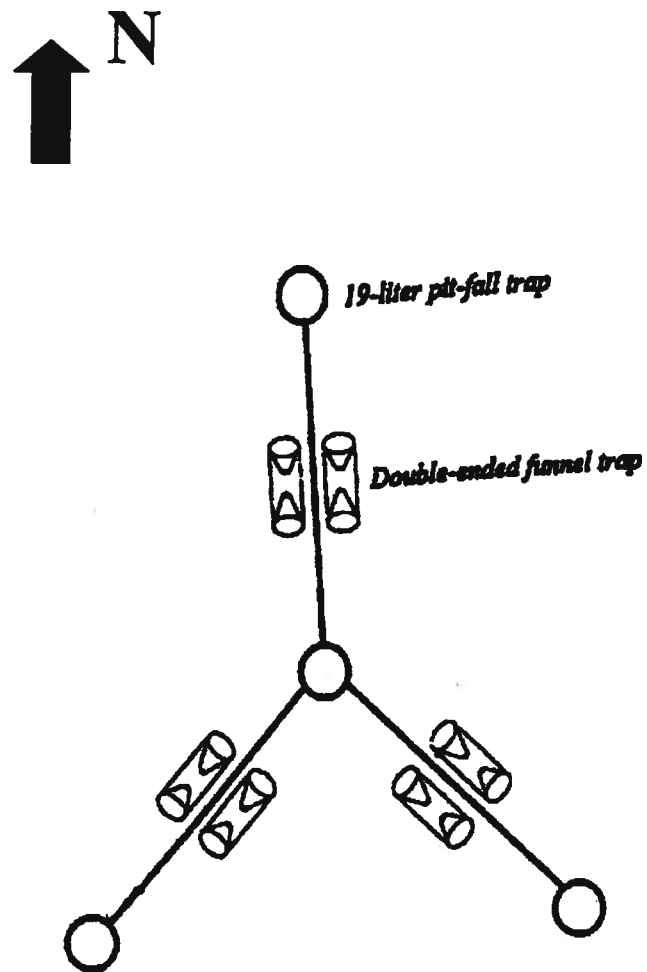


Figure 3: Design and alignment of the pitfall trap array with funnel traps as modified from the designs suggested by Jones (1981) and Bury and Corn (1987) used at Chincoteague National Wildlife Refuge 1998-2000.

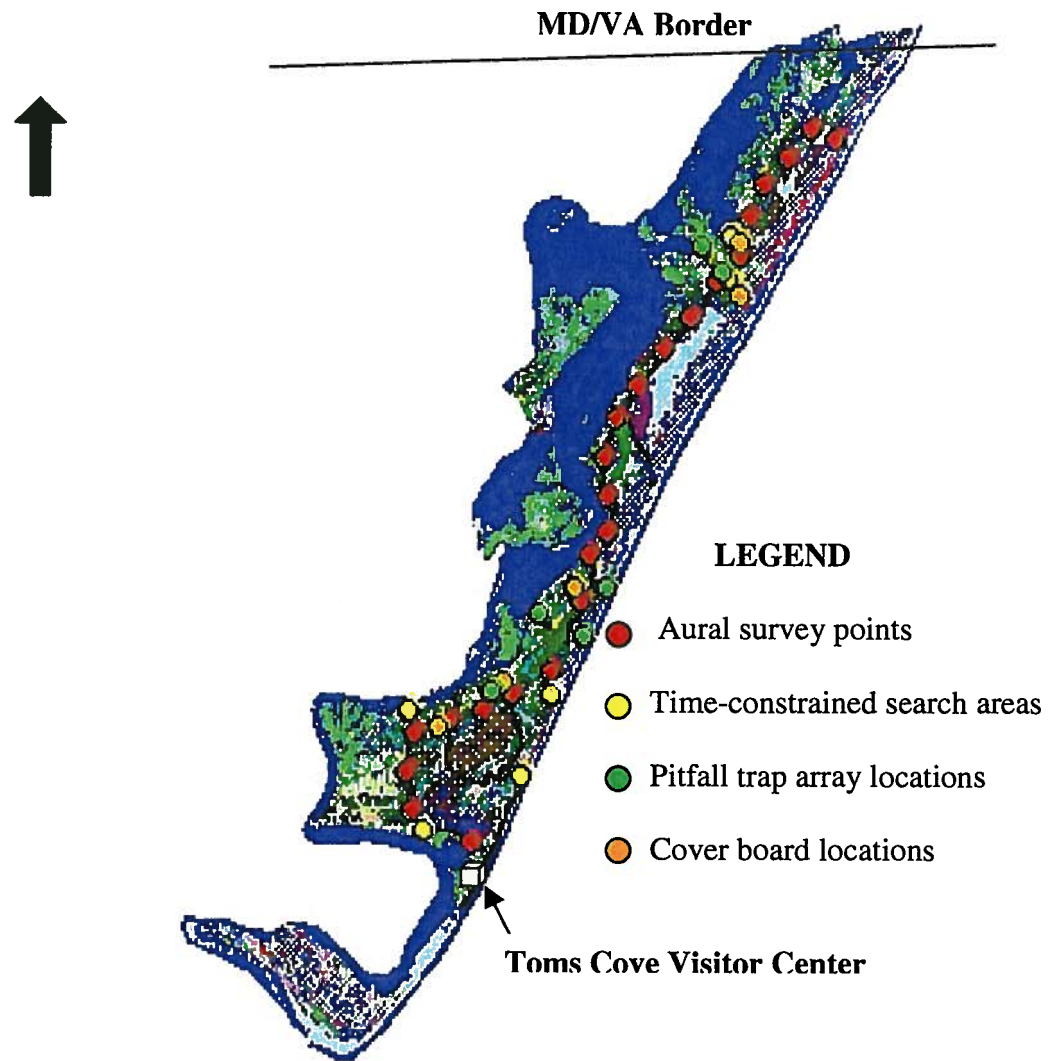


Figure 4: Location of pitfall trap arrays, time-constrained search areas, aural survey points and cover board locations at Chincoteague National Wildlife Refuge, 1998-2000.

