BEFORE THE SECRETARY OF THE INTERIOR

Photo Credit: Karen Powers

PETITION TO LIST THE TRICOLORED BAT *PERIMYOTIS SUBFLAVUS* AS THREATENED OR ENDANGERED UNDER THE ENDANGERED SPECIES ACT

June 14, 2016

CENTER FOR BIOLOGICAL DIVERSITY
&
DEFENDERS OF WILDLIFE

NOTICE OF PETITION

June 14, 2016

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Dear Secretary Jewell:

Pursuant to Section 4(b) of the Endangered Species Act ("ESA"), 16 U.S.C. §1533(b), Section 553(3) of the Administrative Procedures Act, 5 U.S.C. § 553(e), and 50 C.F.R. §424.14(a), the Center for Biological Diversity and Defenders of Wildlife hereby formally petition the Secretary of the Interior to list the tricolored bat, *Perimyotis subflavus*, as a Threatened or Endangered species and to designate critical habitat concurrent with listing.

This petition sets in motion a specific process, placing definite response requirements on the Secretary and the U.S. Fish and Wildlife Service (USFWS) by delegation. Specifically, USFWS must issue an initial finding as to whether the petition "presents substantial scientific or commercial information indicating that the petitioned action may be warranted." 16 U.S.C. §1533(b)(3)(A). USFWS must make this initial finding "[t]o the maximum extent practicable, within 90 days after receiving the petition." Id. Petitioners need not demonstrate that listing is warranted, rather, petitioners must only present information demonstrating that such listing may be warranted. While petitioners believe that the best available science demonstrates that listing the tricolored bat as endangered is in fact warranted, there can be no reasonable dispute that the available information indicates that listing this species as either threatened or endangered may be warranted. As such, USFWS must promptly make an initial finding on the petition and commence a status review as required by 16 U.S.C. § 1533(b)(3)(B).

PETITIONERS:

The Center for Biological Diversity ("Center") is a non-profit, public interest environmental organization dedicated to the protection of native species and their habitats through science, policy, and environmental law. The Center is supported by more than one million members and activists throughout the United States. The Center and its members are concerned with the conservation of endangered species, including the tricolored bat. The Center's members and staff have biological, health, educational, scientific research, moral, spiritual and aesthetic interests in the tricolored bat and its habitat.

Defenders of Wildlife is a non-profit conservation organization that advocates for wildlife and its habitat. Defenders uses education, litigation, and research to protect wild animals and plants in their natural communities. Known for its effective leadership on endangered species issues, Defenders also advocates new approaches to wildlife conservation that protect species before they become endangered. Its programs reflect the conviction that saving the diversity of our planet's life requires protecting entire ecosystems and ensuring interconnected habitats. Founded in 1947, Defenders of Wildlife is a 501(c)(3) membership organization with more than 1,000,000 members and supporters nationwide.

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I. EXECUTIVE SUMMARY

The tricolored bat (*Perimyotis subflavus*) is one of the smallest bats in North America. The species has been devastated by the novel, invasive fungal disease known as white-nose syndrome (WNS) since 2006, when the disease was first documented at hibernacula in upstate New York. As WNS has spread across the continent, numbers of the tricolored bat have plummeted. Biologists report mortality rates up to 100 percent in affected hibernacula. As a result, several eastern states as well as the Canadian government have recently protected or are in the process of protecting the tricolored bat under endangered species laws.

The tricolored bat hibernates for longer periods than other bat species within its range. This behavior, along with the species' preference for hibernating sites within the optimal temperature range for the WNS fungus, may make the tricolored bat particularly susceptible to lethal infection by the disease.

This species appears to have been in gradual decline even prior to the catastrophic advent of WNS. Additional threats posed to the tricolored bat are human disturbance at hibernation and roost sites, wind energy, habitat loss, pesticides, and climate change. The species' low reproductive rate constrains its ability to recover quickly from the dramatic population decline associated with WNS.

The Endangered Species Act states that a species shall be determined to be endangered or threatened based on any one of five factors (16 U.S.C. § 1533 (a) (1)). The tricolored bat is threatened by four of these five factors and demonstrably warrants listing as threatened or endangered.

The loss and curtailment of habitat or range. Habitat essential to the tricolored bat is threatened by residential, agricultural, and industrial development and related activities and by various resource extractive industries.

Disease. The tricolored bat is susceptible to WNS, a novel epizootic that has killed nearly seven million bats in eastern North America since it was discovered in 2006. By many accounts, the tricolored bat is among the species most affected by this disease. The tricolored bat is experiencing declines of up to 100 percent in affected hibernacula, and has been nearly extirpated from several states since the advent of WNS. The possibility that additional bat species warrant listing as a result of WNS-related declines has already been acknowledged by USFWS, with the tricolored bat being recognized as a species that needs to be considered for protection.

Numerous other natural and anthropogenic factors. A multitude of other factors make the tricolored bat highly vulnerable to the more immediate threats outlined above. Among non-migratory bats, the tricolored bat is the species most frequently found dead at industrial wind energy installations in the eastern United States. Environmental contaminants associated with agriculture, mining, and industries have been documented to have both lethal and sub-lethal effects on bat populations, primarily by reducing reproductive success. Climate change is expected to cause significant changes in insect distribution, abundance, and phenology; as an exclusive insectivore, the tricolored bat will be greatly affected by these changes in prey species. Alterations in annual temperature and precipitation regimes may also disrupt the physiological mechanisms governing hibernation and reproduction, and render hibernacula in certain parts of the species' range unsuitable.

The inadequacy of existing regulatory mechanisms. While the species is now listed as endangered, threatened, or of special conservation concern in a few states across its range, these designations afford no significant regulatory protection. National Forest management plans do not protect species that are not federally listed, and sympatry with listed species does not adequately protect the tricolored bat from logging or other threats on these lands. The regulation of oil, gas, mineral, and renewable energy development on federal, state, and private lands is inconsistent and inadequate, and further development of coal and gas reserves in significant bat habitat is probable. Cave-dwelling bats are not sufficiently protected from disturbance in hibernacula by either federal or state cave protection acts. Finally, coherent and successful management of WNS will require more funding, more staff, and better coordination among participating agencies.

Based on the factors outlined above, the tricolored bat warrants protection under the ESA.

II. INTRODUCTION

Bats are of great ecological and economic importance. Insectivorous bats, such as the tricolored bat, provide significant natural insect control, quantifiably benefiting both agriculture and public health. An individual bat may consume 500-1000 insects in an hour, and may consume up to 100 percent of its body mass in insects nightly (Griffin et al. 1960, Kurta et al. 1989). One million bats remove roughly 694 tons of insect prey from the environment each summer (Kunz 2009). Since 2006 the U.S. Fish and Wildlife Service estimates that 5.7-6.7 million bats have died from the bat epidemic caused by white-nose syndrome (USFWS 2012). The primary native predators of night-flying insects in the eastern United States have thus not been available to remove the equivalent of nearly 4,000 tons of insects each year for the last several years.

The long-term consequences of this loss of predation pressure are unknown. However, with regard to human-dominated landscapes, scientists conservatively estimate the value of pest-eating bats to American agriculture as ranging between $3.7 billion and $53 billion annually (Boyles et al. 2010). Another study of the value of bats to agriculture found that in eight counties of south-central Texas, pest control provided by Mexican free-tailed bats (*Tadarida brasiliensis*) to the regional cotton industry was worth $741,000 annually (Cleveland et al. 2006). It is likely that the decline of the tricolored bat, which favors open landscapes more than most other insectivorous bats in eastern North America, has already had real, if as-yet unmeasured, ripple effects on the balance of insect pests in farming areas.

Since the appearance of WNS, the tricolored bat has been declining dramatically. The species is also imperiled by a host of other threats, most of them anthropogenic: Habitat loss and degradation driven by agricultural and residential development, logging, mining and other resource extractive practices, industrial wind energy, environmental contaminants, disturbance by vandalism and recreation, and climate change.

WNS is a fungal disease caused by a newly described, non-native fungal species named *Pseudogymnoascus destructans*. The initial discovery of WNS in New York state hibernacula precipitated a cascade of concern about the future of all hibernating bat species in the United States. Since WNS was first observed in 2006, it is estimated to have killed nearly seven million hibernating bats in the East, and is rapidly spreading south and west (USFWS 2012). The disease is now reported in 29 states and five Canadian provinces (USFWS 2014). Much of the tricolored bat's range is now affected by WNS. High mortality of the species is reported from WNS-affected hibernacula; several aspects of the species' life history may make it especially vulnerable to the disease. A number of state fish and wildlife departments have added the tricolored bat to their state endangered and threatened lists in recent years, because of the population losses caused by WNS.

This petition summarizes the natural history and population status of the tricolored bat and outlines the recognized threats to the species and its habitats. The petition then clearly demonstrates that, in the context of the ESA's five statutory listing factors, the U.S. Fish and Wildlife Service should protect the tricolored bat as Threatened or Endangered.

III. NATURAL HISTORY AND ECOLOGY

A. Taxonomy

The tricolored bat is of the order Chiroptera and family Vespertilionidae (Table 1). The genus and species is *Perimyotis subflavus* (Quinn and Broders 2007). The tricolored bat was previously named the eastern pipistrelle, *Pipistrellus subflavus* (Fujita and Kunz 1984). Many researchers still refer to the species as the pipistrelle or "pips" (Harvey et al. 2011). However, Menu (1984) placed the species in its own genus; later, genetic research confirmed that the species is not closely related to the Old World Pipistrellus bats (Hoofer and Van Den Bussche 2003, Hoofer et al. 2006). The bat historically known as the western pipistrelle, *Pipistrellus hesperus*, now occupies its own genus, *Parastrellus*, as well. There are no species other than the tricolored bat that are members of the genus *Perimyotis* in North America.

Table 1: Classification of Tricolored Bat (NatureServe 2014).

Kingdom	Phylum	Class	Order	Family	Genus	Species
Animalia	Craniata	Mammalia	Chiroptera	Vespertilionidae	*Perimyotis*	*subflavus*

There are four recognized subspecies of tricolored bat in North America—*P. s. subflavus*, the nominal subspecies, *P. s. floridanus*, *P. s. clarus*, and *P. s. veraecrucis*, the latter two of which are found in Mexico (American Society of Mammalogists 1984, Hoofer et al. 2006). There is a possibility that the tricolored bat in Nova Scotia is disjunct (Broders et al. 2003, Quinn and Broders 2007) and warrants subspecies status because of morphometric differences (H. Broders and H. Hunyh, unpub. data), but the taxonomy has not been revised to date.

B. Description

Weighing about 4 to 8 grams, and measuring 77-89 mm from head to tail, the tricolored bat is relatively small compared to other North American bats (Fujita and Kunz 1984). It is the smallest bat found throughout the eastern and midwestern states (Amelon 2006). The wingspan is 220-225 mm (Quinn and Broders 2007). Females are larger than males, and have significantly larger fat deposits than males (Fujita and Kunz 1984). The species' pelage is yellowish-brown, while each hair, when examined individually, is "tricolored": brown at the tip of the hair, yellow in the middle, and dark at the base (Fujita and Kunz 1984). The dorsal pelage is a yellowish brown, and the forearm skin is reddish. The tricolored bat has oblong ears, which when held toward the nose reach just beyond the tip of the nostrils (Quinn and Broders 2007). The tragus, or fleshy projection in the ear, is short and blunt (Long 2008). The lower third of the tail membrane is lightly furred (Chapman and Williams 2010). The tricolored bat is a slow flier, with an erratic and "fluttery" pattern while foraging (Fujita and Kunz 1984); its flight pattern and small size lead people to sometimes mistake it for a moth (Harvey et al. 2011). The tricolored bat is among the earliest bat species to feed in the evening (Fujita and Kunz 1984).

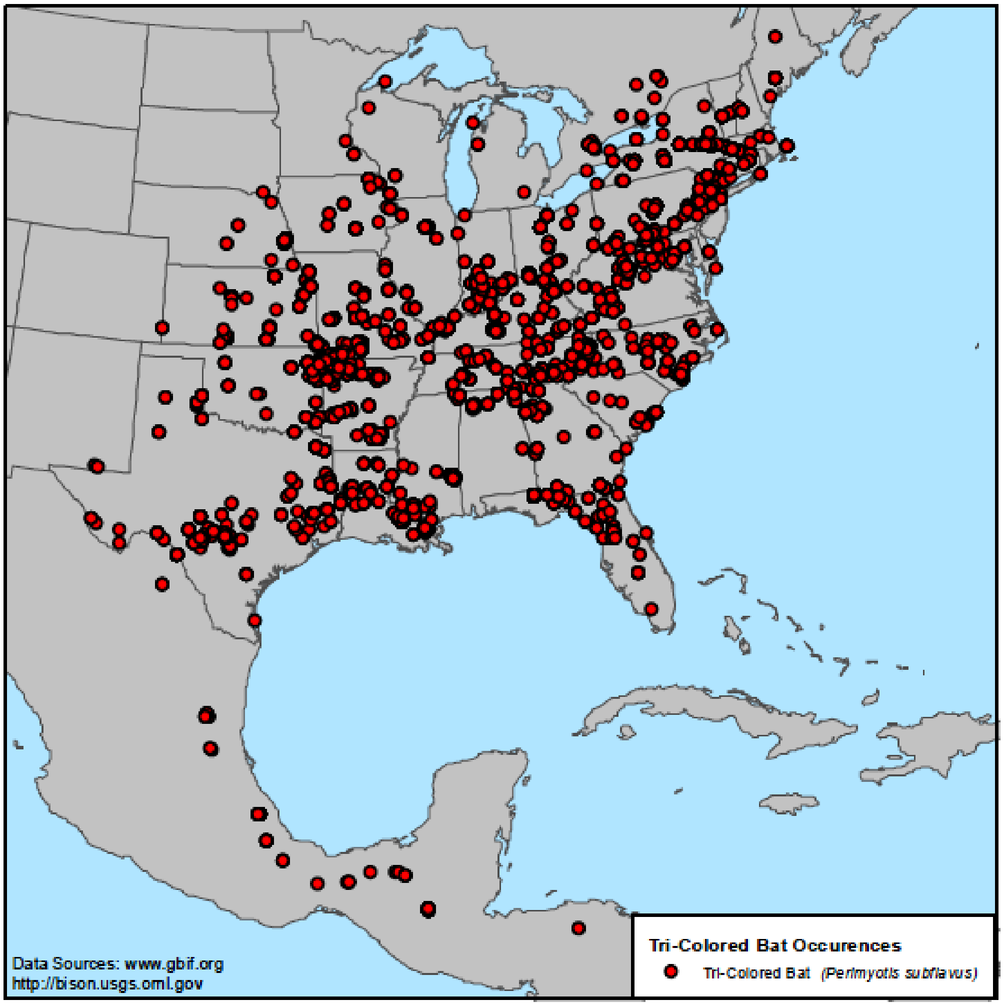
C. Distribution

The tricolored bat ranges across most of eastern North America and into eastern Central America, and it occurs over much of the midwestern United States (Fujita and Kunz 1984). The species' broad range is not well defined, but it has been documented as far north as southwest Nova Scotia, where a small, possibly disjunct population occurs (Quinn and Broders 2007), and south to northeastern Honduras (Fujita and Kunz 1984, Arroyo-Cabrales et al. 2008). Westwards, the bat has been documented in Ontario, Nebraska, Kansas and on the Edwards Plateau of central Texas (Fujita and Kunz 1984, NatureServe 2014). Earlier accounts about the species stated that it was absent from northern New England and southern Florida, as well as northern Indiana, northeast Illinois, eastern Wisconsin, and most of Michigan (Barbour and Davis 1969). However, Kurta (1982) reported minimal records from southeast Michigan and DeGraaf and Yamasaki (2001) reported the tricolored bat throughout Vermont and New Hampshire, though absent in northern Maine. Trombulak et al. (2001) found that low numbers of tricolored bats had been consistently documented in Vermont, pre-WNS, since the 1930s. Kurta et al. (2007) reported range expansion in the Great Lakes region since the 1980s. The tricolored bat appears to be expanding its range in the West as well-- scientists have found it in recent years in west Texas, New Mexico, Colorado, and South Dakota (Geluso et al. 2005, Ammerman 2005, Armstrong et al. 2006, White et al. 2006, Valdez et al. 2009). The species is found in the Florida Panhandle (Davis 1959, Amelon 2006).

As with other cave bats, the availability of suitable hibernacula for tricolored bat apparently influences, and limits, the species' geographic range (Vincent and Whitaker 2007). The species has been considered a relatively short-distance migrant, with summer roosting areas generally within 100 km of hibernacula, and the longest recorded migratory distance for the species at 85 km (Griffin 1940), though new research indicates probable long-distance migration among some tricolored bats.

Prior to the onset of WNS, biologists considered the tricolored bat fairly common in parts of its range, but relatively rare in other areas. The advent of WNS has precipitated a dramatic drop in tricolored bat populations throughout much of its range. Currently, NatureServe (2014) ranks the tricolored bat as: critically imperiled in Pennsylvania, Nebraska, Nova Scotia, and Quebec; imperiled in Michigan and New Brunswick; and vulnerable in Alabama, Massachusetts, Minnesota, New Mexico, New York, North Carolina, Virginia and Ontario. In Vermont, the tricolored bat's status ranges from vulnerable to imperiled, and in Wisconsin, between vulnerable to critically imperiled.

The range of the tricolored bat in Canada is quite limited. It has been recorded in southern parts of Nova Scotia, New Brunswick, Quebec, and central Ontario, southward (van Zyll de Jong 1985; Figure 4). Broders et al. (2003) suggested that the population in southeast Nova Scotia is isolated. Breeding is strongly suspected in Nova Scotia (Broders et al. 2003) but uncertain for New Brunswick (Broders et al. 2001).



D. Habitat

General
Tricolored bats appear to inhabit landscapes that are partly open, with large trees and plentiful woodland edges (Amelon 2006). They are found in a variety of terrestrial habitats, including grasslands, old fields, suburban areas, orchards, urban areas, and woodlands, especially hardwood woodlands. They generally avoid deep woods as well as large, open fields (NatureServe 2014). They require specific habitats for hibernating, roosting, and foraging.

Hibernacula
The tricolored bat is considered to be highly sensitive to overwintering habitat conditions. While it often hibernates in the same sites as other bat species, such as the little brown bat (*Myotis lucifugus*) and the northern long-eared bat (*Myotis septentrionalis*), the tricolored bat tends to occupy the deepest part of caves where temperature is highest and least variable, the walls of the cave are warmer, and humidity levels are higher (Fujita and Kunz 1984, Raesly and Gates 1987, Briggler and Prather 2003).

Biologists report that the tricolored bat uses caves, mines, and buildings for hibernation, but the degree to which the species utilizes different types of hibernacula, and how that may vary with factors such as latitude, is yet unclear.

Quinn and Broders (2007) state that in the northern parts of its range, the tricolored bat hibernates almost exclusively in caves and mines. However, other investigators report that the species also hibernates in buildings (Sandel et al. 2001). In more southerly parts of its range, where year-round food supplies exist, the tricolored bat enters caves or buildings to go into a period of torpor (Briggler and Prather 2003, Quinn and Broders 2007).

Investigators have found that the species prefers larger hibernacula with east-facing openings and high thermal stability (Briggler and Prather 2003). However, in Iowa, tricolored bats appear to prefer caves with vertical entrances. It is unclear whether this is a preference for a particular entrance morphology, or due to a correlation of sinkhole entrances with larger caves (Dixon 2011), which again would indicate the species' preference for greater thermal stability at hibernacula.

The tricolored bat usually hangs singly, rather than in small groups or clusters, and it may occupy the same precise spot from one winter to the next. However, individuals may also have several spots among which they move over the course of a winter (Harvey 2003).

Tricolored bats go deeper into caves than other bat species, again presumably because they prefer more stable as well as warmer temperatures while in hibernation (Vincent and Whitaker 2007). It is thought the warmer pockets in hibernacula, along with more stable temperatures, allow the bat to remain in torpor for longer periods of time (Vincent and Whitaker 2007). However, in Vermont, biologists frequently encounter tricolored bats near the entrances to hibernacula, as well as in deeper locations (VESC 2012). Temperatures in hibernacula used by the tricolored bat have ranged from -6 to 14° C (Barbour and Davis 1969, McNab 1974, Sandel et al. 2001, Amelon 2006). This temperature range is among the greatest recorded for eastern forest hibernating bat species (Rabinowitz 1981).

In terms of humidity, at least one study found that females may prefer sites with higher humidity than sites chosen by males. In Georgia, hibernating females used sites with slightly higher average humidity than males (64.2% versus 61.5% humidity) (Menzel et al. 1999).

Tricolored bats use caves of many sizes. In Iowa, tricolored bats were the most common species in a survey of small caves (less than or equal to 50 m in length); 68 percent of such small caves harbored them, with an average of 1.2 tricolored bats per cave (Dixon 2011). Overall, however, tricolored bats in Iowa appear to favor larger caves over smaller ones.

In terms of location, generally bat biologists have believed that tricolored bats' hibernacula occur within approximately 100 km of summer roosting areas (Griffin 1940). Yet, recent research provides evidence that some tricolored bats, particularly males and those summering at more northern latitudes, migrate on a larger geographic scale more similar to that associated with the non-hibernating tree bats (Fraser et al. 2012).

Summer Roosting Habitat

The tricolored bat appears to roost in a wide variety of places across its range. Some observers note that the species occurs most frequently in barns and other anthropogenic structures (Fujita and Kunz 1984), yet this may be an artifact of where they have been observed most readily. Other investigators have found the species roosting in caves, crevices, and trees, in addition to buildings.

Veilleux has documented roosting females almost exclusively in trees in Indiana (e.g., Veilleux et al. 2003, Veilleux and Veilleux 2004a and b). In Indiana, tricolored bats accounted for only 2.9 percent of bat colonies in buildings, though the species is relatively common in that state (11.5 percent of mist net captures) (Veilluex et al. 2003). Some biologists even assert that across its range, the species favors tree roosts in summer, and rarely occurs in buildings (Harvey et al. 2011).

Female tricolored bats that roost in trees tend to roost in the foliage, rather than in cavities, and the roosting location is usually high up in the canopy (Veilleux et al. 2003). Carter et al. (1999) tracked tricolored bats to roosts in living trees in a bottomland woodland, and approximated the locations of the roosts at mid-canopy. If, in its natural habitat, the tricolored bat is predominantly a tree foliage rooster, as opposed to being predominantly a tree cavity or tree bark rooster, it would be unique in that behavior among north-temperate bat species. Other foliage roosting species, such as *Lasiurus* spp. (hairy-tailed bats), roost individually, unlike the tricolored, which is a colonial rooster (Veilleux et al. 2003).

For roosting, clusters of dead leaves are used more often than clusters of live leaves (Veilluex et al. 2003). In Indiana, the tricolored bat prefers oak trees over other tree species (Veilleux et al. 2003), perhaps because the dead leaves of oaks last longer than the dead leaves of other trees. Dead-leaf roosts are shaped like umbrellas, with a "roof" and hollow core in which bats rest. Live-leaf roosts are generally dense, though bats may also roost on branches with only a few leaves to shelter them (Veilleux et al. 2003). Live trees are used far more than dead trees, though again, reproductive females seem to prefer dead leaves over live foliage or squirrel nests (Veilleux et al. 2003). Other tree species used by the tricolored bat for roosting include yaupon holly (*Ilex vomitoria*), sparkleberry bush (*Vaccinium arboreum*), redbay (*Persea borbonia*), and pines (*Pinus* spp). The tricolored bat has also been reported to roost in Spanish moss (Menzel et al. 1999) and in tree cavities (Menzel 1996). In Nova Scotia, the tricolored bat utilizes conifers preferentially to deciduous trees and roosts exclusively on the north-northwest side of trees, usually in bearded lichen (*Usnea* spp.) (Quinn and Broders 2007).

Some males and non-reproductive females may roost in summer in their winter hibernacula (Carter et al. 1999). In a study of night roosting activity of bats in the central Appalachians, tricolored bats were found to use caves for roosting between bouts of nocturnal foraging activity (Agosta et al. 2005). Adult males were most prevalent in the night roost caves, and numbers for all demographic groups (adult males and females and juveniles) peaked in mid to late August. All the night roosting caves were also used by one or more bat species for winter hibernation.

Males and females may exhibit different roosting behavior. Males overwhelmingly roost alone and on dead leaves of deciduous trees, and can remain at the same roost for extended periods. While they have some site fidelity, it is not uncommon for males to change roosts frequently. Females seek roosts that are warmer and where temperature flux is minimal, compared to roosts used by males (Quinn and Broders 2007). However, roosts with a southern exposure may be avoided (Veilleux et al. 2003).

In Indiana, Veilleux et al. (2003) found upland and riparian habitat types were used more often than expected for roosting. O'Keefe (2009) found that non-reproductive tricolored bats in North Carolina only roosted in forest stands older than 72 years, and preferentially roosted at lower elevations, and closer to non-linear openings and water than expected by random chance. Other researchers have found that at the stand level or greater, tricolored bats seem to roost selectively in more mature forest within riparian buffers or corridors (Perry and Thill 2007, O'Keefe 2009), within a diversity of patch types, and farther than expected from roads (Perry et al. 2008).

Foraging Habitat

Scientists believe that like most insectivorous bats in North America, the tricolored bat concentrates its foraging activity over water, such as rivers and lakes (Quinn and Broders 2007), or in open woods near the edges of water (Arroyo-Cabrales et al. 2008). The scientific literature is somewhat contradictory regarding whether the species forages over open fields, however. Barbour and Davis (1969), for example, say the tricolored bat occurred over watercourses but not over forests or fields. Ford et al. (2005) detected a greater proportion of foraging tricolored bats in riparian areas in a study in West Virginia. Canopy heights tended to be higher where tricolored bats foraged, when compared with other bat species documented in the study. Additionally, other investigators have found evidence of reliance by the tricolored bat on older forest (Perry and Thill 2007, Farrow and Broders 2011). Farrow and Broders (2011) found the species was negatively associated with non-forested land (eg. agricultural and clearcut sites).

Nonetheless, the prevailing scientific view is that the tricolored bat, more so than other bats, gravitates toward more open terrain. Its wing morphology and slow, erratic flight pattern suggest a species adapted to somewhat more open environments. The tricolored bat has a high intensity call with longer frequency, meaning the species' call is stronger and travels farther than that of other bat species. This is indicative of an adaptation to foraging in more open environments (Saunders and Barclay 1992, Quinn and Broders 2007). The species is not usually found in deep forest, but is known to forage above the forest canopy. In northwestern South Carolina, tricolored bats were most likely to be recorded in acoustic surveys in areas with sparse vegetation and early successional habitat (Loeb and O'Keefe 2006). However, vegetation density as a variable was not related to the age class of the forest, suggesting that the sites where tricolored bats were most likely to be detected could have been small gaps in intact forest, as well as larger openings of cleared forest.

The tricolored bat's foraging area is relatively small (Harvey et al. 2011). Distance from roost sites to foraging areas were, at most, 4.3 km, in a study by Veilluex et al. (2003). Similar to other bats, prey abundance and the physical attributes of a site are factors in the tricolored bat's foraging location (Quinn and Broders 2007). In Missouri, the probability of occupancy for tricolored bats was highest in forests with more open understory, except at the lowest layers of the canopy, where dense vegetation was perhaps a result of more light reaching the forest floor (Yates and Muzika 2006).

E. Diet

The diet of the tricolored bat is strictly insectivorous. According to Whitaker (2004), the tricolored bat's diet consists mainly of homopterans (leafhoppers, tree hoppers), coleopterans (beetles), and dipterans (flies). Other insects found in their diet were hymenopterans (wasps, bees, ants), lepidopterans (moths and butterflies), and several other types of insects. However, according to Amelon (2006), lepidopterans were consumed more than relative availability, and coleopterans and homopterans less than relative availability. Prey items range in size from 4 to 10 mm. In a study of eight bats in southern Illinois, the diets of tricolored bats and myotid bats were similar in that they ate the most soft-bodied prey relative to other, larger bat species such as the big brown bat (*Eptesicus fuscus*). In the study, trichopterans (caddisflies) dominated the diet, followed by coleopterans and hymenopterans (Feldhamer et al. 2009).

F. Reproduction

Mating occurs in the late summer and fall in "swarms" in front of cave openings. Females mate with multiple males (Whitaker and Hamilton 1998). Biologists have also observed copulation in hibernacula in later winter (Vincent and Whitaker 2007) and upon emergence from hibernacula (Guthrie 1933, Fujita and Kunz 1984, Dodd and Johnson 2012). Females store spermatozoa in their uteri over the winter, until ovulation occurs in late spring. Tricolored males have a shorter period of sexual dormancy than other North American bats, which may be linked to the apparent capacity for spring mating. Spring mating may provide tricolored females an opportunity for re-insemination if spermatozoa were lost during hibernation (Dodd and Johnson 2012).

Parturition takes place in spring up to early or mid-summer. Gestation lasts approximately 44 days (Wimsatt 1945). In New York, for example, birth takes place half-way between the months of June and July (Wimsatt 1945). Unlike most other cave bats, the tricolored bat usually gives birth to two, rather than one, offspring per year (Wimsatt 1945, Hoying and Kunz 1998). Together, newborn twins weigh up to 52 percent of the mass of the mother (Hamlin 2012). This proportion is at the upper extreme of maternal effort among vespertilionid bats (Amelon 2006). Babies are born hairless and pink, with closed eyes, and the babies can make clicking sounds to help mother bats locate their young (Harvey et al. 2011). Young tricolored bats begin flying at three weeks and can replicate adult flight and foraging capabilities about one week later (Fujita and Kunz 1984). Young are independent at five weeks. Sexual maturity is reached anywhere between 3 and 11 months (Hamlin 2012), and may vary by location within the species' range (Fujita and Kunz 1984).

In spring, females migrate from hibernacula to maternity colonies. In southern portions of their range, females arrive from hibernacula beginning in late April; in more northerly portions of the range, females arrive through late May (Amelon 2006). Tricolored females, like many other bat species, raise their young in colonial maternity roosts. Tricolored maternity colonies are small.

Veilleux and Veilleux (2004a) report an average cluster size of 3.7 individuals. However, Whitaker (1998) found an average maternity colony size of 15 individuals.

Tricolored bat females exhibit a fairly high degree of roost fidelity (Veilleux and Veilleux 2004b), returning to the same small roosting area day after day within a single summer. They also return to the same areas across successive years. However, tricolored females may switch specific roost sites frequently during the maternity period, both with and without volant offspring (Whitaker 1998, Amelon 2006). There is some evidence that female tricolored bats are faithful to their natal roost habitat, as well (Veilleux and Veilleux 2004b). Approximately 44 percent of bats return to previous roosts after having left for new ones (Veilleux et al. 2003). Tricolored bats remain at roosts for about 3.9 days, and pregnant or lactating females may stay as long as 17 days at specific roost sites. Although the tricolored bat has been observed changing roosting sites, pregnant females remain in a single site for longer periods (Veilleux et al. 2003). This time span is longer than other foliage-roosting bat species; for example, eastern red bats (*L. borealis*) and Seminole bats (*L. seminolus*) spend about 1.2 and 1.7 days, respectively, at roosts (Veilleux et al. 2003).

Climate and food availability may play a role in post-natal growth of young (Amelon 2006). Low ambient temperatures may lead to periods of low food availability. Low temperatures in maternal roosts may demand more energy expenditure for thermoregulation of mother bats and young, which could lead directly to low post-natal growth (Amelon 2006).

G. Demography

The tricolored bat has a lifespan of four to eight years in the wild (Nowak 1991). The oldest known record is 14.8 years (Nowak 1991, Whitaker and Hamilton 1998). However, according to Walley and Jarvis (1971), the tricolored bat has been recorded living to over 15 years. Through banding, other researchers have documented tricolored bats ranging in age from seven to eleven years (Fujita and Kunz 1984). Apparent high mortality rates occur between the first and second hibernation season, probably in winter (Davis 1966). Mortality of juveniles during the period of early flight may be as high as 50 percent (Hoying and Kunz 1998).

Some research on tricolored bat indicates that survival rates decline after 3-4 years, with females' longevity lower than males' (Davis 1966). In hibernacula, investigators have found higher numbers of male tricolored bats, with a sex ratio as high as 4:1 in favor of males (Fujita and Kunz 1984). The skewed ratio may be due to lower survival rates for females, and/or females overwintering elsewhere (Davis 1959). However, summer sampling has detected an equal ratio of males to females (Davis 1959), though O'Shea et al. (2004) question Davis' results, based on his correction factors for recaptures.

H. Hibernation

Most temperate bat species spend the coldest months of the year in hibernation. As bats enter a state of torpor, body temperature falls to within 1-2°C of the ambient temperature in the hibernaculum (Geiser 2004). The decreases in body temperature and metabolic rate reduce energy expenditures by approximately 95 percent (as compared to remaining normally active) (Geiser 2004, Dunbar and Tomasi 2006).

In the winter months the tricolored bat must hibernate. This is true even in warmer southern climates (e.g. Florida and Central America), where temperatures and prey resources are suitable for continued roosting year-round (Fujita and Kunz 1984, Briggler and Prather 2003, Broders and Quinn 2007). In fact, the species hibernates in Florida caves in which ambient temperatures are too high for other bat species (McNab 1974, Fujita and Kunz 1984). The tricolored bat enters hibernation earlier and leaves later in the spring than other bat species, and the obligate nature of hibernation for the tricolored bat may be a function of a physiological need for a long period of torpor (Vincent and Whitaker 2007).

In a six-year study in a limestone mine in Ohio, Brack (2007) found that tricolored bats chose hibernation spots with stable temperatures and that this species chose warmer hibernation spots than any other species detected (p. 744). The mean temperature chosen by tricolored bats during all surveys was $9.5 \pm 1.9^{\circ}C$ (p. 743).

The tricolored bat hibernates primarily alone, rather than in clusters (Harvey 2003, Brack 2007), although groups of two or three have been observed in Texas caves (Sandel et al. 2001). Relatively few individuals occur in any given hibernacula (Vincent and Whitaker 2007).

Scientists have reported disproportionate sex ratios in hibernacula, with greater numbers of males than females (Davis 1959, Fujita and Kunz 1984). This may be reflective of an overall skewed sex ratio among tricolored bats, in which males tend to be more long-lived than females.

Tricolored bats frequently hibernate in association with other cave bat species, including the little brown bat (*Myotis lucifugus*), Keen's myotis (*Myotis keenii*), Indiana bat (*Myotis sodalis*), southeastern myotis (*Myotis austroparius*), gray bat (*Myotis grisescens*) and big brown bat (*Eptesicus fuscus*) (Fujita and Kunz 1984).

Bats periodically break torpor during the winter and may become active within or occasionally outside of the hibernaculum. Commonly suggested reasons for breaking torpor include switching hibernacula, feeding, drinking, copulating, and/or enhancing immune function (Speakman and Racey 1989, Thomas and Geiser 1997, Boyles et al. 2006, Luis and Hudson 2006). The energetic cost of these arousals represents up to 75 percent of winter energy expenditures (Thomas et al. 1990).

In Indiana, researchers observed the body masses of male tricolored bats decrease by a mean of 2.65 grams over the winter, representing a loss of about 39 percent of their body masses between September and April. Over the same time period, females lost about 2.5 grams of mass, representing a loss of roughly 29 percent (Fitch 1966, Vincent and Whitaker 2007). Because the presence of human observers itself can be disturbing to hibernating bats, it is possible that loss of mass during hibernation is affected by research (Vincent and Whitaker 2007).