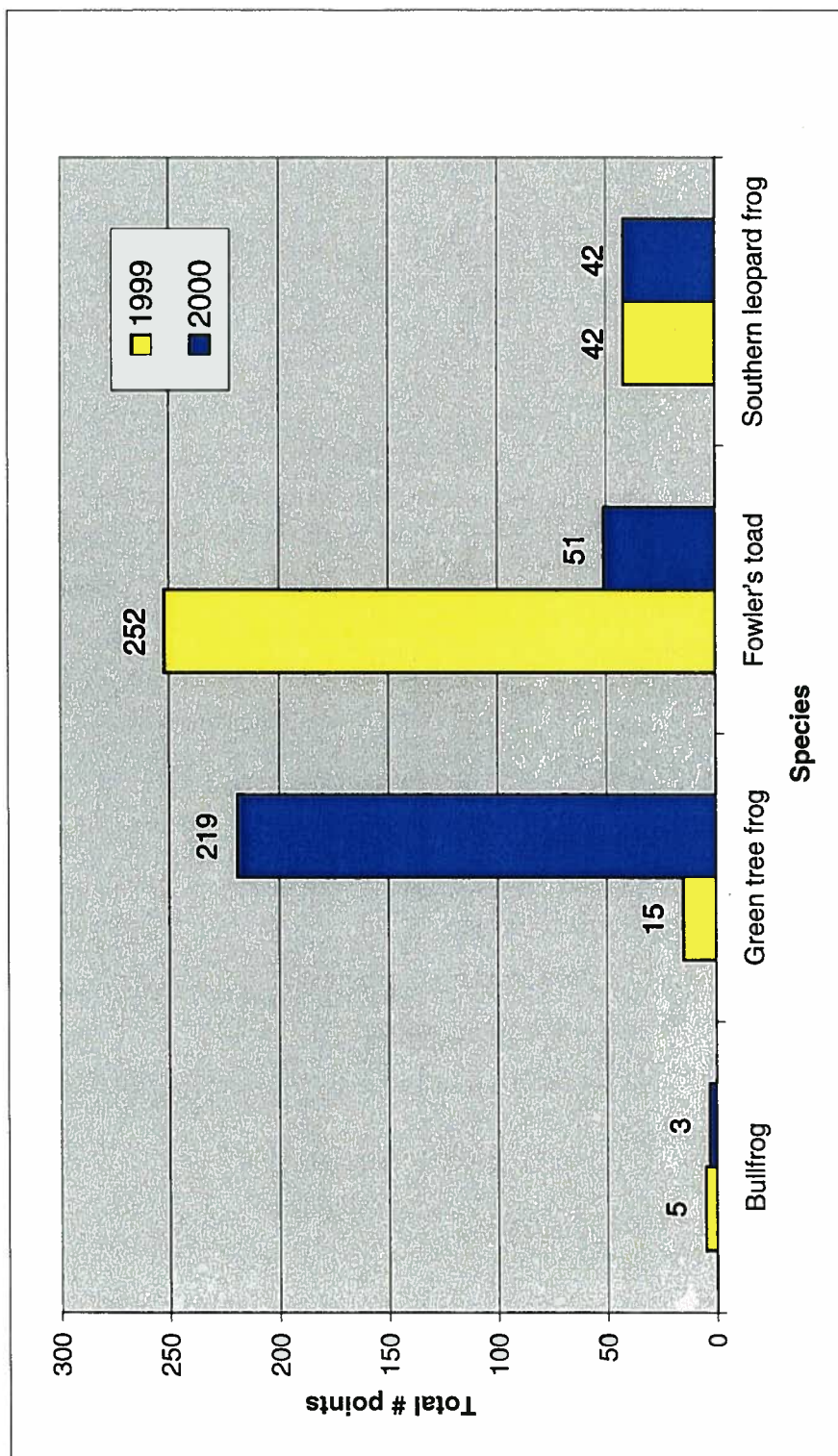


Figure 5: Total number of points each species was detected per season using aural surveys at Chincoteague National Wildlife Refuge 1999-2000.

Basking Survey

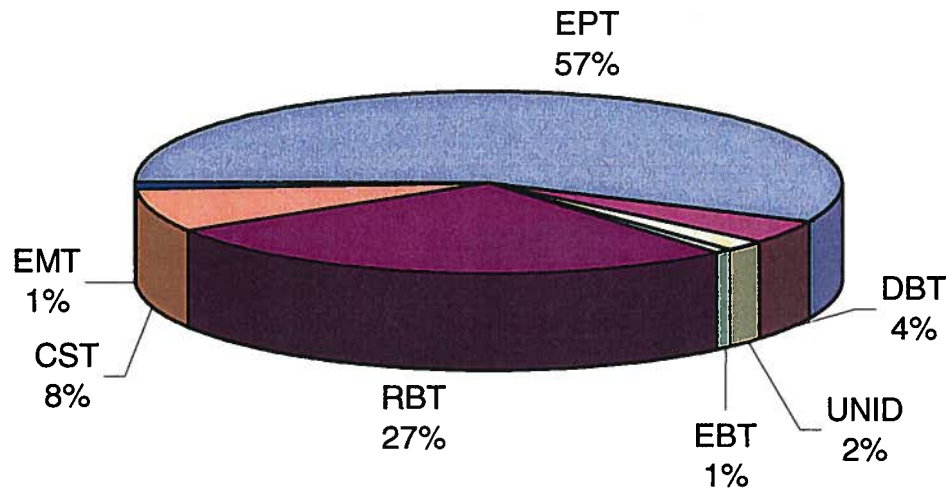


Figure 6: Relative abundance of the species observed using basking surveys at Chincoteague National Wildlife Refuge 1999-2000 (EMT= eastern mud turtle; *Kinosternon subrubrum subrubrum*, EBT= eastern box turtle; *Terrapene carolina carolina*, EPT= eastern painted turtle; *Chrysemys picta picta*, DBT= diamondback terrapin; *Malaclemys terrapin terrapin*, RBT= red-bellied turtle; *Pseudemys rubriventris*, CST= common snapping turtle; *Chelydra serpentina serpentina*, and UNID= unidentified).