The tricolored bat is believed to only rarely fly out of the hibernation site in winter (Whitaker and Rissler 1992), though it can be quite active during hibernation. Vincent and Whitaker (2007) found that the species moved often to various hibernacula. However, other investigators have found that the species, more so than other bats, remains in torpor for longer periods between arousal bouts. A maximum period of 111 days between arousal bouts was found by Brack and Twente (1985).

Hibernation and White Nose Syndrome

Clustered hibernation is known to confer significant thermal benefit by reducing heat loss, and it has been suggested that clustering may reduce the amount of body mass lost during hibernation (Boyles et al. 2008). Though the possibility has yet to be explored, the solitary hibernation behavior of the tricolored bat may thus increase its vulnerability to mortality from WNS by increasing the rate at which energy reserves are depleted. Alternatively, in principle, solitary hibernation could be an advantage with regard to WNS because fungal volumes may be lower with fewer numbers of bats.

The tricolored bat has been subject to some of the greatest mortality rates (greater than 98 percent in the northeastern United States) documented among WNS-affected bats. Both the solo-hibernating northern long-eared bat and the generally colonial little brown bat have experienced similarly high mortality levels. The lack of direct correlation between coloniality and mortality from WNS suggests other factors are at work with regard to the lethality of the fungal disease for different species (Langwig et al. 2012).

Body size may be one factor in species-specific mortality rates. The tricolored bat is one of the smallest bats in eastern North America. Depletion of fat reserves by WNS-induced arousals could become life-threatening for the diminutive tricolored bat before it becomes highly problematic for other, larger bats. The tricolored bat also hibernates for the longest span of time, when compared to other hibernating cave bats. This long hibernation period exposes the tricolored bat to growth of pathogenic fungus for a longer period.

I. Migration

Up until recently, biologists had believed the tricolored bat was a regional migrant, and that like other hibernating bat species, it moved, at most, only moderate distances (e.g., 52.8 km, reported by Griffin 1940) between hibernacula and summer roosting areas. However, recent research suggests that at least some tricolored bats, namely males summering in the northernmost reaches of the species' range, migrate latitudinally more like long-distance migrants such as hoary bats (*Lasiurus cinereus*), and silver-haired bats (*Lasionycteris noctivagans*) (Fraser et al. 2012).

The relatively high mortality of tricolored bats at wind energy sites, like other migratory, non-hibernating bat species and unlike most other hibernating species, indicates that the tricolored may behave differently than other hibernating bats when it comes to seasonal movement. Within its range, the tricolored bat is among the most frequently killed bat species around wind turbines and may account for up to 25 percent of total bat mortality at wind facilities (Fraser et al. 2012, p. 2).

Fraser et al. (2012) analyzed stable hydrogen isotopes in fur samples from tricolored bat museum specimens and found that 33 percent of males and 16 percent of females were collected south of the location where they had been during the fur-growing season, and that in general, the females had not travelled as far as the males. Most migrants were collected at the northern and southern extents of the species' range. Their results show a different pattern of migration for the tricolored bat than had been previously documented, indicating that some individuals undertake annual latitudinal migrations and that migratory tendency varies with latitude and between sexes. The authors suggest that this species' hibernation ecology makes it particularly vulnerable to long winters, making migration from the northern extent of the species' range to more southern hibernacula preferable for some individuals, and that sex-biased differences in migration may be the result of differences in reproductive pressures.

IV. POPULATION STATUS

White-nose syndrome has recently decimated tricolored bat populations in several states, but before the onset of WNS, the tricolored bat was generally believed to be common and secure throughout most of its range in the eastern United States. Historically, little research and monitoring of this species were conducted. However, an analysis of survey data suggests that even prior to WNS, the tricolored bat, along with several other WNS-affected species, was in a state of gradual decline in the eastern United States (Ingersoll et al. 2013). Correcting for biases inherent in hibernacula counts, Ingersoll et al. (2013) found that from 1999 to 2011, (i.e., both pre and post-WNS), the tricolored bat declined by 34 percent in the multi-state study area (New York, Pennsylvania, West Virginia, and Tennessee). Further, while WNS has been assumed to be the sole driver of bat population declines, new research indicates that many factors are likely acting synergistically (Ingersoll et al. 2016, p. 12). Indeed, local declines of major tricolored bat populations always began three to seven years prior to the detection of WNS in those populations (Ibid., p. 13). The causes for the tricolored bat's pre-WNS decline—causes that are presumably ongoing in the post-WNS environment—are not explored in depth in this new study, but the authors suggest that other threats likely contributing to the decline in tricolored bat populations include: loss and disturbance of critical roost (Thomas 1995) and foraging sites (Jones et al. 2009); toxicity from agricultural pesticides and other chemical compounds (Clark 1988); altered roost microclimates, foraging habitats, and prey communities from climate change (Rodenhouse et al. 2009; Frick et al. 2010b); and heightened mortality from inflight collisions with vehicles, buildings, and wind turbines (Arnett et al. 2008; Russell et al. 2009; cited in Ingersoll et al. 2016, p. 15-16).

Due to the declining population trend and increasing threats to the tricolored bat, several entities have reclassified or are in the process of reclassifying its conservation status to reflect the downward trend. The scale of recent loss (34 percent) reported in Ingersoll et al. (2013) puts the tricolored bat at the threshold of "Vulnerable" under the IUCN's Red List criteria, but the species is still categorized as "Least Concern" and an update is warranted, as the IUCN account was last updated in 2008 and still reflects the previous common name of Eastern pipistrelle.

Other conservation assessments of the tricolored bat have also become very out-of-date in the last several years, though some corrections have been made. NatureServe changed the tricolored bat's global conservation status to the more imperiled rank of Vulnerable (G3) in July 2012 (NatureServe 2014). This change reflects the dramatic decline of the species in numerous parts of its range in the last several years. Some state rankings have been updated and some still need to be updated. The bat is ranked as critically imperiled (S1) in Nebraska, Pennsylvania, Vermont, New Hampshire (S1N, SUB), Wisconsin (S1S3), Nova Scotia, and Quebec, as imperiled (S2) in Michigan and New Brunswick, as vulnerable (S3) in Alabama, Massachusetts, Minnesota, New Mexico, North Carolina, Virginia, and Ontario, and as apparently secure or secure throughout the rest of its range.

Five states have classified or are in the process of listing the bat as endangered or threatened. The states of Massachusetts and Vermont classify the tricolored bat as endangered (Massachusetts Division of Fisheries and Wildlife 2012, Vermont Fish and Wildlife Department 2012). In New Jersey the bat is proposed as endangered and awaiting the final rule (New Jersey Division of Fish and Wildlife 2013). In Pennsylvania the bat has been proposed for endangered status, but the final listing has been delayed. The Pennsylvania Biological Survey's Mammal Technical Committee proposed the bat for listing in 2012 but the addition has not yet been finalized and is facing significant opposition from commercial interests including the logging, coal mining, and wind industries (Pennsylvania Game Commission 2013). The tricolored bat is listed as threatened by the state of Wisconsin (Wisconsin Department of Natural Resources 2011).

In Indiana, Maine, Michigan, Minnesota, and Ohio the tricolored bat is considered to be a species of special concern (Indiana Department of Natural Resources 2013, Maine Department of Inland Fisheries and Wildlife 2011, Michigan Department of Natural Resources 2014, Minnesota Department of Natural Resources 2013, Ohio Department of Natural Resources 2012).

Delaware, Rhode Island, South Carolina, and Texas list the bat as a species of greatest conservation need (Delaware Division of Fish and Wildlife 2015, Rhode Island Department of Environmental Management 2015, South Carolina Department of Natural Resources 2015, Texas Parks and Wildlife 2012). Nebraska lists the bat as a Tier II at-risk species (Nebraska Game and Parks 2014).

The tricolored bat is classified as a species of lowest conservation concern in Alabama (Alabama Department of Conservation and Natural Resources 2007). The tricolored bat has no special status in Arkansas, Connecticut, Florida, Georgia, Illinois, Iowa, Kansas, Kentucky, Louisiana, Maryland, Mississippi, Missouri, New Hampshire, New Mexico, New York, North Carolina, Oklahoma, Tennessee, Virginia, or West Virginia (Arkansas Game and Fish Commission 2013, Connecticut Department of Energy and Environmental Protection 2010, Florida Fish and Wildlife Conservation Commission 2014, Georgia Wildlife Resources Division 2014, Illinois Department of Natural Resources 2011, Iowa Department of Natural Resources 2012, Kansas Department of Wildlife, Parks, and Tourism 2005, Kentucky Department of Fish and Wildlife Resources 2014, Maryland Department of Natural Resources 2005, Mississippi Department of Wildlife Fisheries and Parks 2005, Missouri Department of Conservation 2014, New Hampshire Fish and Game 2011, New Mexico Game and Fish 2007, New York State Department of Environmental Conservation 2014, North Carolina Wildlife Resources Commission 2014, Oklahoma Department of Wildlife Conservation 2014, Tennessee Wildlife Resources Agency 2005, Virginia Department of Game and Inland Fisheries 2010, West Virginia Division of Natural Resources 2012).

Most of the Canadian population of the tricolored bat has already been exposed to WNS. The decline of the species in Canada is estimated at 94 percent, and the species was listed as endangered by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) in November 2013 (COSEWIC 2014).

V. THE TRICOLORED BAT WARRANTS PROTECTION UNDER THE ESA

Under the ESA, 16 U.S.C. § 1533(a)(1), USFWS is required to list a species as threatened or endangered if it is in danger of extinction or threatened by possible extinction in all or a significant portion of its range. In making such a determination, USFWS must analyze the status of the tricolored bat in light of five statutory listing factors:

A. Present or threatened destruction, modification, or curtailment of its habitat or range;
B. Overutilization for commercial, recreational, scientific, or educational purposes;
C. Disease or predation;
D. Inadequacy of existing regulatory mechanisms;
E. Other natural or manmade factors affecting its continued existence.

1533(a)(1)(A)-(E); 50 C.F.R. § 424.11(c)(1) – (5).

Four of these five factors threaten tricolored bats. The species is imperiled by habitat modification and loss, disease, the inadequacy of existing regulatory mechanisms, and other natural and manmade factors. Threats to the tricolored bat in light of each of these factors are discussed in detail below. Because of the species' susceptibility to WNS, along with other imminent threats that likely act in synergy with WNS, the tricolored bat clearly warrants protection under the Endangered Species Act.

VI. THREATS

Tricolored bats are threatened by loss of habitat through development, logging, and mining, wind turbines, WNS, environmental contaminants, and climate change. While WNS is the gravest threat to the bats' survival, the impacts of this disease seem to be exacerbated by the other factors described below (Ingersoll et al. 2016, p. 15). Thus, immediate protection is needed to safeguard the species and its entire habitat while scientists gain a better understanding of each threat and how to mitigate them.

A. Present or threatened destruction, modification, or curtailment of its habitat or range

Numerous factors threaten the habitat of the tricolored bat including residential, commercial, and agricultural development, logging, and energy development including coal mining, oil and gas development, and wind turbines.

Residential, Commercial, and Agricultural Development

Tricolored bats are threatened by habitat loss due to residential, industrial, commercial, and other development activities that cause conversion of habitat. Between 1982 and 2010, 43 million acres of land in the United States were newly developed, bringing the total acreage of developed land to approximately 113 million acres, a 58 percent increase in developed land over a roughly 30-year period (U.S. Department of Agriculture 2013, p. 8). Of note, more than 37 percent of developed land was developed since 1982, with every one of the 48 conterminous states showing statistically significant increases in developed land area since 1982 (U.S. Department of Agriculture 2013, p. 8).

Expansion of suburban habitats is fragmenting natural habitats that are important to the tricolored bat including forests and grasslands near water. For example, one of the most rapidly developing areas is the southeastern United States including many states in the bat's range where urbanized land is expected to double or triple within the next fifty years at the expense of agricultural and forested landscapes. Terando et al. (2014) developed models to project growth for the Southeast and predicted that the extent of urbanization in the region will increase by 101 to 192 percent by 2060 (p. 1). Increases in the urban development footprint are occurring even in areas where population numbers alone are not showing large increases due to increasing sprawl, indicating that population growth models alone may under-predict the future extent of urban areas (Terando et al. 2014, p. 1). Theobald (2005) reports that the footprint of developed land in the United States increased from 10.1 percent to 13.3 percent from 1980 to 2000, and outpaced the population growth rate by 25 percent (p. 22).

In terms of the footprint of developed land, the mean size of urban land patches is expected to increase by 79 percent, while the mean size of natural land types will decrease as fragmentation increases. For example, agricultural land in the Southeast is expected to undergo the greatest fragmentation with a mean decline of 46 percent in the largest patch size and a decline of 22 percent in mean patch size. The authors predict that in the Southeast by 2060, from 11 to 21 percent of current agricultural land will be urbanized, 9 to 17 percent of current grassland will be urbanized, and from 7 to 12 percent of current forested land will be urbanized (Terando et al. 2014, p. 5).

The loss of natural habitats to urban development is not unique to the Southeast, but is occurring throughout the bat's range. Across the United States, human population growth and the expansion of development into formerly rural or undeveloped lands has been rapidly accelerating over the past fifty years. In 1950, less than 1 percent of the conterminous United States was classifiable as urban, and 5 percent was classifiable as exurban, but by 2000, 2 percent of the total land area was urban, and 25 percent exurban (Brown et al. 2005, p. 1855). Since 1950, exurban area increased sevenfold to eightfold in the non-metropolitan counties of the Eastern Temperate ecoregion (p. 1856), which occupies the majority of the tricolored bat's range. The Great Plains, which occupies the westernmost portion of the tricolored bat's range, also experienced increases in exurban area (Brown et al. 2005, p. 1856). Every eastern ecoregion has experienced losses of agricultural land since 1950, most due to urbanization (Brown et al. 2005, p. 1857).

Though no statistics are available that specifically relate bat decline to habitat loss caused by development, this is due to a lack of research on bats rather than to the species' tolerance of such habitat loss. The devastating effects of habitat loss and fragmentation on wildlife populations in general are well documented in the scientific literature (eg. Johnson and Klemens 2005, entire, Dirzo et al. 2014, entire, Martinuzzi et al. 2015, p. 160). In the United States, destruction of habitat is the most pervasive threat to vertebrate species, impacting more than 92 percent of imperiled mammals, fish, amphibians, reptiles, and birds (Wilcove et al. 1998 cited in Martinuzzi et al. 2015, p. 161). Riparian areas and temperate forests, two habitat types essential to bat species, are particularly affected by expanding residential development (Smith and Wachob 2006, p. 431, 432). The tricolored bat uses forested habitat for summer roosting and the formation of maternity colonies, and relies on the insect abundance fostered by riparian habitats to meet the energetic requirements associated with reproduction. Reduced connectivity among requisite habitat types (e.g., roosting and foraging) increases general energy expenditures and may contribute to local declines.

Expansion of industrial agricultural development can also degrade habitats that are important to the tricolored bat. Lemly (2000) reports that intensive agriculture can negatively affect wildlife habitat both by draining wetland areas and by polluting surface waters (p. 1). Wickramasinghe et al. (2003) identify agricultural intensification as a major cause of the decline in European bat populations (p. 984). They found that total bat activity was significantly higher on organic farms than on conventional farms and recorded more bats passing over water on organic farms than on conventional farms. They report that bat foraging activity was significantly higher on organic farms than on conventional farms. The authors conclude that bats are bioindicator species and that declining bats are “victims of agricultural change” (p. 984).

To evaluate the impact of agricultural intensification on bat foraging, Wickramasinghe et al. (2004) quantified the availability of bat prey by comparing nocturnal aerial insects captured within habitats on 24 matched pairs of organic and conventional farms in the United Kingdom. They found that insect abundance, species richness, and moth species diversity were significantly higher on organic farms than on conventional farms, and that insect abundance was significantly higher in pastural and water habitats on organic farms than in the same habitats on conventional farms (p. 1283). They conclude that that agricultural intensification has a profound impact on nocturnal insect communities, and that because bats are resource limited, reduction in prey availability due to agricultural intensification adversely affects bat populations (p. 1283).

Logging

The loss of forested habitat threatens the tricolored bat. Logging affects bat populations through the loss of roosting and foraging habitats, changes in forest structure, and changes in insect distribution and abundance (Hayes and Loeb 2007, entire, Yates and Muzika 2006, p. 1238). A substantial proportion of land within the range of the tricolored bat is privately owned and subject to logging activity. State and federal lands within the species’ range are also subject to frequent logging. The most commonly employed silvicultural practices may be incompatible with bat habitat conservation for several reasons. Tricolored bats appear to roost primarily in trees (Veilleux and Veilleux 2004a), and they exhibit high site fidelity (Veilleux and Veilleux 2004b). It appears that many species of North American bat have experienced population losses due to degradation of or disturbance at summer roosting sites (Veilleux et al. 2003, p. 1068). Tree-roosting bats require a relatively large number of suitable trees during the roosting season, as they tend to relocate from one tree to another every few days (Veilleux and Veilleux 2004a). Logging of roosting areas to which bats return throughout a summer season, or from one year to the next, may displace bats and precipitate energetic costs, greater exposure to predation, disruption of social bonds with other colony members, and loss of socially-transmitted knowledge and behaviors that enhance survival (Veilleux and Veilleux 2004b).

The resources that intact, mature stands provide (e.g., diversity of potential roosting sites, proximity to foraging habitat) are necessary to many parts of bat life history, and are essentially excluded by most silvicultural management. For example: the use of short-rotation cuttings (20-40 years) does not allow development of the mature stands with complex vertical structure. The density of snags and large-diameter trees is consistently lower in managed forests: snags with a diameter at breast height (dbh) greater than 50 cm were more than 50 times more abundant in old-growth stands than in 21-40 year old managed stands in western Oregon (cited in Hayes and Loeb 2007, p. 209). Several other studies show similar trends: the proportion of snags and large-diameter trees is dramatically reduced in managed stands, even those on relatively long rotation times (40 years or more) (e.g., Duvall and Grigal 1999, Nelson and Lambert 1996, Graves et al. 2000).