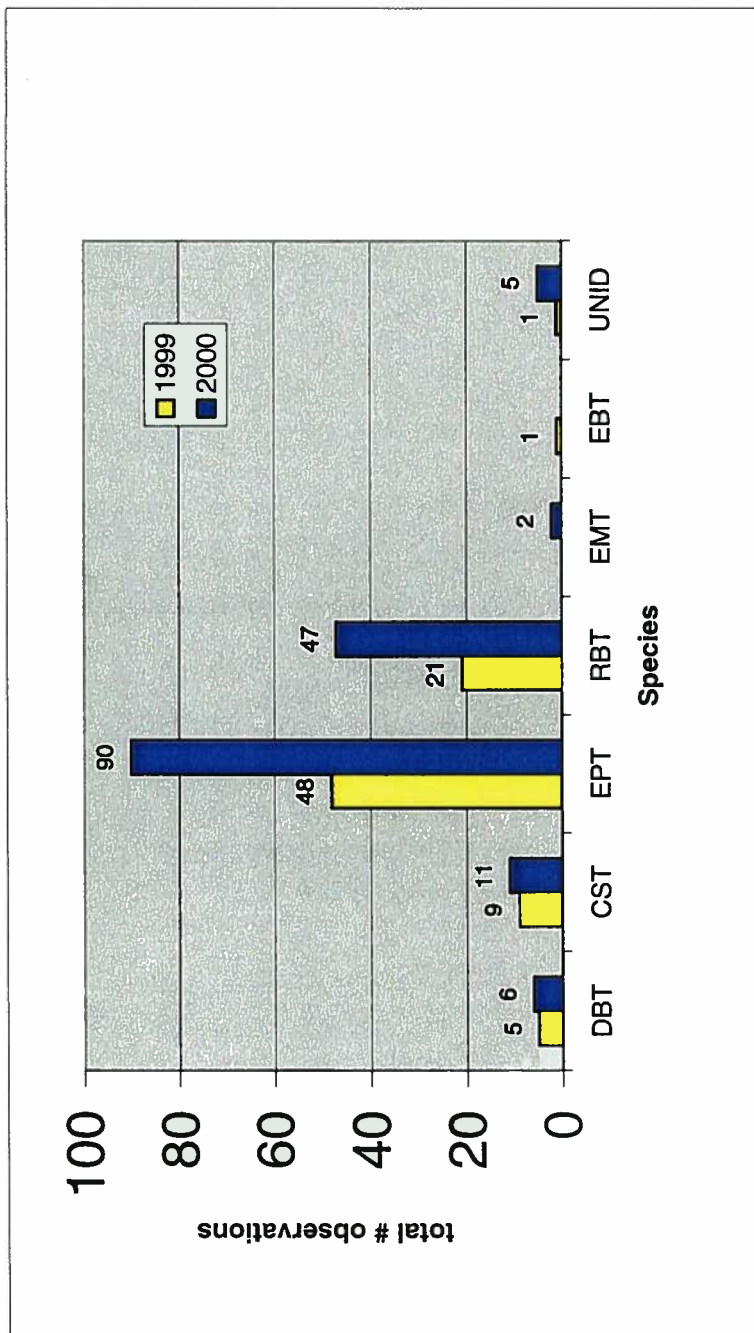


Figure 7: Total number of observations made using basking surveys at Chincoteague National Wildlife Refuge 1999-2000. (DBT= diamondback terrapin; *Malaclemys terrapin terrapin*, CST= common snapping turtle; *Chelydra serpentina serpentina*, EPT= eastern painted turtle; *Chrysemys picta picta*, RBT= red-bellied turtle; *Pseudemys rubriventris*, EMT= eastern mud turtle; *Kinosternon subrubrum subrubrum*, EBT= eastern box turtle; *Terrapene carolina carolina*, and UNID= unidentified.

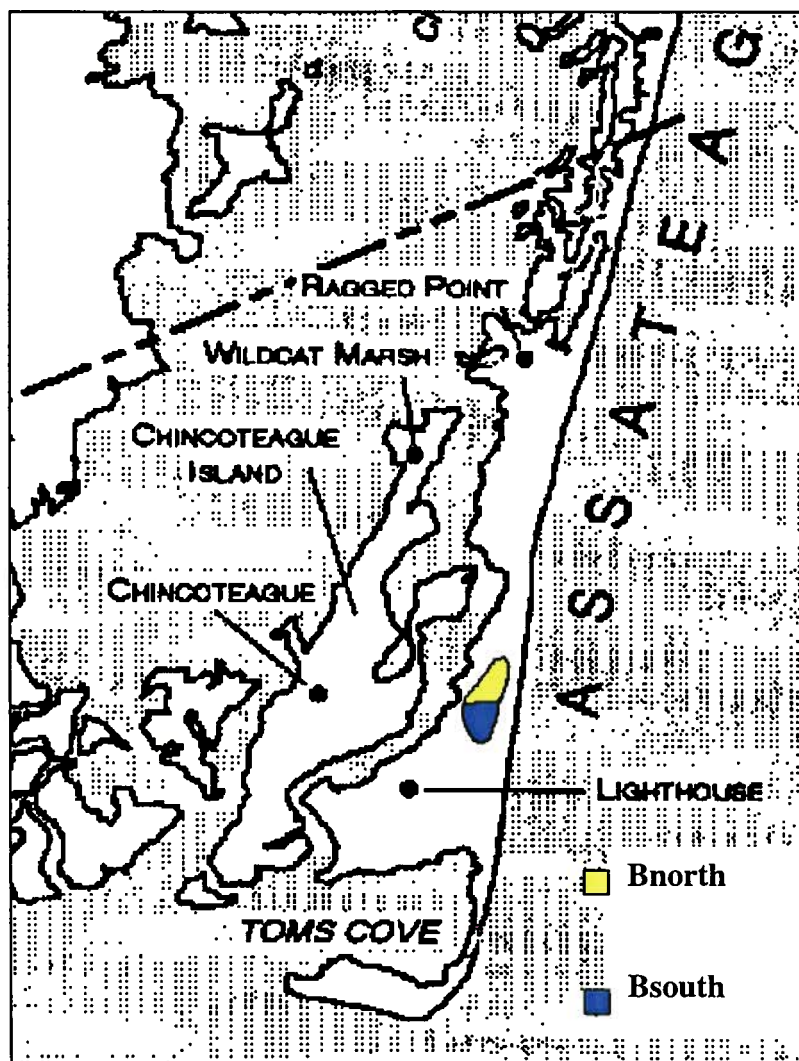


Figure 8: Map of moist soil management units “Bnorth” and “Bsouth” at Chincoteague National Wildlife Refuge 1999-2000

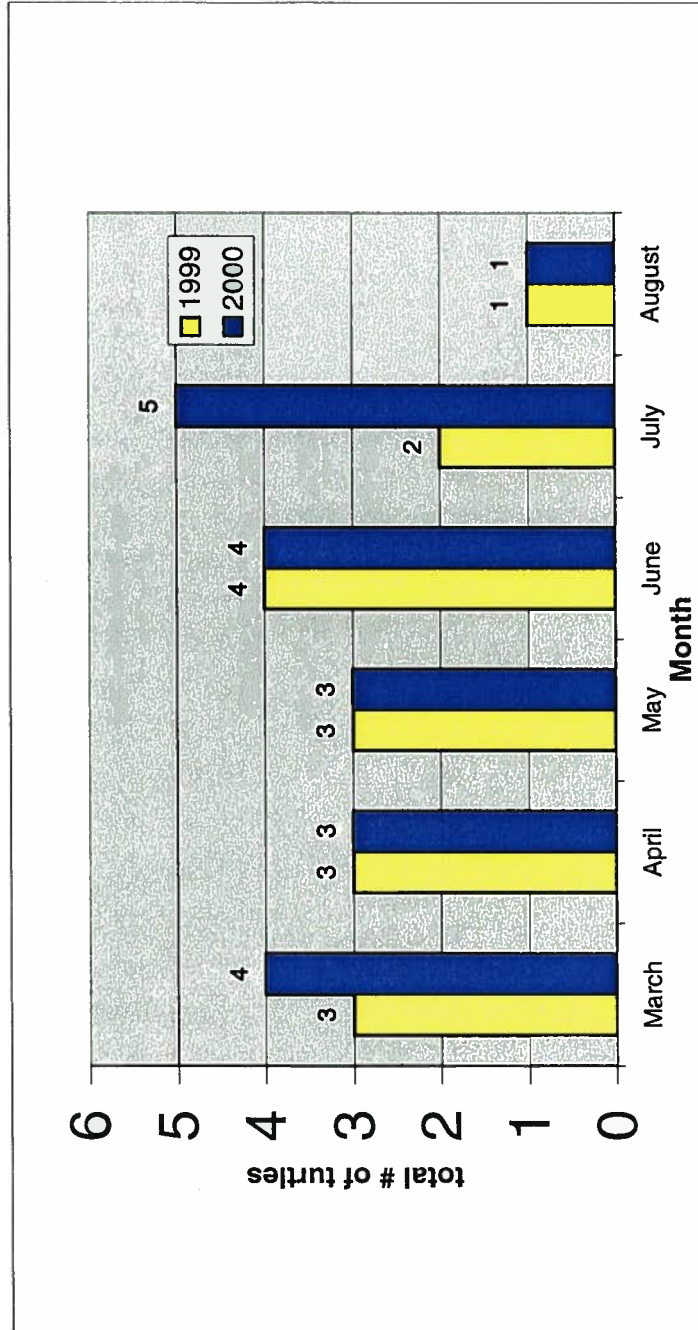


Figure 9: Total number of species detected by month using basking surveys at Chincoteague National Wildlife Refuge 1999-2000.

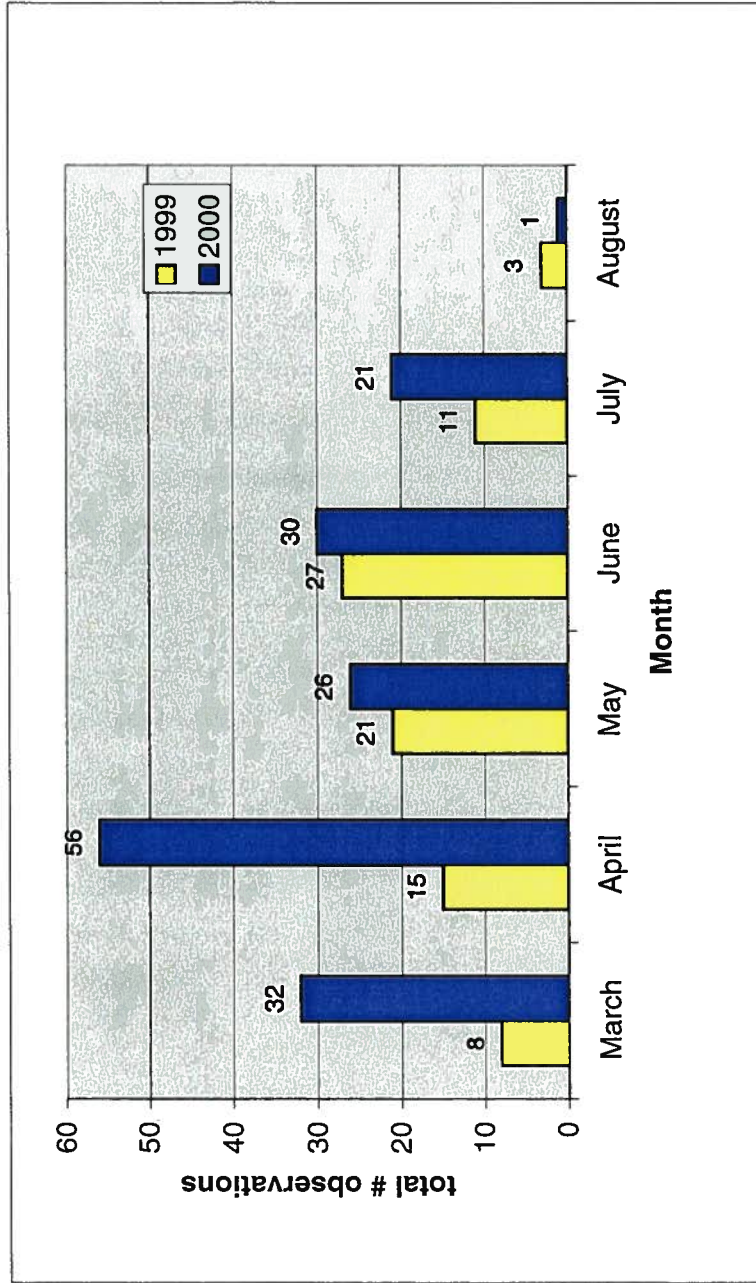


Figure 10: Total number of observations by month using basking surveys at Chincoteague National Wildlife Refuge 1999-2000.

Road Survey

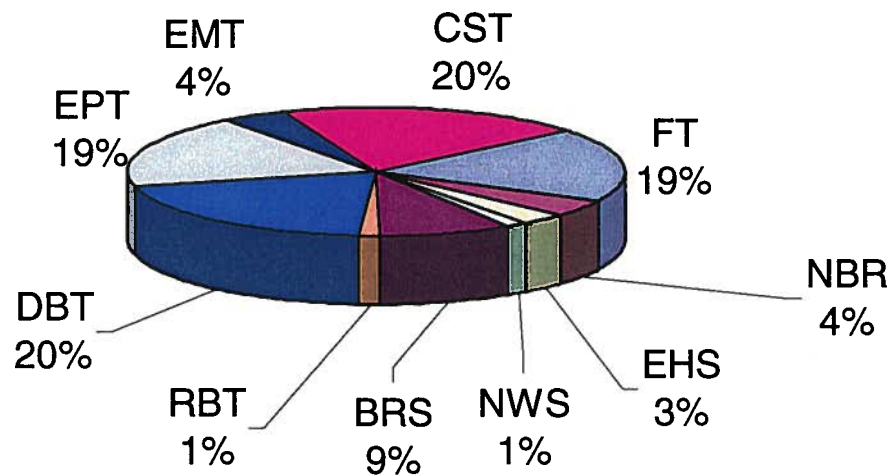


Figure 11: Relative abundance of the species detected using road surveys at Chincoteague National Wildlife Refuge 1999-2000 (FT= Fowler's toad; *Bufo woodhousii fowleri*, NBR= northern black racer; *Coluber constrictor constrictor*, EHS= eastern hognose snake; *Heterodon platirhinos*, NWS=northern water snake; *Nerodia sipedon sipedon*, BRS=black rat snake; *Elaphe obseleta obseleta*, RBT= red-bellied turtle; *Pseudemys rubriventris*, DBT= diamondback terrapin; *Malaclemys terrapin terrapin*, EPT= eastern painted turtle; *Chrysemys picta picta*, EMT= eastern mud turtle; *Kinosternon subrubrum subrubrum*, and CST=common snapping turtle; *Chelydra serpentina serpentina*).