Commercial forestry occurs throughout the tricolored bat's range. New England's Northern Forest spans 26 million acres in central and northern Maine, northern New Hampshire and Vermont, and the Adirondack and Tug Hill regions of New York. It is one of the largest contiguous forested regions in the United States, and 84 percent (21.8 million acres) is controlled by private landowners (Dobbs and Ober 1995). Forestry is the dominant industry in the Northern Forest, and has been since the late 18th century. Commercial forestry companies own more than 60 percent of Northern Forest lands, and the majority (70 percent) of commercially owned land is controlled by a few paper corporations (Dobbs and Ober 1995). Much of the Northern Forest was cut for construction and to accommodate pasturelands in the early years of European settlement, but has significantly regenerated since the mid-19th century; the historical effects of this reforestation on regional bat populations are not documented, but it seems likely that bats would benefit from the regrowth of forested habitat. The early 20th century was a period of some interest in "sustained yield" forestry, and as the timber and pulp industries expanded in the Southeast and Western states, the Northern Forest was largely unaffected (Dobbs and Ober 1995). Beginning in the 1960s, however, the advent of industrialized, motorized logging brought renewed levels of harvesting to the North: the ease of whole-tree harvesting and clear-cutting, and the rising value of mature trees drove logging into many previously uncut or mature second-growth forests. Real estate interests in much of the Northern Forest also spiked in the 1980s, and many timber holdings were liquidated for the development of vacation homes and recreation areas (Dobbs and Ober 1995). The state of Maine has the highest proportion of land controlled by timber interests in the Northeast: 90 percent of Maine's 17.5 million acres is forested; nearly half of this land is owned by Fortune 500 paper and timber companies, and only five percent is public land, leaving nearly 98 percent of the state's forests open to timber management (Dobbs and Ober 1995).

Likewise, the Southeast is also a major industrial forestry region. The Southeast currently supplies 70 percent of the nation's pulp and paper (Buckner et al. 2002, p. 9). These products do not require wood of any specific size or quality and are therefore most efficiently provided by clearcutting. Short-rotation clearcutting is standard forestry practice in Southeastern forests, the vast majority of which are privately owned (Wear and Greis 2011, p. 5). Folkerts (1997) reports that the rate of deforestation in this region exceeds that of any comparably-sized tropical area; this destructive harvesting practice has altered more than one-third of the forested area in the Coastal Plain (Folkerts 1997). A high proportion of forested lands in the Southeast are privately owned and therefore not subject to environmental regulation that would enforce the use of management practices designed to minimize negative environmental effects (Morse et al. 1997). Regulation of harvesting activities on privately owned timberlands is virtually nonexistent.

Intensive logging can negatively impact tricolored bats in several ways due to reduced habitat availability and reduced fitness (Veilleux and Veilleux 2004, Yates and Muzika 2006, Hein et al. 2009, Perry et al. 2011, Sheets et al. 2013).

Perry et al. (2011) demonstrated fidelity of individual tricolored bats over multiple years to specific summer forested areas for foraging and drinking. In an eight-year study in the Ouachita National Forest, the authors determined that some tricolored bats, as well as other species such as eastern red bats, Seminole bats, evening bats, and northern long-eared bats, either utilized the same forested streams throughout the year or returned to the same sites each summer (p. 113). The authors conclude that intra- and inter-annual site fidelity by forest-dwelling bats to individual forest stands or foraging areas, including particular stream pools, may be a common phenomenon. There are likely benefits for remaining or returning to the same areas such as knowledge of satisfactory roosting, foraging, and drinking locations, and enhanced predator avoidance, improved mating opportunities, and reduced search time for resources (p. 115). Because returning to the same forested area confers advantages and likely improves fitness for those individuals that exhibit site fidelity (Perry et al. 2011, p. 115), logging would eliminate these advantages, reduce the bats' fitness, and increase energetic costs and predation risk.

Sheets et al. (2013a) studied bat use of habitat in forests in Indiana prior to logging treatments and found that even though tricolored bats forage in many types of forest habitats, they used forest interior habitat at relatively high levels (p. 213). The tricolored bat is considered a clutter-adapted species (Menzel et al. 2005b cited in Sheets et al. 2013a), meaning it can adeptly fly through areas of dense foliage.

Yates and Muzika (2006) examined the influence of forest composition, structure, and arrangement at multiple scales on the occupancy of bat species across two forested watersheds in the Ozark Highlands of Missouri. They found that at the local-site scale, the model that best predicted tricolored bat occurrence consisted of variables describing structural complexity of the forest, with total live basal area being inversely related to occurrence, but overstory canopy density being directly related to occurrence (p. 1242). The second-most-important model for tricolored bat included live basal area and understory density, with understory density from 1-2 m being directly related, but understory density from 2-3 m being inversely related to probability of site occupancy (p. 1242). Increased density of vegetation from 2-3 m represented a greater amount of shrubs and midstory vegetation in the forest, creating additional obstacles during commutes from roosting sites to foraging areas. Meanwhile, increases in vegetation density from 1-2 m represented greater density of lower shrubs, which may indicate a less dense midstory and greater light levels reaching the forest floor. The authors conclude that changes in structural complexity beneath the forest canopy impact the occupancy of a site by tricolored bats (p. 1245).

At the landscape scale, none of the models were significantly better than the null model at explaining the occupancy of tricolored bats.

Hein et al. (2009) investigated the relationship between bats and forested corridors in an intensively managed landscape in South Carolina. They examined the influence of site-level characteristics to determine which corridors provide suitable habitat for bats. They recorded 1,736 tricolored bat call sequences at 122 sites (p. 1202). They found that in fragmented environments, corridors provide important habitat features for bats. This is consistent with previous studies which have documented that linear landscape features are used by commuting and foraging bats for navigational references, commuting lanes, suitable foraging areas, and protection from wind and predators (Hein et al. 2009, p. 1204-1205). Small bats in particular, like the tricolored bat, travel farther distances along tree-lined paths, rather than flying directly to foraging areas by crossing open stands (Murray and Kurta 2004 cited in Hein et al. 2009, p. 1205). The authors conclude that corridors are an important habitat component for bat commuting and foraging and should be maintained in managed landscapes to provide habitat features needed by bats, and that maintaining these forested corridors is a practice that is currently lacking in intensively managed landscapes in the southeastern United States (p. 1206). Thus throughout the majority of the tricolored bat's range, logging presents an ongoing habitat threat and existing regulatory mechanisms do not provide adequate protection for the bat's habitat.

Natural Gas Development

The threats posed by oil, gas, and mineral development are somewhat localized but present across a significant portion of the tricolored bat's range and poses a substantial threat to many populations. New technologies developed around 2000 have allowed companies to access oil and natural gas reserves previously inaccessible, such as from shale and coalbeds (EPA 2015, p. ES-3). As a result, extraction activities have greatly expanded, especially in the eastern United States. There has been a 6,000 percent increase in shale natural gas development in the United States from 2007 to 2013 (Malakoff 2014, cited in Entreken et al. 2015, p. 2), and the Marcellus Shale in the Appalachian Basin is the most rapidly growing source of natural gas in the country (Souther et al. 2014, p. 330). The construction of wells and associated infrastructure and the use of vast quantities of water and an array of toxic chemicals in the extraction process will degrade tricolored bat habitat (Souther et al. 2014, p. 335).

The primary method of natural gas development is called hydraulic fracturing ("fracking" or "hydrofracking"). This involves fracturing rock formations using highly pressurized fluids consisting of water and various chemicals (Hein 2012, p. 1). The Marcellus and Utica shales underlie the Appalachian Basin from the Mohawk and Hudson Rivers of New York, through Pennsylvania, Ohio, West Virginia, and parts of Maryland, Virginia, and Ontario (Kiviat 2013, p. 1). Overall, high-volume horizontal fracking (HVHHF) could potentially occur over 280,000km^2 in the eastern United States. For reference, that is roughly the size of New York, Pennsylvania, and New Jersey combined.

Each fracking installation constitutes a wellpad, access road, storage areas for water, chemicals, sand, and wastewater, a compressor station, and a collector pipeline (Kiviat 2013, p. 2). The average size of a forest well installation in Pennsylvania in 2008 was 3.56ha; given the associated edge effects, that constitutes approximately 15ha of disrupted habitat per well (Johnson 2010, cited in Kiviat 2013, p. 3). In Pennsylvania alone, 2,000 Marcellus shale wells have been drilled or permitted (Johnson 2010, p. 8, 13), and an estimated 60,000 new wells will be in place by 2030 (Davis and Robinson 2012, cited in Kiviat 2013, p. 3). Fracking activities will therefore result in a significant amount of habitat loss for the tricolored bat in the eastern United States (Souther et al. 2014, p. 335).

The magnitude of shale gas development over the next couple decades will have the same impacts that other anthropogenic activities have had on bat habitat, but potentially at much greater levels (Hein 2012, p. 11). Bats rely on forests for foraging and roosting activities (Fenton 2003, Safi and Kerth 2004, Lane et al. 2006, Henderson et al. 2008, cited in Hein 2012, p. 9), and the negative impacts on bats from forest cover loss are well documented in studies on logging (Grindal 1996, Patriquin and Barclay 2003), urban expansion (Evelyn and Stiles 2003, Duchamp et al. 2004, Sparks et al. 2005a), and agricultural development (Russ and Montgomery 2002, Lesinski et al. 2007, cited in Hein 2012, p. 10). Furthermore, tricolored bats exhibit site fidelity, especially among pregnant females (Kalcounis and Hecker 1996, Sasse and Pekins 1996, Brigham et al. 1997, O'Donnell and Sedgley 1999, Weller and Zabel 2001, Menzel et al. 2002, Willis and Brigham 2004, Perry and Thill 2007, cited in Hein 2012, p. 10-11). The loss of forested habitat puts additional stress on already struggling females emerging from their hibernacula at a time of the year when prey availability is already low (Humphrey et al. 1977, Kurta et al. 1996, Murray 1999, cited in Hein 2012, p. 11).

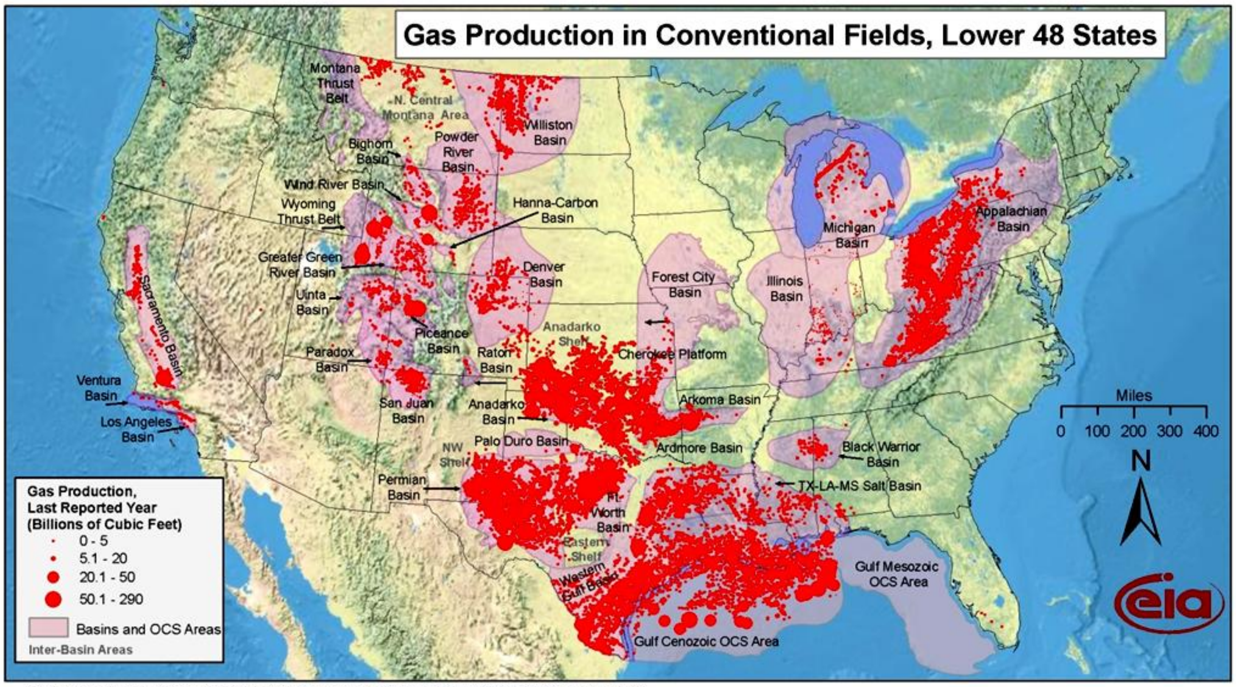
Fracking operations require extraordinary amounts of water as well, resulting in the loss or degradation of wetland and other aquatic sites in areas where water is less available or where drought is occurring (Hein 2012, p. 2). Aquatic habitats provide both a main source for insect prey as well as water and therefore play a critical role in the ecology of tricolored bats (Racey and Swift 1985, Grindal et al. 1999, Downs and Racey 2006, Hayes and Loeb 2007, cited in Hein 2012, p. 7). Bats may drink up to 26 percent of their daily water intake from open water sources, and these sources are especially important for reproductive success to provide sufficient nutrition for females' young (Adams and Thibault 2006, Adams and Hayes 2008, Johnson et al. 2011, cited in Hein 2012, p. 7-8). Thus, the extensive withdrawal of water from tricolored bat habitat, especially in sensitive areas, will impact site-selection, reproductive success, and prey availability (Hein 2012, p. 8).

In addition to direct habitat loss, it is also becoming increasingly clear that fracking contaminates groundwater and poses serious ecosystem health risks. The EPA recently identified 1,076 chemicals used in hydrofracking fluids, including acids, alcohols, aromatic hydrocarbons, bases, hydrocarbon mixtures, polysaccharides, and surfactants such as lead, ethylene glycol, benzene, toluene, and xylene compounds (EPA 2015, p. ES-11; Hein 2012, p. 2). The majority of 453 chemicals that were measured for their physiochemical properties associated strongly with soils and organic materials, meaning that they have the potential to persist in the environment for long periods of time (EPA 2015, p. ES-12). At least 65 of these are listed as hazardous by the federal government (EPA 2004). As much as a third of the injected drilling fluids remain in the ground following drilling, and numerous incidences of related water contamination have been reported across the country. However, the type and number of chemicals used in any given well are unidentified (Kiviat 2013, p. 3). Contamination from wastewater can occur at any time during fracking operations, and extensive transport and use of these chemicals in various supplies, vehicles, and equipment increase the risk of spills or leaks (Hein 2012, p. 4).

Cadmium, mercury, and lead are three well-studied and commonly associated heavy metals that can be found in HVHHF operations (Hein 2012, p. 9). These metals cause a wide array of health issues in mammals, including reproductive and renal failure, reduced immune function, hormonal changes, impaired function to the central nervous system, motor skill impairment, and hematological issues (reviewed in Hein 2012, p. 9); though these metals' direct impacts on bats is poorly studied. These toxins, as well as others mentioned above, often end up in wastewater ponds or inadequately treated in wastewater treatment plants (Kiviat 2013, p. 13, 18). As a result, many aquatic insects and insectivores such as bats are at an increased risk of exposure to these toxins by being attracted to these water sources, which then become ecological traps (Kiviat 2013, p. 3). Bats are known to congregate and drink from other industrial and toxic holding ponds (Huie 2000, cited in Hein 2012, p. 8). Similar to the cyanide poisoning of bats drinking from gold mine sites (Clark and Hothem 1991), HVHHF operations can easily result in tricolored bat mortality from poisoning (Hein 2012, p. 8). They could also suffer from reduced prey availability as insect populations decrease due to contamination. At least 80 percent of Marcellus Shale gas wells are located within 200m of riparian areas and 100 percent are within 300m (Entrekin et al. 2011, cited in Hein 2012, p. 4).

Finally, the noise and light pollution associated with natural gas extraction could impair bats' ability to forage (Legakis et al. 2000, Jones 2008, Schaub et al. 2008, Seimers and Schaub 2011, Francis et al. 2012, cited in Kiviat 2013, p. 5). HVHHF diesel compressors run 24 hours a day and can be heard from long distances (Kiviat 2013, p. 5), and installations are very brightly lit through all hours of the night (Ibid.).

The geographic coincidence of the Marcellus Shale and the current distribution of white-nose syndrome is striking (Hein 2012, p. 7). While drilling, mining, and other subsurface exploration have the potential to harm bat populations under normal circumstances, the presence of WNS greatly amplifies the threats these activities pose by compromising the health and stability of these populations (USGS 2009, cited in Hein 2012, p. 7). The habitat loss, direct disturbance, and environmental contamination associated with natural gas extraction are significant, and have the potential to further imperil regional bat populations.



Source: Energy Information Administration based on data from HPDI, IN Geological Survey, USGS
Updated: April 8, 2009

Coal Mining

The nature and spatial extent of coal mining has expanded considerably since 1990 (Wickham et al. 2013, p. 335). The development of draglines – which can excavate 100 cubic yards of earth in one scoop – and amendments to the Clean Air Act that favored the use of low-surface coal (Milici 2000, Copeland 2005, cited in Wickham et al. 2013, p. 335) have caused a dramatic uptick in coal extraction since its beginnings in 1960 (Copeland 2005, cited in Wickham et al. 2013, p. 335). The size of individual operations has increased (many individual permits now occupy more than 3,000 acres, and the largest contiguously permitted area extends over 21,700 acres), and novel industrial extraction technologies have facilitated the development of what is known as mountaintop removal (MTR) or mountaintop mining (MTM) (USGAO 2009, p. 9).