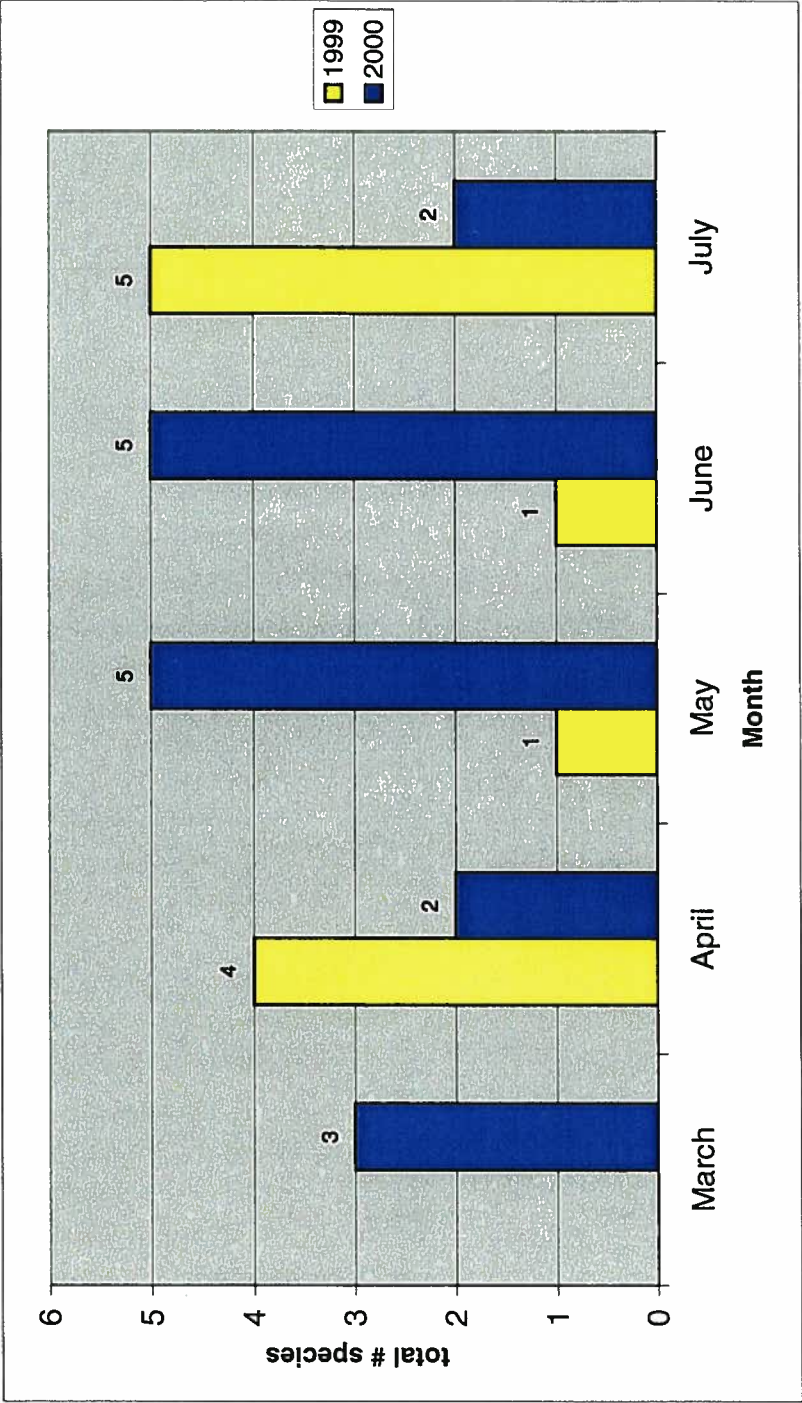


Figure 12: Total number of species detected by month using road surveys at Chincoteague National Wildlife Refuge 1999-2000.

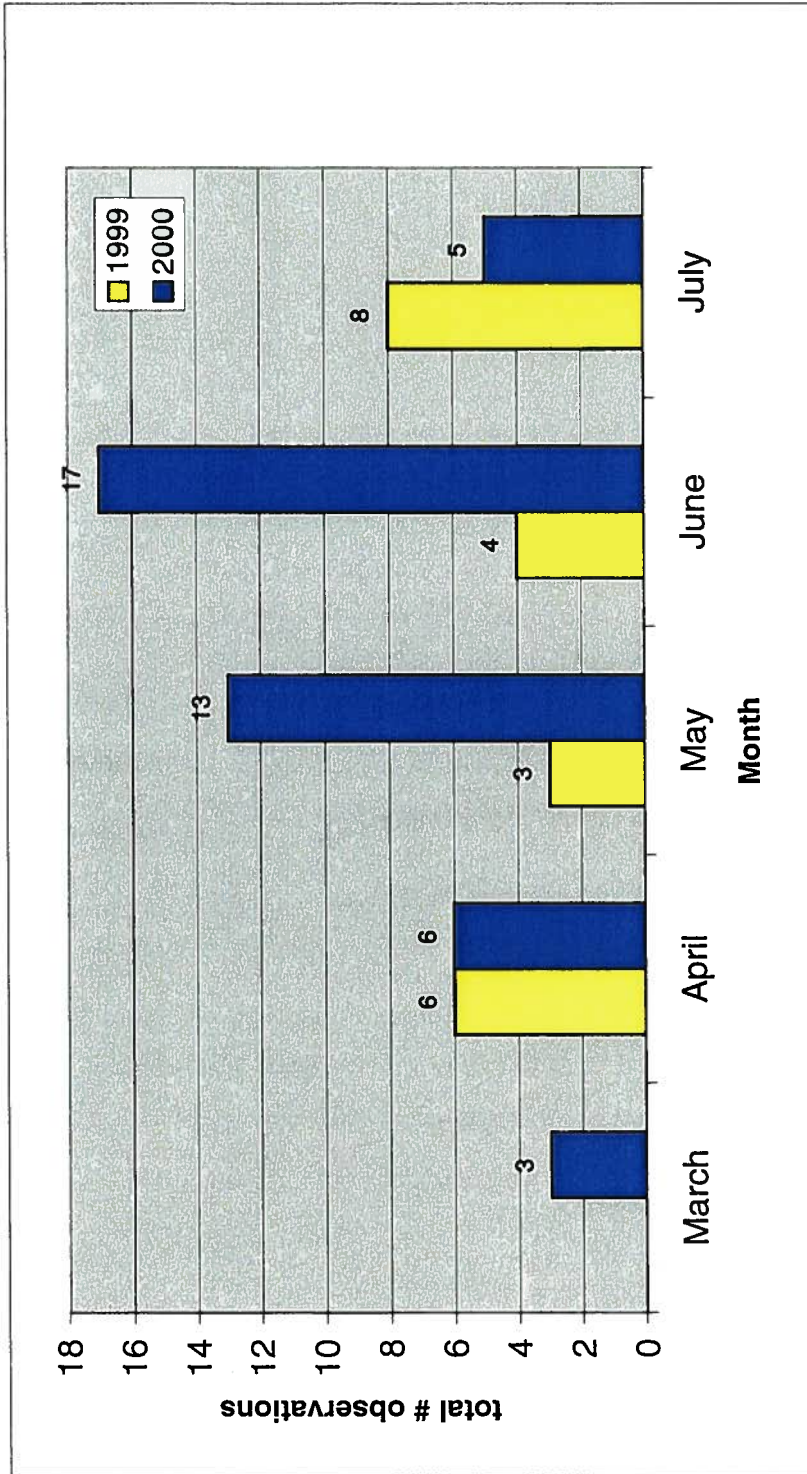


Figure 13: Total number of observations made by month using road surveys at Chincoteague National Wildlife Refuge 1999-2000.

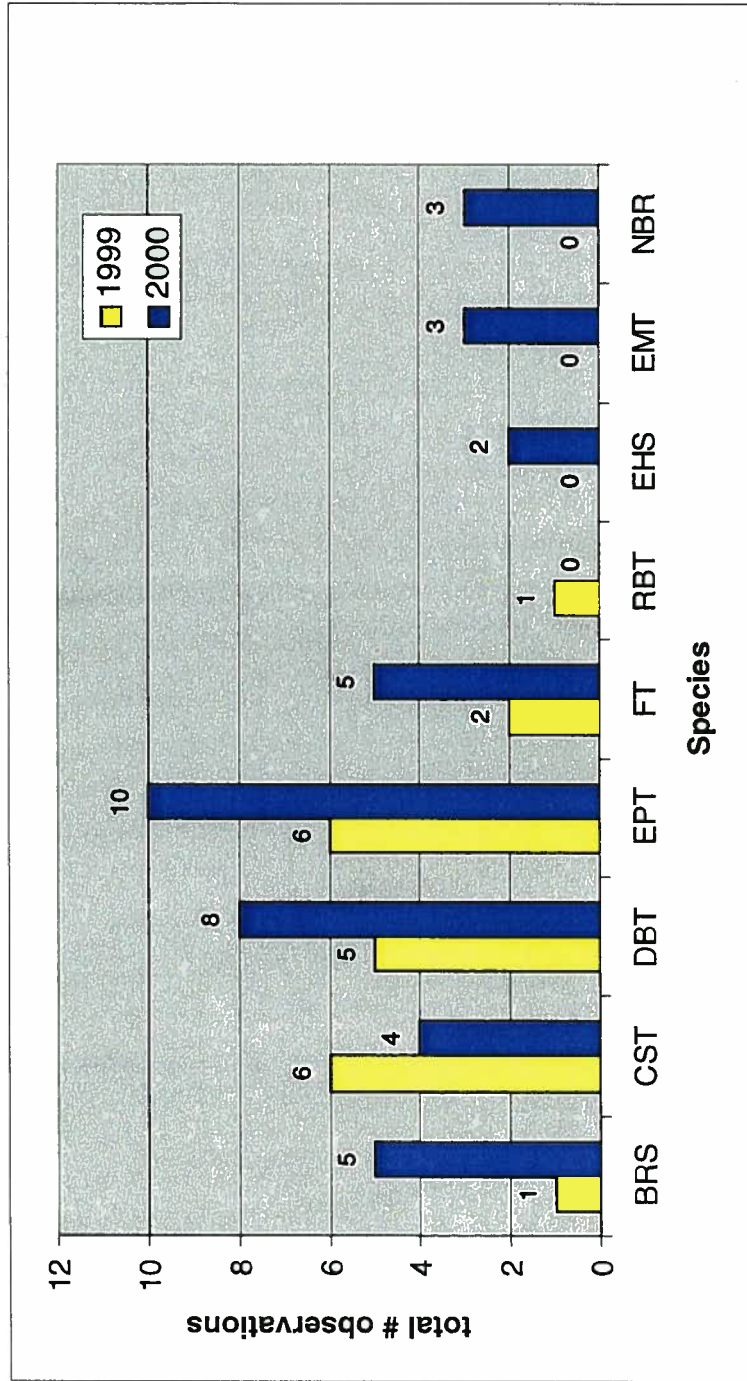


Figure 14: Total number of observations for each species using road surveys at Chincoteague National Wildlife Refuge 1999-2000. (BRS= black rat snake; *Elaphe obseleta obseleta*, CST= common snapping turtle; *Chelydra serpentina serpentina*, DBT= diamondback terrapin; *Malaclemys terrapin terrapin*, EPT= eastern painted turtle; *Chrysemys picta picta*, FT= Fowler's toad; *Bufo woodhousii fowleri*, RBT= red-bellied turtle; *Pseudemys rubriventris*, EHS= eastern hognose snake; *Hererodon platirhinos*, EMT= eastern mud turtle; *Kinosternon subrubrum subrubrum*, NBR= northern black racer; *Coluber constrictor constrictor*.

Time constrained search

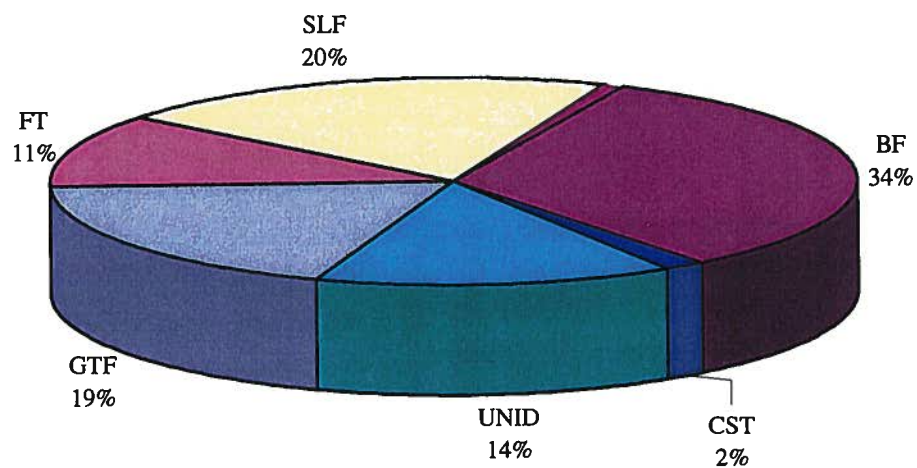


Figure 15: Relative abundance of the species captured using time-constrained searches at Chincoteague National Wildlife Refuge 1999-2000 (GTF= green tree frog; *Hyla cinerea*, FT= Fowler's toad; *Bufo woodhousii fowleri*, SLF= southern leopard frog; *Rana sphenoccephala*, BF= bullfrog; *Rana catesbeiana*, CST= common snapping turtle; *Chelydra serpentina serpentina*, and UNID= unidentified).