MTR is a highly destructive form of surface coal mining in which explosives are used to blast away mountain summits, exposing underlying coal seams. Rock and soil removed by this process are typically disposed of in neighboring valleys, a practice known as valley fill (VF), which buries existing streams (Palmer et al. 2010, p. 148, cited in FWS 2013, p. 61060). Coal slurry, or sludge, a byproduct of post-extraction processing, is either injected into nearby abandoned mines or stored in lagoons or surface impoundments, where contaminants leach into groundwater, poisoning wildlife and people. MTR mostly occurs in portions of West Virginia, Virginia, Kentucky, and Tennessee, and the surface area under open permit for MTR in Kentucky and West Virginia has increased annually by approximately 2% since 1990. MTR is now responsible for over 40 percent of coal production in Appalachia (USGAO 2009, p. 1), and it is one of the major drivers of land cover change in the central Appalachian region (Sayler 2008, cited in FWS 2013, p. 61060).

Surface coal mining results in high levels of disturbance in aquatic and terrestrial habitats. Bats are impacted by the loss of forested habitat, the creation of toxic wetlands on mine sites, and the removal of natural streams (Buehler and Percy 2012, p. 17). The hardwood forests of Appalachia are critical summer habitat for the tricolored bat (Veilleux et al. 2003), and in addition to the total loss of habitat represented by mined areas, MTR/VF fragments remaining forest, reducing its suitability as habitat for this species (Hayes and Loeb 2007, p. 217). The filling of streams, especially, has been found to significantly lower the densities of aquatic insects (Hartman et al. 2005, p. 96), which is a primary food source for tricolored bats. Indeed, FWS determined surface coal mining to be a significant threat to eastern small-footed and northern long-eared bats (FWS 2013, p. 61060).

Wind Energy

Wind energy has risen to being one of the top two causes of bat mortality events since 2000 (the other being WNS) (O'Shea et al. 2016, p. 2). Among North American bat species, the tricolored bat is uniquely unfortunate in its sensitivity to both the ravages of WNS and the impacts of wind energy. Wind turbines may kill 600,000 (Hayes 2013, p. 977) to nearly 900,000 bats each year (Smallwood 2013, p. 27), and within its range, the tricolored bat is the most commonly killed species, after the non-hibernating, long-distance migrants known as the "tree bats" (eastern red bat, hoary bat, silver-haired bat) (Arnett et al. 2008, p. 64).

Fraser et al. (2012, p. 2), in a study of seasonal migration in tricolored bats, write that "Within its range, *P. subflavus* is among the most frequently killed species around wind turbines and may account for up to 25 percent of total bat mortality, a much higher proportion than known regional migrants." Given that the tricolored bat is less common than the little brown bat, the next most frequently killed cave bat at wind facilities, it seems likely that the proportional mortality, as well as absolute number of tricolored bats killed, is far greater than for any other hibernating bat species.

Tricolored bats appear to migrate longer distances, along a north-south gradient, than other hibernating bat species (Fraser et al. 2012, p. 5). This propensity to travel greater distances, as the tree bats do, may at least partially explain the frequency with which tricolored bats are killed at wind energy sites. They are exposed to more wind energy projects simply by virtue of covering greater distances during the migratory period (Arnett et al. 2008, pp. 70-71).

Studies of bat fatalities at wind energy installations have identified several patterns. Peak mortality occurs from mid-summer to fall, particularly during the fall migration. Most bat fatalities occur during nights of low wind speed (Ellison 2012, p. 11; Arnett et al. 2008, p. 69). Also, most fatalities occur among adult bats, rather than juveniles Arnett et al. 2008, p. 69). Within the range of the tricolored bat, bat fatalities at wind facilities are highest on forested ridges in the eastern United States (Ellison 2012, p. 10). Larger, taller turbines kill more bats on a per turbine basis than smaller turbines (Ellison 2012, p. 10).

Scientists have theorized that bats are drawn to turbines as potential roost sites (Horn et al. 2008, p. 129) and/or to the abundance of insect prey where turbines are located (e.g., ridgelines) (Rydell et al. 2010, p. 826). Bats may perceive turbines as trees, and approach them searching for insect prey (Cryan et al. 2014, p. 4). This last hypothesis may explain why tree bats make up the majority of bats killed at wind energy sites, and why, secondarily, the tricolored bat, which shares behavior traits with tree bats as a tree/foliage rooster and as a long-distance migrant, is the most commonly killed hibernating bat at wind turbines.

Other reasons that wind turbines may be so deadly to bats is that they are frequently the highest features in a local landscape, so they may be attractive places for bats to seek prospective mates during the fall mating season (Arnett et al. 2008, p. 70). In addition, collisions with turbines may occur as a result of being located in migratory pathways (e.g., on forested ridgelines) (Ellison 2012, p. 4).

The proximate causes of mortality for bats encountering wind turbines are either direct collision (blunt force trauma due to hitting or being hit by a turbine blade) or barotrauma (abrupt air pressure changes which causes tissue damage to air-containing organs) (Rollins et al. 2012, p. 362). Some fatalities may be due to combination of both types of injury (Ellison 2012, p. 4; Grodsky et al. 2011; p. 922)).

As wind energy facilities continue to proliferate across the country, they pose a mounting threat to the tricolored bat as well as other North American bat species. Since 2008, wind energy capacity in the United States has tripled (EERE 2015a). In 2013, wind supplied about 4.5 percent of electrical demand in the United States (EERE 2015b, p. 1), and at the end of 2015, installed capacity was approximately 70,000 MW (EERE 2015c). A federal government study projects that wind energy could supply 20 percent of the nation's electricity demands by 2030 (EERE 2015b, p. 5), or approximately 350,000 MW (Bat Conservation International 2016).

Smallwood (2013, p. 29) estimated the number of bats (all species) killed in 2012 in the United States to be between 650,538 and 888,036, based on an installed wind energy capacity at the time of 51,630 MW. His estimated rate of bat mortality in the United States ranged from 12.6 to 17.2 dead bats per MW. Hayes (2013, p. 977) arrived at similar results, with an estimated total bat mortality of between 604,860 and 912,900 at 51,000 MW installed capacity in 2012. His estimated rate of bat mortality ranged between 11.86 to 17.9 bats killed per MW. Based on these estimated rates of mortality per MW, the installed capacity of 70,000 MW in 2015 may have killed between roughly 830,000 and 1.25 million bats. Thus, a future installed capacity of 350,000 MW of wind energy could mean that over 6 million bats could be killed in one year by the wind energy industry. This is an astonishing number, similar to the number the FWS estimated had been killed by WNS by 2012, six years after the disease was first discovered.

Regional differences in bat mortality from wind energy are important to consider in terms of the impact of wind industry on the tricolored bat. Within the range of the species, the rate of bat deaths (all species) is much higher than the national estimates produced by Smallwood and Hayes. For example, based on past studies, the rate of mortality in the Midwest is 31 dead bats/MW. In the Northeast the rate is approximately 32 dead bats/MW, and in the South, nearly 40 dead bats/MW (Arnett and Baerwald 2013, p. 18). Again, while the majority of these deaths are among the tree bat species, the tricolored bat is the next-most commonly killed bat species at wind energy sites.

The potential for offshore wind in the United States is estimated to be nearly three times as great as onshore wind. Currently there are proposals for wind energy projects totaling 4,900 MW offshore of nine states (EIA 2015). However, despite its placement at sea, offshore wind may still be a threat to bats, including the tricolored bat, which is among six bat species that have been detected offshore (Johnson et al. 2011 and Sjollema et al. 2014, from Arnett and Baerwald 2013, p. 35).

As both a hibernating bat, and as a long-distance migrant, the tricolored bat occupies the worst of both worlds with regard to the leading threats—WNS and wind energy—facing North American bats today. In the face of rapid growth in the wind industry, and in the absence of operational restrictions and other limits on wind energy to protect bats, it is likely to become a significant source of harm to both local populations of tricolored bats and the species as a whole.

Mine Closures

Physical barriers to closed mines and caves to keep the public safe may also have negative impacts on tricolored bats. Abandoned underground mines provide habitat for threatened bat species which have lost many of their natural cave sites to urban development, deforestation, and recreational exploration of caves (Burghardt 2001, unpaginated). Backfilling, plugging, and solid bulkhead exclosures can therefore further reduce habitat availability. In an effort to address this issue, the National Park Service has made an effort to erect bat-compatible gates and other barriers to prevent public entrance into these sites but have them available for bats (ibid.). However, this is only occurring on NPS lands, and many sites located elsewhere are not being closed properly. FWS noted that many types of doors and gates can alter the thermal regime of the cave or mine, restrict flight and movement of bats, and change the airflow, thus reducing the ability for a site to support bat populations (Hemberger 2011, unpub. data, cited in FWS 2013, p. 61058). Mining operations, passage collapse, and mine reclamation also impacts bats and their hibernacula and are considered threats to their populations (FWS 2013, p. 61058).

B. Overutilization for commercial, recreational, scientific, or educational purposes

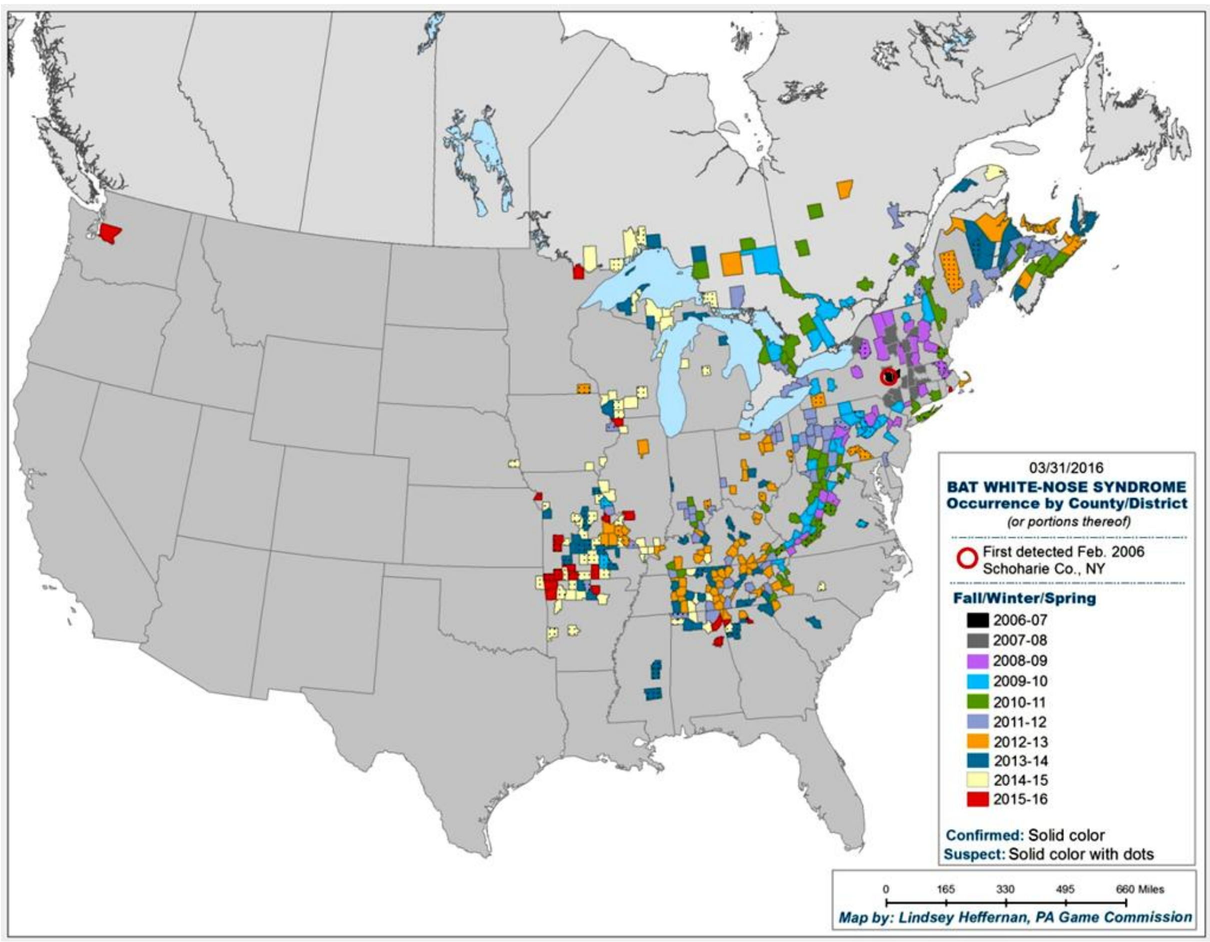
Overutilization is not known to threaten the tricolored bat at this time.

C. Disease or predation

White-nose syndrome (WNS)

The U.S. Fish and Wildlife Service has called white-nose syndrome "the worst wildlife health crisis in recent memory." WNS is a disease of North American hibernating bats and was first observed in a hibernaculum in February 2006, near Albany, New York (Blehert 2012, p. 1). WNS was named soon after its discovery for its most apparent symptom: a characteristic bloom of white fungus on the muzzle, ears, and/or wing membranes of infected bats. To date, the disease has spread to a total of 29 states in the East, Midwest, South, and Pacific Northwest, and five provinces in eastern Canada (USFWS 2016a).

The states where WNS is presently documented are New York, Vermont, Massachusetts, Connecticut, New Hampshire, Maine, New Jersey, Pennsylvania, West Virginia, Virginia, Maryland, Delaware, Tennessee, North Carolina, Ohio, Indiana, Kentucky, Alabama, Georgia, South Carolina, Illinois, Missouri, Arkansas, Michigan, Wisconsin, Iowa, Minnesota, Rhode Island, and Washington (USFWS 2015a). In Canada, scientists have documented WNS in Ontario, Quebec, New Brunswick, Nova Scotia, and Prince Edward Island (USFWS 2015a), all of which are a part of the range of tricolored bats.



The fungal pathogen, now known as *Pseudogymnoascus destructans* (or *Pd*, originally named *Geomyces destructans*) (Minnis and Lindner 2013, p. 646) has been documented in another three states (Mississippi, Nebraska, Oklahoma) (USFWS 2016a), although bats have yet to display signs of illness in these states.

Biologists consider WNS to be a grave threat to the survival of multiple bat species. Thus far, seven species are affected: tricolored bat, little brown bat (*Myotis lucifugus*), Indiana bat (*Myotis sodalis*), northern long-eared bat (*Myotis septentrionalis*), eastern small-footed bat (*Myotis leibii*), big brown bat (*Eptesicus fuscus*) and gray bat (*Myotis grisescens*) (USFWS 2015b). Mortality rates have reached 100 percent in some hibernacula in the Northeast and Mid-Atlantic States. The disease has continued to spread to new states every winter since it was first discovered. Scientists predict that WNS will eventually spread across virtually the entire contiguous United States (Maher et al. 2012, p. 7). One study projected that the disease would reach the Rocky Mountains by 2016, and would be in 93 percent of all counties with caves by the year 2105 (Maher et al. 2012, p. 4).

Actual events are outpacing predictions, however. In March 2016, WNS was discovered in the West for the first time on the West Coast, near Seattle, Washington (WDFW 2016). A sickened bat, found by a hiker, was taken to a wildlife rehab center, where it subsequently died. Lab analysis confirmed that the cause of death was WNS. The next closest known locations of *Pd* at the time were in eastern Nebraska and Minnesota. This constitutes a jump of approximately 1,300 miles. A genetic analysis of the infected bat indicated that it was a western subspecies of little brown bat, so it was likely a bat native to the location, and not a displaced eastern subspecies that brought the disease from infected regions in the eastern half of the continent (Ellison 2016).

The number of bats dead from WNS is staggering. In early 2012, the USFWS estimated that between 5.5 million and 6.7 million bats had died from WNS (USFWS 2012). Populations of bats have dropped precipitously in the northeastern United States and Canada since the advent of WNS. Bat counts in mid-Atlantic and southern Appalachian regions indicate that populations in these areas are following a similar trend. In the Northeast, by 2011 the number of little brown bats counted at hibernacula had declined by 91 percent (Turner et al. 2011, p. 18). Summer acoustic surveys in Massachusetts similarly indicated a drastic decline: Calls of *Myotis* species were down by 72 percent between 2004-2006 and 2010 (Brooks 2011, p. 2539). By early 2013, Herzog and Reynolds reported that hibernacula counts for little brown bats in Maryland, Virginia, and West Virginia were down by 72 to 99 percent. Fall-swarm capture rates for northern long-eared bats in Virginia dropped from an average of 3.6 bats/hour in 2009 to 0.5 bats/hour in 2011 (Herzog and Reynolds 2013).

More recent bat survey data are nearly as dismal. In 2013, winter surveys of three mines and one bunker in New Hampshire turned up 78 big brown bats, but only three eastern small-footed bats and just two tricolored bats. No little brown bats or northern long-eared bats were detected. The following year, in 2014, just one northern long-eared and one little brown bat were found in two mines (Preston 2015).

In New Jersey, the largest hibernaculum in the state had a total of over 26,500 bats (mostly little brown bats) pre-WNS. After WNS, the size of the hibernating colony declined to 476 by 2014 (Hall 2015).

In New York, summer acoustic surveys for *Myotis* species (mostly little brown bats) continue to show declines, despite winter reports of increased numbers of little brown bats at some hibernacula (Herzog and O'Connor 2015).

Among Pennsylvania hibernacula surveyed at least twice since WNS, the majority of bat counts are still down, although some colonies appear to have bottomed out and are now showing slight gains (Scafini and Turner 2015).

Hibernacula counts of Maryland's little brown, northern long-eared, and tricolored bats have all dropped by close to 99 percent or greater since the onset of WNS (Feller 2015).

WNS is the greatest, most immediate threat to the survival of the tricolored bat. Among WNS-affected species, the tricolored bat has suffered some of the worst mortality rates, along with the little brown bat and northern long-eared bat. After initial onset of WNS, peak mortality seems to occur a year or two later for tricolored bats than for other bat species. This phenomenon may lead to the erroneous conclusion that the species is not harmed to the same extent as other WNS-affected species. For example, in 2011 tricolored bat counts were down by 75 percent in the Northeast. At the same time, little brown bats were down by 91 percent and northern long-eared bats had declined 98 percent (Turner et al. 2011, p. 18, 19).