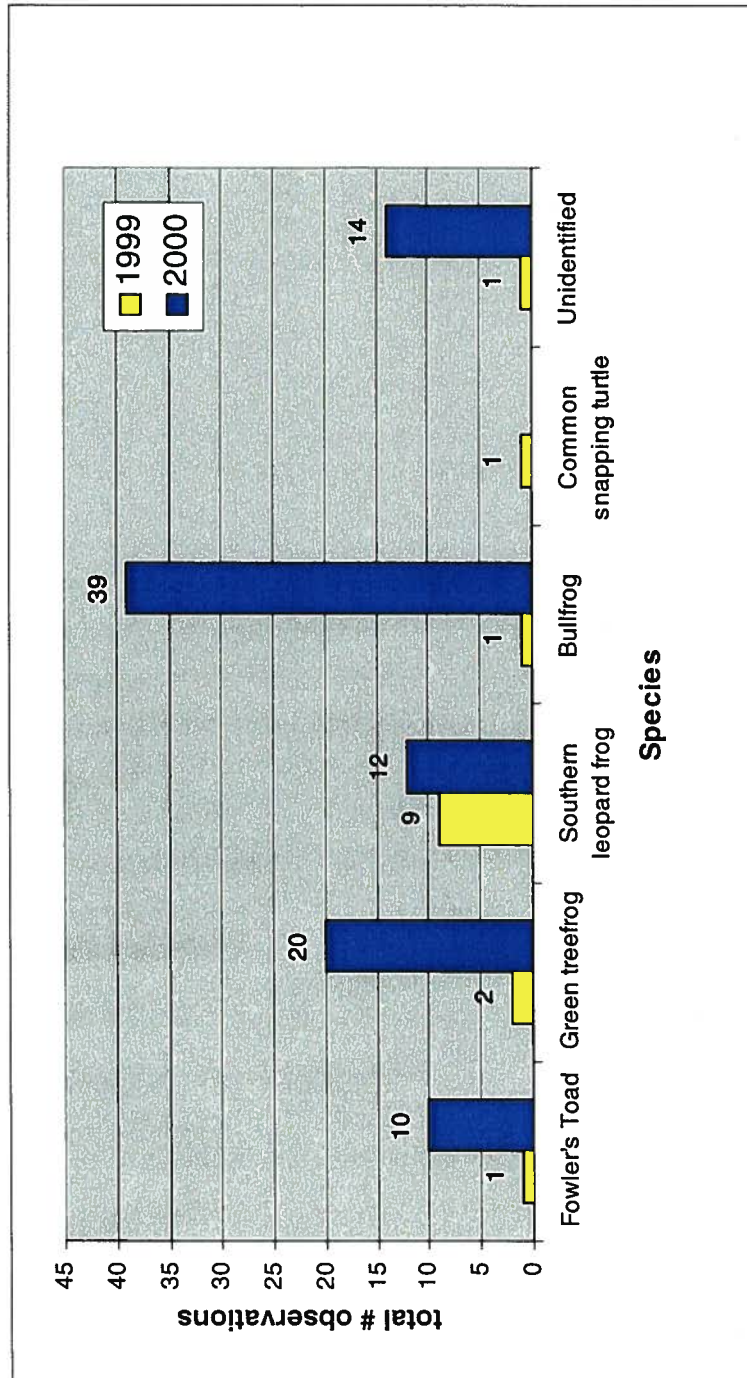


Figure 16: Total number of observations made for each species using time-constrained searches at Chincoteague National Wildlife Refuge 1999-2000.

Pitfall trap arrays

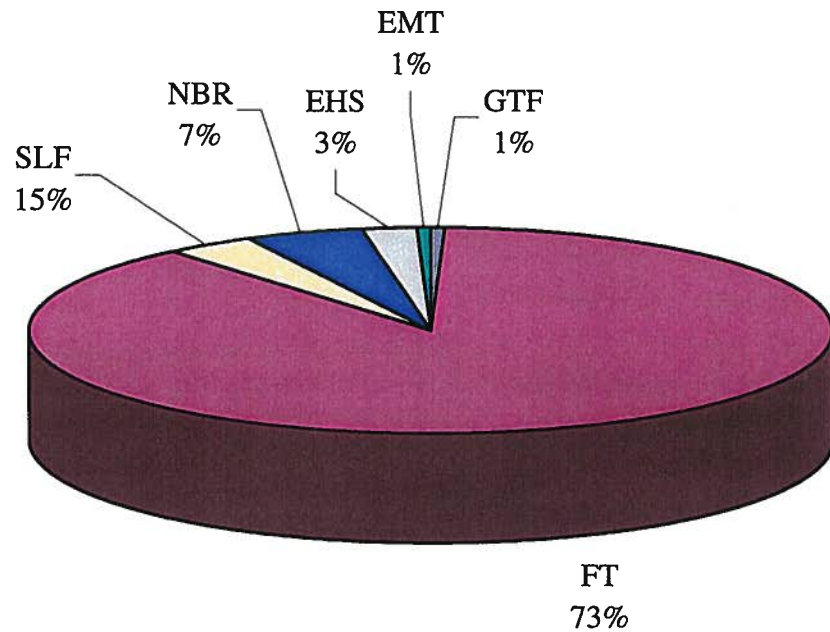


Figure 17: Relative abundance of the species using pitfall trap arrays at Chincoteague National Wildlife Refuge 1999-2000 (GTF= green tree frog; *Hyla cinerea*, FT= Fowler's toad; *Bufo woodhousii fowleri*, SLF= southern leopard frog; *Rana sphenoccephala*, NBR= northern black racer; *Coluber constrictor constrictor*, EHS= eastern hognose snake; *Heterodon platirhinos*, and EMT= eastern mud turtle; *Kinosternon subrubrum subrubrum*).

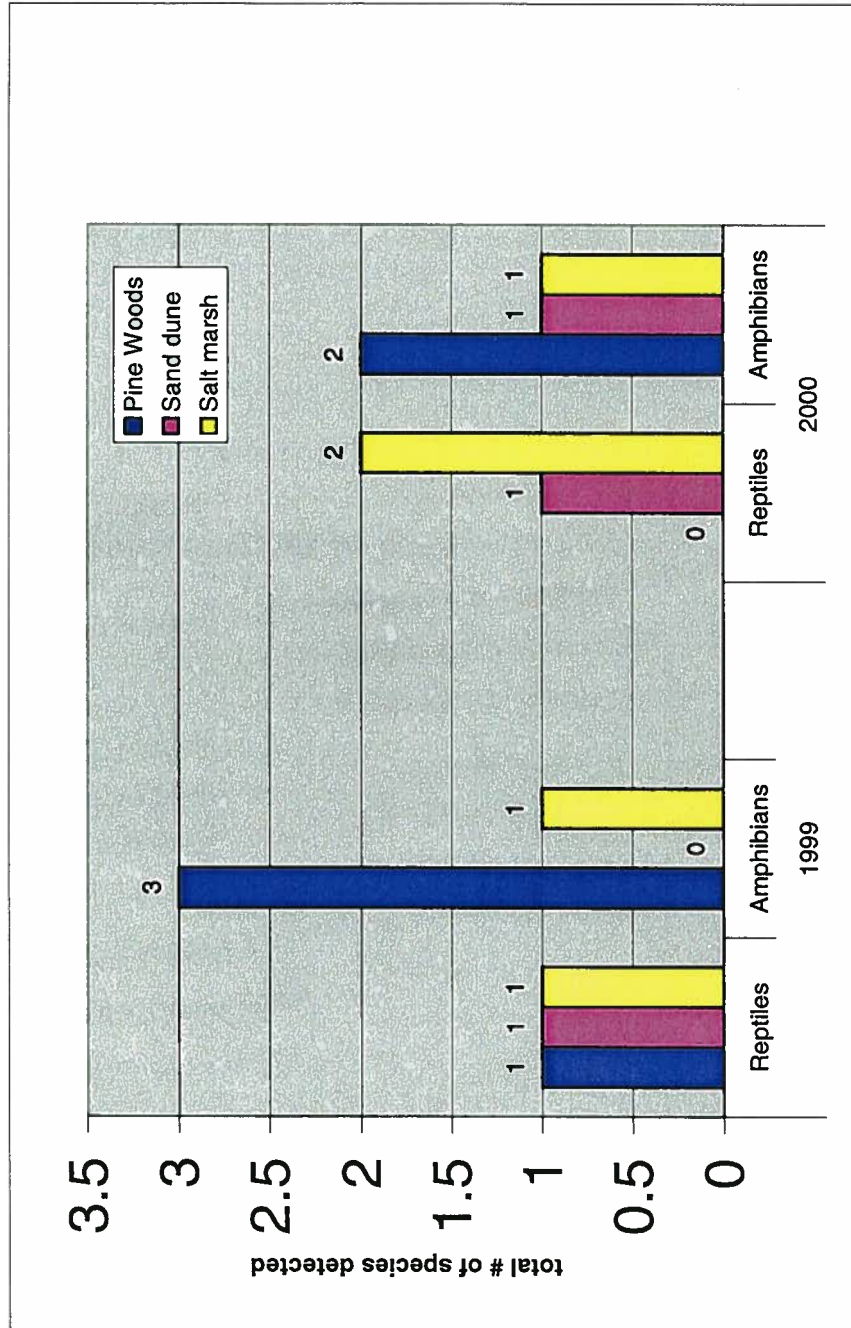


Figure 18: Total number of amphibian and reptile species captured in each habitat type using pitfall trap arrays at Chincoteague National Wildlife Refuge 1999-2000.

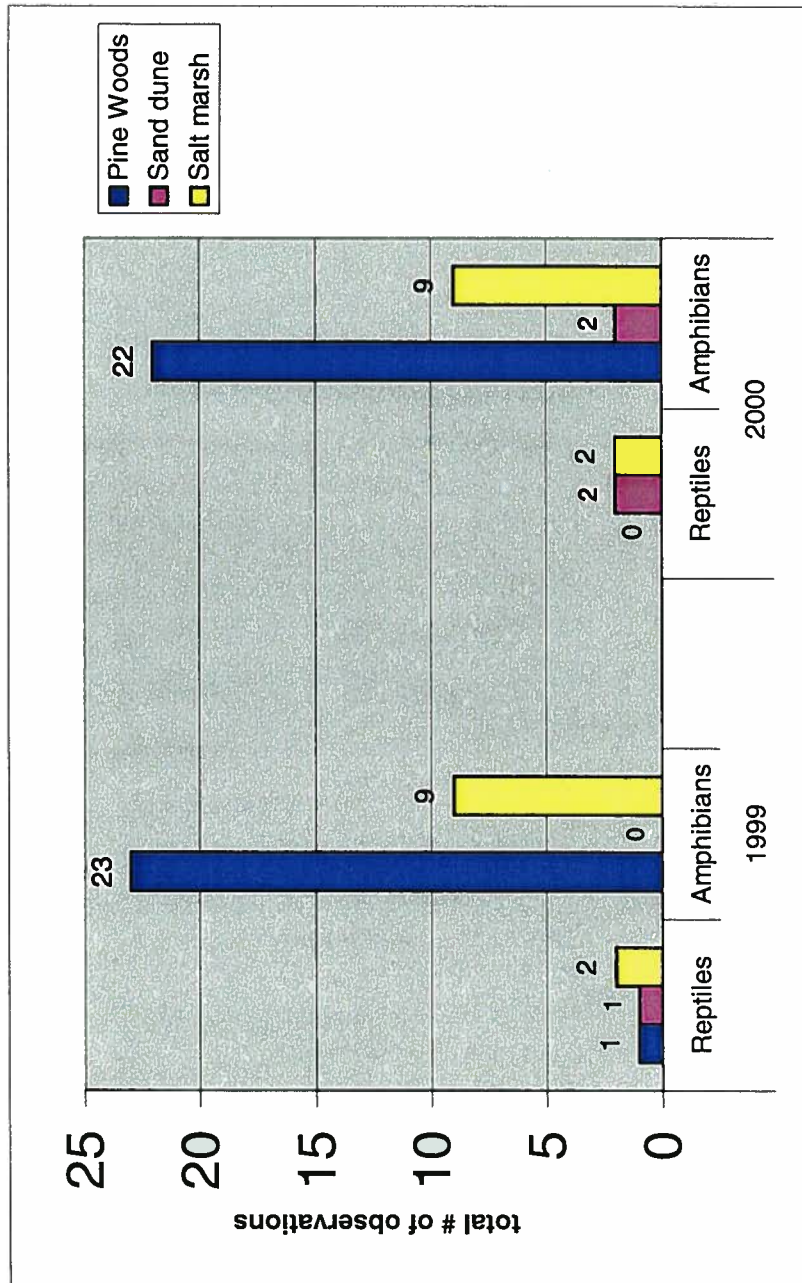


Figure 19: Total number of observations of amphibians and reptiles per habitat for the pitfall array technique at Chincoteague National Wildlife Refuge in 1999 and 2000.

Coverboard survey

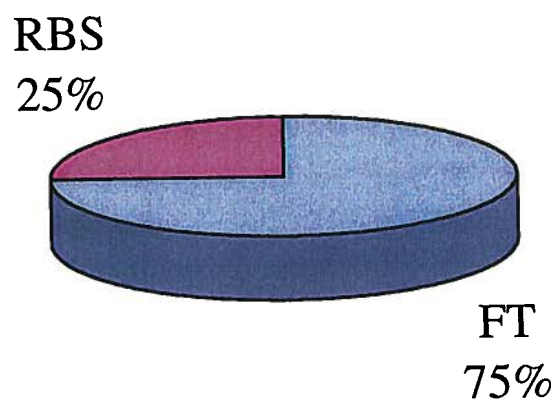


Figure 20: Relative abundance of the species detected using the coverboard survey at Chincoteague National Wildlife Refuge 1998-2000 (FT= Fowler's toad; *Bufo woodhousii*, and RBS= Red-backed salamander; *Plethodon cinereus*).

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