However, later data indicate tricolored bat declines as severe, or nearly so, within another couple years. For example, the Akron Mine in New York, which had the most sizeable colony of tricolored bats in the state, went from 968 tricolored bats in 2008 to 7 in 2014, a decline of over 99 percent. Meanwhile, Benson Cave, which had 39 tricolored bats in 2006, had none in 2014—a 100 percent mortality rate (Herzog and O'Connor 2015). In Virginia, the tricolored bat declined by nearly 90 percent between 2009 and 2012 (VDGIF 2012). More recent data from Virginia has shown that tricolored bats hibernating at sites where little brown bats were absent declined 81 percent post-WNS. However, declines were worse for tricolored bats that shared hibernacula with little brown bats. At these sites, tricolored bats declined 94 percent (Reynolds 2015). In Pennsylvania, the tricolored bat population had dropped by 98 percent by 2012 (PGC 2012). In one West Virginia cave, a colony of tricolored bats dropped by 99 percent after the arrival of WNS (Herzog and Reynolds 2013). In another study, post-WNS capture rates for the tricolored bat in West Virginia were just 22.9 percent of the historical rate, on par with decreased capture rates for the northern long-eared bat and little brown bat (Francl et al. 2012, p. 35).

Mortality rates similar to those seen in the Northeast are now emerging in the southern Midwest (Halley 2015). For example, in Ohio, three years after WNS first appeared, counts of hibernating tricolored bats have declined 98 percent. In other areas more recently hit by the disease, declines are less severe to date, but it remains to be seen what the impact of WNS will be over the next several years. In Indiana, for example, the tricolored bat has declined by 45 percent (Kocer et al. 2015). As has been seen in the Northeast, mortality of the tricolored bat may "catch up" after a few years.

One possible explanation for a delayed peak of mortality in tricolored bats is that they normally tend to position themselves deeper in hibernacula than other species, and are thereby often overlooked in winter surveys. However, tricolored bats move closer to the entrances after the onset of WNS. In the first couple years, this influx of tricolored bats from deeper in a hibernaculum offsets the loss of individuals closer to the entrance. A colony of tricolored bats may actually appear to grow or at least remain stable in the first year, while other WNS-affected bat species decline. However, after the passage of several years, there are no more recruits of tricolored bats from deeper in the site, and the tricolored bats evidence a dramatic drop (pers. comm. Greg Turner).

Various reports on hibernation behavior of the tricolored bat in WNS-positive states underscore the apparently “restless” behavior of this species as a response to disease onset. In Tennessee in 2012, tricolored bats moved around a great deal within hibernacula; they also experienced high mortality (pers. comm. Brooke Hines). In Kentucky, the tricolored bat displayed frequent roost switching, again apparently as a response to the arrival of WNS. In North Carolina, biologists observed a similar pattern of the tricolored bat’s frequent movement within hibernacula (pers. comm. Brooke Hines). In Virginia, winter counts of tricolored bats spiked in 2011 at hibernacula recently documented with *Pd*. A year later, tricolored bat numbers dropped precipitously (Powers et al. 2015, p. 119).

One other possible explanation for the lag in mortality seen in tricolored bats is their tendency to hibernate alone or in only very small groups. Initially, this behavior may confer protection as tricolored bats would be less susceptible to contracting the fungus from either conspecifics or other bat species than more social bats (e.g., Indiana bats and little brown bats). Indeed, biologists have suggested that larger bat colonies may amplify fungal growth and spread (Halley 2015). Langwig et al. (2012, p. 6) found that white-nose syndrome related declines are greater in larger winter colonies of tricolored bats, suggesting that, indeed, proximity to other bats is a disadvantage to this species when WNS appears at a site. However, tricolored bats hibernating singly and some distance from other bats may still be exposed to *Pd* over a period of several years, as the fungus spreads more completely throughout a hibernaculum, covering walls, ceilings, floors and other substrates.

Various researchers have documented not only that *Pd* is present on cave or mine surfaces (e.g., Lorch et al. 2012, p. 1299; Puechmaille et al. 2011, P. 7), but that it can grow in a wide variety of cave soils (Reynolds et al. 2015, p. 3) and on multiple, complex organic substances (e.g., insect exoskeletons, wood) that may be found in such subterranean sites (Raudabaugh and Miller 2013, p. 4; Reynolds and Barton 2014, p. 7). Thus, it seems that some tricolored bats may be belatedly exposed to infectious doses of *Pd* via the cave environment, having eluded the disease the first year or two of infection.

Interestingly, in Virginia, tricolored bats have higher mortality rates in the presence of little brown bats, as compared to tricolored bats hibernating in sites without little brown bats. Little brown bats, which may be somewhat more susceptible to WNS than tricolored bats and/or display greater sociality and clustering behavior in hibernacula, may amplify the growth and spread of *Pd*, to the detriment of the tricolored bats that share sites with them. Tricolored bats hibernating alongside a large number of little brown bats may have a higher prevalence of the disease, higher fungal loads, or earlier exposure to the fungus—all of which could result in increased mortality rates.

Other scientists have speculated that the tricolored bat’s preference for warmer hibernation environments (Perry 2013, p. 30) is disadvantageous once the species becomes infected with *Pd*. This is because *Pd* grows optimally at the higher end of the range of hibernacula temperatures (12.5-15.8°C, Verant et al. 2012, p. 4). Therefore, the tricolored bat is likely subject to higher fungal loads than species such as the eastern small-footed bat that tend to occupy colder hibernation sites (pers. comm. Kate Langwig). Tricolored bats also seem to seek out more humid conditions in hibernacula (Perry 2013, p. 30). Greater humidity is associated with higher mortality among WNS-affected bats (Wilder et al. 2011, p. 953). Fungal load may be at least a partial explanation of the high lethality of WNS in the tricolored bat.

No population modeling has been done for the tricolored bat, but the catastrophic reductions projected for two other WNS-affected bats do not bode well for the tricolored, which is likely to go down a similar path. For example, Frick et al. (2010, p. 681) predicted the regional extirpation of the little brown bat in the Northeast by 2026. In a demographic model of little brown bats for the entire eastern United States, Russell et al. (2015, p. 116) found "the minimum population size reached during the scenarios was estimated at <100,000 (<1.5 percent of starting populations) for all scenarios." The authors (p. 116) further noted that at such reduced numbers, "populations are more likely to be extirpated due to stochastic events such as poor weather conditions, human mediated landscape development, or Allee effects."

Thogmartin et al. (2013, p. 167) projected that by 2022, only 13.7 percent of Indiana bats would remain under an "acquired immunity" scenario. Under a "persistent mortality" scenario, where bats did not develop immunity to the disease, Indiana bats would fare even worse.

Prospects for the tricolored bat are likely to be at least as bad as they are for the little brown bat or Indiana bat. Frick et al. (2010, p. 682) stated: "Our analysis focused on little brown myotis in the northeastern United States, but several other bat species are experiencing similar mortality from WNS and may also be at significant risk of population collapse or extinction." Prior to WNS, the tricolored bat was not nearly as numerous as the little brown bat. In the wake of the disease, its mortality rate has been nearly as high as the little brown bat's, and considerably higher than the Indiana bat's, in most locations. There appear to be no reports of tricolored bats surviving multiple winters post-WNS, unlike little brown bats (Coleman and Reichard 2014, p. 375). All these factors indicate a demographic collapse for the tricolored bat that will be at least as severe and long-lasting as that projected for the little brown bat and Indiana bat.

Scientists have established that WNS is a fungal disease, caused by the novel species *Pseudogymnoasus destructans* (*Pd*) (Blehert et al. 2009, p. 227; Gargas et al. 2009, p. 152; Lorch et al. 2011, p. 3; Minnis and Lindner 2013, p. 646). *Pd* is psychrophilic (Chaturvedi et al. 2010, p. 5), meaning that it is cold-adapted. It is found on bats, but also on cave substrates, including in sediments (Lindner et al. 2011, p. 245) and on cave walls (Puechmaille et al. 2011, p. 7). *Pd* appears able to persist indefinitely in both cave (Lorch et al. 2012, p. 1300) and lab (Hoyt et al. 2015, p. 332) environments, in the absence of bat hosts. *Pd* produces enzymes capable of degrading non-bat sources of organic compounds (Reynolds & Barton, 2014 p. 7), and has demonstrated an ability to grow on a variety of substrates found in caves (Radabaugh & Miller 2013, p. 8; Smyth et al. 2013, p. 19). In fact, *Pd*'s apparent capacity to grow in a wide variety of cave soils, and even in substrates with low organic carbon content, may be a crucial factor in disease progression (Reynolds et al. 2015, p. 11) and the likelihood that bat colonies are fully extirpated by the disease (Reynolds et al. 2015, p. 13). Persistence of *Pd* in hibernacula without bats suggests that infected sites can act as environmental reservoirs for the pathogen, diminishing prospects for successful recolonization by bats and hindering long-term recovery (Reynolds et al. 2015, p. 13; Hoyt et al. 2015, p. 332).

Scientific consensus has gathered around the novel pathogen theory of WNS, in which the white-nose fungus originated elsewhere—possibly Europe--and recently invaded North America (Warnecke et al. 2012, p. 4). Molecular and spatio-temporal investigations point to *Pd* being a new arrival in North America that has spread rapidly, rather than an indigenous organism that was recently activated as a pathogen (Coleman and Reichard 2014, p. 375). After the discovery of WNS in the northeastern United States, scientists went looking for the disease elsewhere. The pathogenic fungus, *Pd*, was soon found on bats in caves throughout Europe (Puechmaille et al. 2011, p. 2). Although European bats acquire cutaneous infections like North American bats infected with WNS, no mortality attributable to the disease has been reported there (Coleman and Reichard 2014, p. 375). Biologists speculate that European bats co-evolved with the fungus, and may have innate resistance to it (Warnecke et al. 2012, p. 3). More recently, scientists discovered *Pd* on six species of bat in caves in three northeast China provinces (Hoyt et al. 2016, p. 140), lending support to a theory that *Pd* may have originated in Asia, moved to Europe, and then on to North America.

Laboratory research further supports the theory that *Pd* has European origins. Inoculation of little brown bats with either the European or North American strains of the fungus caused high mortality in both cases, with the European strain affecting the bats even more quickly than the fungus obtained from North America (Warnecke et al. 2012, p. 3). The greater virulence of the European strain to North American bats suggests that since *Pd* has been in North America, its lethality has attenuated, at least to a very minor degree, due to rapid evolution of the host–pathogen interaction between *Pd* and bats (Warnecke et al. 2012, p. 3).

There is evidence that the highest mortality rates for bats occur in warmer as well as more humid hibernacula (Hayman et al. 2013, pp. 4-5; Langwig et al. 2012, p. 6; Wilder et al. 2011, p. 953). Temperatures in WNS-affected hibernacula range annually from 2 to 14°C, well within the fungus' range of thermal tolerance, allowing it to persist and proliferate year-round (Blehert et al. 2009, p. 227). *Pd* can also grow and survive at higher temperatures. In fact, optimal growth occurs at 12.5-15.8°C. The upper critical temperature for growth lies between 19 and 19.8°C (Verant et al. 2012, p. 4). While vegetative growth is inhibited at high temperatures, fungal spores are not denatured by these conditions, which in fact induce germination.

Scientists have believed WNS could be less destructive in warmer climates, where hibernation is shorter. For example: "…southern populations may be shielded from sharp WNS declines by a short hibernation season" (Reynolds et al. 2015, p. 12). However, as white-nose syndrome has continued to spread in the United States, it is unclear whether the disease will be less harmful to bats in more southern latitudes. More moderate winters in the southern Appalachians of Virginia (compared with the Northeast) apparently have not protected several WNS-affected bats, including the tricolored bat. Between 2008 and 2013, winter counts of the tricolored bat declined by 89.5 percent (meanwhile, little brown bats declined 99 percent and Indiana bats 33.5 percent) (Powers et al. 2015, p. 115). In contrast, in Kentucky, summer bat counts indicate that the northern long-eared bat has declined significantly, while the tricolored bat appears stable thus far (Martin and Derting 2015, p. 17).

Biologists have also theorized that caves in more southern latitudes would be too warm for *Pd* to grow. As a rule of thumb, cave temperatures approximate the mean annual surface temperature

(MAST) (Perry 2013, p. 30). However, bats in southern caves seem to be dying at essentially the same rates as bats in more northern hibernacula. One reason why may be that while temperatures in most caves are equivalent to MAST, bats in southern regions are selecting the specific sites that offer temperature and humidity levels they need for successful hibernation. I.e., caves and mines chosen as hibernacula in southern regions are cooler than MAST, and therefore still provide suitable growing conditions for *Pd*. Perry (2013, p. 31) states: "…physical characteristics, such as cave structure and topographic setting of individual caves, may reduce or increase temperatures compared with MAST."

Similarly, as WNS continues to spread westward, the expectation that the disease will be less destructive in more arid climates may also prove false. A study of microclimates in bat hibernacula in New Mexico showed that both temperatures and relative humidity in some hibernacula were within the range for optimal growth of *Pd* (Buecher 2011, p. 16).

WNS is a cutaneous fungal infection caused by *Pseudogymnoascus destructans* (Blehert et al. 2009, p. 227). Bats affected by WNS may display some or all of the following symptoms (Kunz and Reichard 2010, pp. 14-16):

- White fungus on nose, ears, and wing membranes
- Depleted white and brown fat reserves by mid-winter
- Reduced capacity to arouse from deep torpor, particularly in response to disturbance
- More frequent arousal than normal during hibernation, and/or premature emergence from hibernacula in mid-winter
- Ulcerated, scarred, or necrotic wing membrane tissue

Pd penetrates the dermis of affected bats, eroding wing and ear tissue. Hyphae may extend into and replace hair follicles and sebaceous glands, compromising their function and eventually reaching the tissue beneath, though no local inflammation or immune response has been reported (Blehert et al. 2009, p. 227; Gargas et al. 2009, p. 148). Damage of wing tissue appears to be the most biologically significant aspect of the disease, affecting a range of physiological functions as well as flight (Willis et al. 2011, p. 365). Infected bats may fly out of their hibernacula day or night, prior to normal spring emergence, and die out on the landscape (Kunz and Reichard 2010, p. 16). Or sickened bats may perish in situ, dropping to the floor of hibernacula (Kunz and Reichard 2010, p. 13).

Initial theories about how WNS kills bats centered on energetic explanations, in which infected bats, arousing much more frequently than normal, ran out of fat reserves prematurely (Reeder et al. 2012, p. 8). However, this explanation does not address the cause of more frequent arousals (Coleman and Reichard 2014, p. 375). More recently, researchers have focused on the extensive wing damage caused by WNS, which may cause dehydration (Willis et al. 2011, p. 370; Cryan et al. 2010, p. 6) and electrolyte depletion (Cryan et al. 2013a, p. 400; Cryan et al. 2013b, p. 2; Warnecke et al. 2013, p. 1). Wings cover a disproportionally large surface area in bats, relative to other mammals, and they may play an important role in physiological homeostasis, particularly during hibernation (Cryan et al. 2010, p. 3). Dehydration resulting from damage to wings may prompt bats to arouse more frequently as they seek water. During this time they burn off fat reserves prematurely. Wing damage appears to also lead to decreased concentrations of electrolytes such as sodium (Warnecke et al. 2013, p. 3) and chloride (Cryan et al. 2013a, p. 400).

Bats face a double whammy in terms of immune response to WNS. Hibernating bats do not mount an effective immune system reaction to *Pd* colonization. The lower body temperatures of bats in torpor, nearly matching the ambient temperatures of hibernacula, appear to be associated with reduced immune function (Meteyer et al. 2012, pp. 618-19). Studies of other mammals show that hibernation leads to significant changes in behavior of T and B lymphocytes. These cells of the adaptive immune system appear to be sequestered in secondary lymphoid organs during hibernation, and do not move out to sites of infection (Meteyer et al. 2012, p. 585). In keeping with these observations in other hibernating mammals, hibernating bats infected with *Pd* show no gross evidence of pathology, no inflammation, and no recruitment of immune cells to fungal invasion sites. The lack of effective immune response to *Pd* during hibernation likely explains conditions leading to mortality during that time.

However, bats that survive to spring emergence may yet succumb once they return to euthermic conditions. As bats' immune systems become operational again, they detect *Pd* infection, which then triggers a dramatic, and potentially fatal inflammatory response. Over-responsive immune cells destroy host tissues as well as the invading pathogen, sometimes with lethal results for the host (Meteyer et al. 2012, p. 585). A similar immune response in humans is called Immune Reconstitution Inflammatory Syndrome, or IRIS. Scientists theorize that WNS may be a form of IRIS (Meteyer et al. 2012, p. 585).

A model of WNS as a multi-stage disease with various factors contributing to mortality is emerging (Verant et al. 2014, p. 4). Cutaneous infection causes some bat species to arouse more frequently from torpor, utilizing more of their fat stores. Increased arousal, with its vastly increased demand on fat stores, explains 58 percent of the morbidity associated with WNS (Field et al. 2015, p.2). Destruction of wing tissue, dehydration and electrolyte imbalance, and under-response and over-response of the immune system are among the other factors contributing to WNS pathology and bat mortality (Field et al. 2015, p. 2).

Differential susceptibility to dehydration among bat species may explain why some species fare worse with WNS than others (Willis et al. 2011, p. 370, Cryan et al. 2013b, p. 2). For example, little brown bats, like tricolored bats, have declined by well over 90% in northeastern states. Healthy hibernating little brown bats have high rates of evaporative water loss compared to a similar-sized congener in Europe (Willis et al. 2011, p. 368). The three species most frequently diagnosed with WNS (little brown bat, northern long-eared bat, tricolored bat) are the species most consistently roosting in the most humid parts of hibernacula, and are frequently seen with condensation on their fur, suggesting that these species have greater susceptibility to evaporative water loss than the species with lesser, or more variable, mortality to WNS: Indiana bat, eastern small-footed bat, and big brown bat. The latter three species are rarely seen with condensation on their fur, and often select drier areas of caves and mines for hibernation (Cryan et al. 2010, p. 4). It is possible that a species' higher baseline of evaporative water loss during hibernation correlates with greater susceptibility to WNS. Experimental evidence for this is not yet available, however (Willis et al. 2011, p. 371).

WNS affects not only the bats it kills, but also the survivors. Some bats emerge from hibernation with visible wing damage (Reichard and Kunz 2009, p. 458). Many bats captured in the summer months show substantial and sustained damage that likely compromised flight (Reichard and Kunz 2009, p. 462). However, some survivors appear able to heal the wing damage (Fuller et al. 2011, p. 5). The bats with the worst wing damage have significantly lower body mass than those with little or no WNS-induced wing damage, which could reflect lower foraging success, and possibly lead to reproductive decline (Reichard and Kunz 2009, p. 458). Scientists have indeed found evidence that bats have lower reproductive success post-WNS (Ford et al. 2011, p. 128; Francl et al. 2012, p. 36). Obviously, if true, this makes prospects for recovery, including of the tricolored bat, even more uncertain.

D. Inadequacy of existing regulatory mechanisms

No existing regulatory mechanisms can adequately protect the tricolored bat from WNS, the single biggest threat the species faces across its range, as detailed throughout this petition. While the tricolored bat is currently listed as endangered, threatened, or of special conservation concern pursuant to state law in a few states, these designations afford no significant regulatory protections (Snape and George 2010). Existing federal laws are also inadequate to protect the species. On national forest lands managed by the U.S. Forest Service under the authority of the National Forest Management Act, national forest management plans do not protect species that are not federally listed. While protections conferred by management for listed species sympatric with the tricolored bat may be beneficial to co-occurring tricolored bat populations, they are not sufficient to protect the tricolored bat because neither the life histories nor the ranges of such sympatric species are identical to those of the tricolored bat. Further, the tricolored bat's sympatry with other ESA-listed bat species does not adequately protect the tricolored bat from logging or other threats on these lands. The regulation of oil, gas, mineral, and renewable energy development on federal, state, and private lands is inconsistent and inadequate, and further development of coal and gas reserves in significant bat habitat is probable. Cave-dwelling bats are not sufficiently protected from disturbance in hibernacula by either federal or state cave protection acts. Finally, and most significantly, although the federal government is helping to coordinate a federal, state, academic, NGO, and foreign response to WNS, *see, e.g.*, http://www.whitenosesyndrome.org, these coordination efforts in and of themselves do not constitute sufficient regulatory mechanisms. In sum, while no existing regulatory mechanisms address, or can adequately address, the threat of WNS, federal listing could help address the threat of the disease by providing for regulation of activities likely to spread the disease to the species' roost sites.

In the context of its recent decision to list the northern long-eared bat (NLEB) as threatened, the Service concluded that there are insufficient regulatory mechanisms in place to address WNS, the primary threat to hibernating bats in North America such as the NLEB and the tricolored bat. 80 Fed. Reg. 17,974, 18,001 (Apr. 2, 2015). In that listing decision, after surveying existing federal, state, and foreign laws and regulations in detail, the Service concluded that these laws and regulations would not adequately protect the northern long-eared bat from WNS. 80 Fed. Reg. at 18,000–01. Because the tricolored bat shares most of its habitat and range with the NLEB and is at least as vulnerable (if not more vulnerable) to WNS, the Service should likewise conclude here that "no existing regulatory mechanisms have been shown to sufficiently protect the species against WNS" and that "despite regulatory mechanisms that are currently in place…the species is still at risk, primarily due to WNS." 80 Fed. Reg. at 18,000.

The majority of the tricolored bat's range overlaps with the NLEB's range. Because FWS concluded that no adequate regulatory mechanisms exist to protect NLEB from WNS in these areas of overlap, it should reach the same conclusion for the tricolored bat. Where the ranges of the tricolored bat and NLEB do not intersect, then it is clear that no NLEB regulations will protect the tricolored bat. In the majority of the overlapping range of the two bat species, both species have the same levels of state protection. However, the tricolored bat has even less protection than the northern long-eared bat in seven states: Alabama, Iowa, Oklahoma, Louisiana, Missouri, New Hampshire, and North Carolina (Alabama Department of Conservation and Natural Resources 2007, Iowa Department of Natural Resources 2012, Oklahoma Department of Wildlife Conservation 2014, Louisiana Department of Wildlife and Fisheries 2005, Missouri Department of Conservation 2014, New Hampshire Fish and Game 2011, North Carolina Wildlife Resources Commission 2014). The tricolored bat's range also extends beyond the range of the northern long-eared bat to an additional three states: New Mexico, Florida, and Texas. The tricolored bat receives no legal protection in any of these states (Texas Parks and Wildlife 2012, New Mexico Game and Fish 2014, Florida Fish and Wildlife Conservation Commission 2014).

As described *supra* Section IV, Population Status, a handful of states have classified or are in the process of classifying the tricolored bat as endangered or threatened pursuant to state law (Massachusetts (endangered, 2012), Vermont (endangered, 2012), Connecticut (endangered, 2015), New Jersey (proposed endangered, 2012), Wisconsin (threatened, 2015)). However, the majority of states within the tricolored bat's range accord it no protection under state law (Alabama, Arkansas, Florida, Georgia, Illinois, Iowa, Kansas, Kentucky, Louisiana, Maryland, Mississippi, Missouri, New Hampshire, New Mexico, New York, North Carolina, Oklahoma, Pennsylvania, Tennessee, West Virginia). The tricolored bat is considered a species of "special concern" or "greatest conservation need" in ten states (Delaware, Indiana, Maine, Michigan, Minnesota, Ohio, Rhode Island, South Carolina, Texas, Virginia). However, these categorizations do not provide any regulatory protection. In its definition of "species of concern," FWS specifies that "such species receive no legal protection and use of the term does not necessarily imply that a species will eventually be proposed for listing." (USFWS 2015c). The tricolored bat is also listed as endangered in Canada, but Canada makes up only ten percent of its global range (Species at risk public registry 2016).

Finally, the listing of the NLEB does not serve as an adequate regulatory mechanism to protect the tricolored bat from WNS. First, the Service's 4(d) Rule for the northern long-eared bat, finalized January 14, 2016, essentially eviscerates any protection the species would have enjoyed due to its listing as a threatened species (81 Fed. Reg. 1900). The final 4(d) rule removes almost all protections from incidental take that would otherwise have applied per 50 C.F.R. §§ 17.31 and 17.32. Incidental take is subject to restrictions only in counties within a 150-mile zone from counties in which WNS is currently present (81 Fed. Reg. 1900). Within this zone, liability for take is further shielded – liability can arise only for activities within 0.25 miles of a "known" hibernacula, or for tree removal within 150 feet of a "known occupied" maternity roost tree between June 1 and July 31 (81 Fed. Reg. at 1921). Additionally, there are no survey requirements to determine whether, indeed, there are occupied hibernacula or maternity roost trees within these small, protected zones. Due to the somewhat inconspicuous nature of hibernacula and roosts, the lack of a survey requirement makes this existing "protection" even less effective. Second, WNS is spreading at such a rapid rate that the idea of a "WNS free zone" is likely already out of date. In April 2016, the Service reported that WNS had jumped the Rocky Mountains and ended up in Washington State (USFWS 2016a). The best available scientific data seems to indicate that any "WNS free zone" will quickly become meaningless. Finally, on April 26, 2016, the Service announced that it was declining to designate critical habitat for the NLEB. The Service explained that a critical habitat designation was "not prudent" because WNS was the primary threat and a critical habitat designation would likely increase human disturbance at hibernacula (USFWS 2016b). The failure of the Service to designate critical habitat for the NLEB further weakens any derivative protection the tricolored bat might receive from the listing of the NLEB.

It is also worth noting that while WNS is the biggest threat to both the northern long-eared bat and the tricolored bat, the tricolored bat may be in even greater peril than the northern long-eared bat. The tricolored bat is particularly susceptible to wind turbine-related mortality and the species may account for up to 25% of total turbine-related bat mortality (Fraser et al. 2012, p.2), and as Ingersoll et al. (2016) demonstrated, this and other threats cause decline in tricolored bat populations often even before WNS is detected (p. 15-16). Further, the tricolored bat's tendency to hibernate for longer periods than other bats coupled with its preference for hibernating at sites located within the optimal temperature range for *Pd* may increase its vulnerability to WNS (Vincent and Whitaker 2007). Scientists have observed that the tricolored bat has been subject to some of the highest mortality rates documented among WNS-affected bats (greater than 98 percent in the northeastern United States) (Langwig et al. 2012).

E. Other natural or manmade factors

Environmental Contaminants

Tricolored bats are highly susceptible to the accumulation of heavy metals, pesticides, and other pollutants found in their environment (WIDNR 2013, p. 4), and the effect of environmental contaminants on bat populations is of significant conservation concern. Several features of the tricolored bat's life history and ecology heighten its susceptibility to contamination (Ibid.).

The unusually long life spans of bats compared to other small mammals facilitates the bioaccumulation of toxins in their tissues (Secord et al. 2015, p. 1; O'Shea and Clark 2002, p. 238; O'Shea et al. 2000, *PD*F p. 4; Walker et al. 2007, p. 3; Osborne et al. 2011, p. 67). Additionally, bats may be more susceptible than other mammals to the effects of low doses of bioaccumulative contaminants due to their annual life cycles requiring significant fat deposition followed by extreme fat depletion during hibernation or migration, at which time contaminants may be mobilized into the brain and other tissues (Clark and Shore 2001, cited in Secord et al. 2015, p. 1). The tricolored bat is also exclusively insectivorous; their primary prey is contaminated by pesticides, heavy metals, and other toxins accumulating to higher degree in bats than other non-exclusive insectivores (Secord et al. 2015, p. 1; O'Shea and Clark 2002, p. 238). Their high metabolic rate also results in the ingestion of a larger amount and quicker rate of contamination (Secord et al. 2015, p. 1). Lastly, tricolored bats often roost and hibernate in abandoned mines and drilling sites that act as faux shelter (Ducummon 2000, entire; Harvey 2003, *PD*F p. 6). In fact, the tricolored bat inhabits more caves and mines than any other species in eastern North America (Harvey 2003, PDF p. 6). Mine sites typically have higher levels of contaminants than what would be expected elsewhere.

Three primary sources of environmental contamination threaten bats: pollutants or toxins associated with agriculture, non-point source industrial pollutants (e.g., mercury), and mine-related contaminants.

Agriculturally-associated contaminants are known to be a significant threat to many bat populations. Use of agricultural pesticides (which here encompasses herbicides, insecticides, fungicides, nematocides, rodenticides, and all of the other biocides) has become widespread and toxins such as organochlorines, organophosphates, pyrethroids, neonicotinoides, and other "contaminants of emerging concern", or CECs, have long lasting lethal and sub-lethal impacts on bat populations. Bats may ingest and accumulate these toxins from their insect prey or from contaminated water sources.

The legacy of the organochlorine pesticides (OCs) used in the 20th century is well known, and they are the most intensively studied class of contaminants in bats (O'Shea and Clark 2002, p. 2). Though they are no longer used in the United States, DDT, dieldrin, and other related pesticides persist in many ecosystems and may continue to harm to bat populations (Clark et al. 1980, p. 139; Schmitt et al. 2001, p. 18, McFarland 1998, cited in Eidels and Sparks 2007, p. 51; O'Shea and Clark 2002, p. 239). OCs are neurotoxic, synthetic chemicals that are taken up by lipids and bioaccumulate in the food chain. Insects that are not killed outright by OCs can be eaten by bats which are then exposed to the toxins (O'Shea and Clark 2002, p. 2). In addition to accumulating OCs from their prey, these lipophilic compounds are especially harmful to hibernating bats because they accumulate in stored fat, and as these reserves are depleted, the chemicals are mobilized and often cause death (Secord et al. 2015, p. 1). OCs pose additional threats to pups; stored fat used for energy to provide milk exposes the young to OCs early on (O'Shea and Clark 2002 p. 2). Plus, bats maximize foraging during their reproductive season, which also maximizes their exposure to environmental contaminants (Ibid.).

Remnant OCs also have many sub-lethal effects on bats, including loss of coordination, increases in metabolic rates, and reduced levels of food consumptions (Swanepoel et al. 1999, Clark and Shore 2001, cited in O'Shea and Clark 2002, p. 240). In vitro, brain tissue of tricolored bats was sensitive to DDT and DDE levels far below environmental levels (Esher et al. 1980, cited in Thompson 2006, p. 17). While many of the studies of pesticide contamination in bats discuss the role of now-banned DDT in major historical bat declines, several more contemporary studies link other agrochemicals to bat decline. Given OCs' bioaccumulative properties and persistence in the environment, combined with bat behavior and life history, OCs continue to pose threats to tricolored bats (Schmidt et al. 2001, p. 20).

More contemporary classes of pesticides (e.g., organophosphates, pyrethroids, neonicotinoides) have also been determined to have sub-lethal effects on many bat populations (Eidels and Sparks 2007, p. 55; Mason et al. 2013, p. 7-8; WIDNR 2013; Secord et al. 2015, p. 2). Carbamate and organophosphate pesticides are the most heavily used insecticides in the world (O'Shea and Johnson 2009, p. 502), and they inhibit the neurotransmitter cholinesterase (O'Shea and Clark 2002, p. 4), affecting thermoregulation, reproduction, immune function, motor coordination, and foraging behavior (Grue et al. 1997, cited in O'Shea and Johnson 2009, p. 502) These are therefore considered to be toxic to mammals. Compromised neurotransmitter function may negatively influence navigational and foraging abilities, which may in turn affect bats' ability to meet energy requirements or expose them to predators, excessive sun, inclement weather, or drowning (Grue et al. 1997, Wilkinson 1976, Hoffman et al. 2001, cited in Eidels and Sparks 2007, p. 51; O'Shea and Johnson 2009, p. 502). Studies in Missouri and Indiana confirmed that a significant number of Indiana bat carcasses collected from hibernacula contained measurable concentrations of chlorpyrifos and dichlorvos, respectively (Eidels and Sparks 2007, p. 52), but more studies are needed on species-specific impacts.

Pyrethroid pesticides, which have recently increased in usage (O'Shea and Johnson 2009, p. 502), are also neurotoxins which result in tremoring during acute poisoning (Ibid.). They are highly toxic to aquatic organisms, but laboratory testing indicates that these compounds may be rapidly metabolized and are of low toxicity in mammals (Peterle 1991, cited in O'Shea and Johnson 2009, p. 502). However, there is concern that pyrethroids may persist in the environment (Clark and Shore 2001, cited in O'Shea and Johnson 2009, p. 502), and there has been little research on the presence or effects of these toxins in bat species (O'Shea and Johnson 2009, p. 502). Pyrethoid compounds have been reported in the guano or carcasses of Brazilian free-tailed bats (*Tadarida brasiliensis*) in Texas, little brown bats (*Myotis lucifugus*) in Missouri, and northern long-eared bats (*Myotis septentrionalis*) in Missouri (Sandel 1999, McFarland 1998, cited in O'Shea and Johnson 2009, p. 502), but the toxicological significance of these compounds is unknown. Given the extensive and growing use of these compounds, more studies are urgently needed to better understand the impacts they will have on tricolored bats.

Neonicotinoids are a relatively new class of insecticides, introduced in the 1990s, which irreversibly block post-synaptic nicotinic acetylcholine receptors (nAChRs) in the central nervous system of insects and other animals (Jeschke and Nauen 2008, p. 1084; Jeschke et al. 2011, p. 2897). Neonicotinoids include imidacloprid, clothianidin, thiamethoxam, acetamiprid, thiacloprid,and dinotefuran, and they are now one of the most widely used classes of agricultural chemicals in the United States (Hopwood et al. 2012, p. v; Hopwood et al. 2013, p. 4). These compounds may negatively impact bats by reducing prey availability and inevitably becoming cumulatively exposed to these toxins through consumption of contaminated insects (Mason et al. 2013, p. 8).

In general, correlations between bat decline and pesticides seem to be largely mediated by insects; pesticide use causes both the diversity and abundance of insects to decline, reducing the local diversity and abundance of bat species. Accordingly, organic farms have a significantly higher abundance and diversity of nocturnal insects, which supports the higher bat activity found on these facilities than on conventional farms (Wickramasinghe et al. 2003, p. 991). Finally, it has been posited that pesticides or other environmental contaminants might play some role in WNS, by compromising metabolic, neurologic, or immune function or otherwise predisposing them to infection by another agent (Cornelison et al. 2014, Hoyt et al. 2015, cited in Secord et al. 2015, p. 2, 7-8).

Finally, there is a whole host of other contaminants that potentially threaten bat populations generally referred to as contaminants of emerging concern, or "CECs". These can include detergents/surfactants, antibacterials, pharmaceuticals, personal care products, plasticizers and polybrominated diphenyl ethers (PBDEs) (Secord et al. 2015, p. 2). CECs such as PBDEs may interact with thyroid functions in bats, and therefore manipulate their hibernation timeframes, weakening their immune systems and further exacerbating the impacts of WNS (Meteyer et al. 2012, Thuvander and Darnerud 1999; Martin et al. 2007; Liu et al. 2012, cited in Secord et al. 2015, p. 9). PBDEs were found in the brain and fat tissues of little brown bats (*Myotis lucifugus*) in New York (Kannan et al. 2010, cited in Secord et al. 2015, p. 2).

Other CECs have been directly linked between sewage plant effluent and insects eaten by bats (Park et al. 2009, cited in Secord et al. 2015, p. 2). Twelve endocrine disrupting chemicals were detected in the insects at sewage treatment plants used as foraging sites by common pipistrestrelles (*Pipistrellus pipistrellus*) (Ibid.). The researchers hypothesized that the concentration of EDCs found could have detrimental effects on the foraging bats (Ibid.). Antibacterials and antibiotics may directly interfere with the fungal-bacterial balance in bats fighting WNS (Cornelison et al. 2014, Hoyt et al. 2015, cited in Secord et al. 2015, p. 2). Agriculture is a proven source of antibiotics to the environment through livestock rearing practices and could negatively impact bat populations (Sarmah et al. 2006, cited in Secord et al. 2015, p. 2). Effects of these contaminants warrant further study, especially in how they relate to bats' ability to fight WNS (Cornelison et al. 2014, Hoyt et al. 2015, cited in Secord et al. 2015, p. 7-8).

Non-point source pollutants are a threat to tricolored bats. Mercury is a neurotoxin that has been linked to numerous adverse health impacts in mammals; reduced immune function, impaired function of the central nervous system (sensory and motor skills), and compromised reproductive ability are among the most pernicious of the documented consequences of mercury contamination (Schweiger et al. 2006, p. 6). The primary sources of mercury contamination in the United States are coal-fired electric utilities, utility oil boilers, municipal waste combustors, medical waste incinerators, pulp and paper production, chlorine production, hazardous waste incinerators, metal production, and landfills (Driscoll et al. 2007, p. 20). Although mercury emissions have been heavily curbed as a result of recent regulations, most of the reduction comes from incinerators; mercury emissions from electric utilities, mainly coal-fired power plants, remain largely unchanged (Driscoll et al. 2007, p. 18). In fact, emissions from electric utilities have increased from comprising 25 percent of total emissions to 40 percent (Ibid.). Overall, total anthropogenic Hg emissions from all sources in the United States equal 103 metric tons of mercury per year.

Furthermore, the northeastern United States – which comprises a significant portion of the tricolored bat's range – has a combination of geographic features which make it a hotspot for mercury contamination (Driscoll et al. 2007, p. 18; Evers et al. 2007, p. 41). Deposition of acid rain is higher in the region resulting from industry pollution in the Midwest (Ibid.). Forested regions with a high proportion of wetlands, as are found in the Northeast, also seem to be especially sensitive to mercury contamination; ionic mercury (Hg) deposited in water is converted to methyl mercury (MeHg), a more bioavailable form of mercury that is ultimately found at toxic concentrations in the highest trophic levels (Driscoll et al. 2007, p. 18).

Though researchers have conducted most of their studies on fish and fish-eating predators, recent evidence shows that high levels of this heavy metal also affect insectivores (O'Shea et al. 2000, *PD*F p. 4; Schweiger et al. 2006, p. 12). Eleven out of 13 bat species sampled in New England and Mid-Atlantic States were contaminated with Hg levels which exceeded the level of concern (10 ppm, *See* Osborne et al. 2011, p. 59), including the tricolored bat, which was sampled in Virginia and West Virginia (Ibid. at 63). Hazardous levels of mercury have been found in other tricolored bat populations as well, such as in Arkansas and Kentucky (Evers and Yates 2006, p. 8; Massa and Grippo 2000, Powell 1983, cited in Osborne et al. 2011, p. 63, 68-69).

Aquatic nymphs of flying insects are major sources of Hg in aerial insectivores (Osborne et al. 2011, p. 68). Tricolored bats favor wetlands and riparian areas as foraging habitat (Owen et al. 2004, Ford et al. 2005, Menzel et al. 2005, cited in O'Keefe 2009, p. 24) and are at higher risk to contamination when feeding in these areas (Osborne et al. 2011, p. 68). The effects of Hg in aquatic and terrestrial food webs are detrimental to tricolored bat populations. With the majority of bats exceeding mercury contamination levels that resulted in adverse impacts in rodents in Osborne et al.'s 2011 study, it is clear that mercury poisoning threatens the tricolored bat (Evers et al. 2012; Osborne et al. 2011, p. 69).

Mining-related contaminants are an acknowledged threat to bats. As stated earlier, the tricolored bat inhabits more mines than any other eastern North American species (Harvey 2003, *PD*F p. 6), so the potential to be adversely impacted by the toxins prevalent in mines is severe. More generally, leaching and spills from mining and other extraction activities have annually caused substantial wildlife mortality, and bats have been disproportionately affected (Eisler and Wiemeye 2004, p. 35).

The mine-related contaminant that has received the most research attention to date is cyanide. Many gold mining operations use cyanide extraction techniques to chemically separate gold from other minerals; the contaminated solution that remains is then stored in sludge ponds or heaps (O'Shea et al. 2000, *PD*F p. 2). Wild animals are often attracted to cyanide ponds to drink, a behavior that may be fatal as cyanide is absorbed quickly and acts as a rapid asphyxiant. Unlike many other environmental toxins, however, cyanide does not persist in ecosystems, and does not biomagnify; sublethal doses may be ingested, detoxified and eliminated easily (Eisler et al. 1999, cited in O'Shea et al. 2000, *PD*F p. 2).

Climate Change

Climate change is widely recognized as an imminent threat to individual species and entire ecosystems alike. Temperate zone bats may be particular vulnerable because their prey availability, reproductive cycles, hibernation patterns, and migrations are closely linked to temperature and climate (Burles et al. 2009, cited in Sherwin 2012, p. 3; Racey 1982, Humphries et al. 2002, Jones et al. 2009, Newson et al. 2009, cited in Loeb and Winters 2013, p. 103). Ecological alterations wrought by climate change will vary by region, but alterations of precipitation, stream flow, and soil moisture will impact the distribution and abundance of insects, which is already changing in some areas (e.g. Hughes 2000, Bale et al. 2002, Menendez 2007; Rodenhouse et al. 2009, p. 250). Small mammals with high energy demands such as bats are more vulnerable to changes in prey availability (Rodenhouse et al. 2009, p. 250), and increased foraging costs would likely result in further population declines for the tricolored bat (Frick et al. 2010, cited in Loeb and Winters 2013, p. 104).

Changes in temperature may directly impact tricolored bat survival. Milder winter conditions may permit bats to enter hibernacula later than usual; however, less food is available later in fall, which would cause them to enter hibernation with fat reserves inadequate to last the winter. Warmer or more variable winter temperatures may also induce more frequent arousals in hibernating bats. Energy requirements are minimized at approximately 2º C, while warmer or cooler temperatures greatly increase energetic costs; rising or fluctuating temperatures may thus cause arousal frequencies unsustainable for these species (Humphries et al.2004, cited in Rodenhouse et al. 2009, p. 254). This will further enhance their susceptibility to pathogens and infections such as WNS (Jones et al. 2009, Boyles and Willis 2010, cited in Sherwin 2012, p. 12). Tricolored bats are particularly sensitive to overwintering habitat conditions. They tend to occupy the deepest parts of caves where the temperature is highest and least variable (Fujita and Kunz 1984, Raesly and Gates 1987, Briggler and Prather 2003). Climatic shifts could be extra burdensome on their fat reserves.

Climate change is expected to result in range shifts for several bat species (Loeb and Winters 2013, p. 104). The Indiana bat's maternity range is forecasted to shift from Midwestern U.S. to northeastern U.S. and the Appalachian mountains (Loeb and Winters 2013, p. 108). Similarly, the winter distribution of little brown bats is expected to shift northward based on preferred hibernation temperatures (Humphries et al. 2002, cited in Loeb and Winters 2013, p. 104). This demonstrates not only the sensitivity of bat species to climatic changes, but also the possibility of increased competition among species already in more favorable and stable habitats. The Appalachian Mountains, for example, likely provide numerous climate refugia due to their topographic complexity (Loeb and Winters 2013, p. 110), but these areas may become overcrowded, especially given the loss of many of these habitats due to natural gas and coal extraction operations.

Finally, extreme weather events such as droughts and floods are expected to increase in frequency as a result of climate change which may also impact bat survival (Jones et al. 2009, cited in Loeb and Winters 2013, p. 104; Rodenhouse et al. 2009, p. 250). For bats, water needs increase during pregnancy, so droughts, especially coupled with extreme low or high temperatures, would have negative ramifications for bats' reproductive success (Bourne and Hamilton-Smith 2007, Adams 2010, cited in Loeb and Winters 2013, p. 104).

VII. CONSERVATION RECOMMENDATIONS

The tricolored bat should be expeditiously listed as a Threatened or Endangered species, and critical habitat designated concurrent with listing. Given the grave threat of WNS to the species' survival, it is essential to fully protect all peripheral populations from other threats to their habitat and survival.

The identification and protection of hibernacula and maternity roosts used by tricolored bats should be a priority. Foraging habitat nearby should be included in protected areas. Protection and restoration of riparian zones, late-successional forest stands, and snag habitat is critical. Caves and mines perceived as hazardous to public safety should be gated using bat-appropriate methods so as to preserve and protect important habitat.

Boyles and Willis (2009) suggested two temporary management strategies to preserve WNS-affected bat populations until more permanent solutions are devised or discovered. They had suggested that managing bats' energy budgets may provide an intermediate conservation solution and proposed two ways in which this could be accomplished: (1) Provide source of energy (food) to hibernating bats, and (2) Reduce amount of energy needed to survive hibernation by providing thermal refugia within hibernacula. We would add (3) Provide a source of drinking water to hibernating bats.

While these proposals seem highly impractical as long-term, widely applied solutions, and to our knowledge, have not been implemented in any field location, they could offer a stopgap means of protecting bats, by supporting a limited number of colonies. WNS-infected bats that have been taken to wildlife rehabilitators, and provided warmth, hydration, and food, have survived and recovered, indicating that these proposals could work in a natural site (SCDNR 2011). An advantage of these techniques is that they may pose relatively low risk to cave ecosystems. No other treatments currently being researched, such as vaccines and microbial biocontrol agents, are yet available for widespread application in the field. In addition, some of these other methods involve introduction of organisms not naturally occurring in cave environments, which could cause harm to native microbes or other members of a cave natural community.

We recommend field trials of the above suggestions, to determine whether they could ameliorate mortality caused by WNS until other treatments, potentially more amenable to widespread use, are proved effective, as well as safe for cave environments.

In order to decrease the risk of anthropogenic spread of *Pd*, the following measures should be adopted. Private landowners should be encouraged to close caves, especially priority bat hibernating and roosting sites, to recreational and other non-essential human access. On public lands, cave recreation should be suspended in priority bat caves, at minimum. In addition, cave recreation should be generally discouraged, and where allowed, should be contingent on a registration system that includes education about the threat of WNS and the correct procedures for minimizing risk of *Pd* spread. Reducing recreational use of bat caves will also diminish the amount of human disturbance, a known threat to bat populations.

Detailed protocols for fungal decontamination have been researched and developed (USFWS 2016c). They should be made mandatory by the BLM, Forest Service, National Park Service, and U.S. Fish and Wildlife Service on the federal lands under their jurisdiction. Cave access should be contingent on compliance with decontamination and other disease management measures.

Population monitoring and improved WNS surveillance (Ingersoll et al. 2016, p. 16) should become a priority of the highest order, as an understanding of the temporal and spatial patterns of decline is essential to tracking the progressions of the disease and understanding the behavioral and environmental factors influencing bat susceptibility (Ibid.). Identifying and protecting both high-risk and more stable populations should be part of a comprehensive management strategy, particularly because most hibernacula host multiple species which may not respond synchronously to infection.

Finally, the additional protections conferred by ESA listing are necessary. Wildlife management agencies have no set precedent for managing an epidemic such as WNS, and while solutions are being developed, the protections conferred by Endangered or Threatened status are vital to the persistence of the tricolored bat.

VIII. CRITICAL HABITAT

Federally listed species with designated critical habitat are more likely to make progress toward recovery than species lacking it (Taylor et al. 2005). Critical habitat designation provides the most effective means of assuring that a listed species' habitat is managed to ensure the species' survival and recovery.

Protecting hibernacula and maternity roosts of the tricolored bat from disturbance and degradation is the highest habitat conservation priority for the species. Protecting nearby foraging habitat is also essential. A single hibernaculum should not be considered adequate habitat for local bat populations as changes in climate, hydrology, or other environmental conditions may make certain hibernacula more or less suitable in a given year; the availability of alternative habitat is critical to bat survival (NatureServe 2009). Summer roosting habitat should provide numerous suitable roosts to accommodate roost-switching behavior, and should be located near still water and/or other suitable foraging habitat.

The ESA mandates that when the USFWS lists a species as endangered or threatened, the agency generally must also concurrently designate critical habitat for that species. Section 4(a)(3)(A)(i) of the ESA states that, "to the maximum extent prudent and determinable," the USFWS: shall, concurrently with making a determination . . . that a species is an endangered species or threatened species, designate any habitat of such species which is then considered to be critical habitat . . . . 16 U.S.C. § 1533(a)(3)(A)(i); see also id. at § 1533(b)(6)(C). The ESA defines the term "critical habitat" to mean:

> i. the specific areas within the geographical area occupied by the species, at the time it is listed . . . on which are found those physical or biological features (I) essential to the conservation of the species and (II) which may require special management considerations or protection; and ii. specific areas outside the geographical area

> occupied by the species at the time it is listed . . . upon a determination by the Secretary that such areas are essential for the conservation of the species. Id. at § 1532(5)(A).

Protection of the tricolored bat's roosts will promote the species' conservation, because the tricolored bat, along with most other cave-dwelling bats in North America is at risk, in part, because of human disturbance of roosts, particularly hibernacula and maternity sites. In fact, prior to the onset of WNS, bat biologists saw inadequate cave management as the leading threat to North American hibernating bats. Only with focus on cave protection, which included installation of protective gates and exclusion of human visitors, did some bat colonies begin to recover (Kunz & Pierson 1994).

Biologists recognize that protection of bat roosts and hibernating sites plays a key role in the recovery of bat species (Racey & Entwistle 2003). Indeed, the West Virginia Department of Natural Resources cites protection of critical caves as the reason why Virginia big-eared bats have increased in number since the 1980s (WVDNR 2006). Similarly, protection of hibernacula and roosts of the endangered Ozark big-eared bat, via cave gating, habitat acquisition and easements, is responsible for the increase of this species (Graening et al. 2011). Finally, in an interagency assessment of the status of gray bats, biologists considered protection of important hibernacula to be one of the most crucial steps for recovery of the species (Martin 2007), and in contrast, lack of protection at certain caves continues to be a reason for decline of gray bats at those particular sites (Crozier & Mengak 2012).

Recognizing the crucial importance of protecting hibernacula as critical habitat, the USFWS designated hibernacula and roosting sites for the Indiana bat and Virginia big-eared bat as critical habitat (Clawson 2004, USFWS 1979) in order to safeguard caves and mines, and the listed bats that use them, from purposeful and accidental harm. The Service states:

> Protection of Indiana bat hibernacula has been recognized as a high priority in the species' critical habitat designation (USFWS 1976) and Federal recovery planning documents (USFWS 1983). Consequently, the Service and its state and private cooperators have concentrated their recovery efforts on providing appropriate protection to these sites (USFWS 2007).

Despite all the evidence that protection of bat hibernacula and roost sites is vital to bat survival and recovery, in 2016, the USFWS denied critical habitat designation for the northern long-eared bat, declaring it not prudent because it could "increase the threat to the northern long- eared bat from vandalism and disturbance at hibernacula and could, potentially, increase the spread of WNS" (USFWS 2016c). The Service provided no evidence to back up this assertion. In fact, the agency's rationale flies in the face of USFWS policy regarding other listed bat species, and runs counter to the wealth of evidence that protection of critical hibernacula is key to recovery of listed bats.

In an attempt to justify the denial of critical habitat for the northern long-eared bat on the basis of possible vandalism, the Service lists incidents in which vandals, recreationists, cavers, and others have harmed hibernacula, roosts, and bats themselves. However, the agency does not argue nor attempt to document that these incidents occurred more frequently at caves that were designated as critical habitat or otherwise formally protected, or that critical habitat status led to the incidents of vandalism.

On the contrary, there is abundant evidence that protection of bat roosts and hibernacula promotes bat survival and recovery, and no evidence that it increases harm. Withholding protective status for important hibernacula and roosting sites, and hoping that people with poor intent won't notice them or go in them is not an effective conservation approach. Vandalism and bat disturbance are longstanding, ongoing problems at many caves and bat-occupied mines worldwide (Kunz & Pierson 1994, Martin et al. 2003). The vast majority of these events have occurred in caves that are not designated as critical habitat. Physical barriers and protective policies clearly make a positive difference for bats, but no strategy is foolproof. Trespassers sometimes go to great lengths to breach cave gates. Sometimes no signs, laws, or any amount of common sense will deter determined ne'er do wells.

Based on the evidence, it is reasonable to conclude that while bats in any cave are at risk from trespass and human disturbance, bats in caves lacking protection are at greater risk. Therefore, the responsible course of action is to safeguard the most important bat sites using a variety of conservation tools, including critical habitat status.

In the case of the tricolored bat, the Center for Biological Diversity and Defenders of Wildlife expects that USFWS will comply with the unambiguous mandate of the Endangered Species Act and designate critical habitat concurrently with the listing of the tricolored bat.

IX. CONCLUSION

The tricolored bat faces imminent threats throughout its range and is in danger of extinction or likely to become so in the foreseeable future. White nose syndrome has wiped out bat populations across the eastern United States and continues to spread. The decimation caused by WNS, combined with habitat loss and degradation, environmental contaminants, climate change, industrial wind turbines, and the inadequacy of existing regulatory mechanisms, threatens the tricolored bat with extinction. According to the best available science, the tricolored bat warrants protection under the Endangered Species Act.

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Appendix A: Conservation Status of Tricolored Bats by State

State	Conservation Status	Citation
Alabama	Lowest Conservation Concern	Alabama Department of Conservation and Natural Resources. 2007. Tricolored bat species page. Available at: http://www.outdooralabama.com/tricolored-bat Accessed December 9, 2014.
Arkansas	None	Arkansas Game and Fish Commission. 2013. Arkansas endangered, threatened, and species of special concern. Available at: http://www.agfc.com/licenses/Documents/SpecialConcernSpecies.*Pd*f Accessed December 9, 2014.
Connecticut	None	Connecticut Department of Energy and Environmental Protection. 2010. Endangered, threatened, and special concern mammals. Available at: http://www.ct.gov/deep/cwp/view.asp?a=2702&q=323480&deepNav_GID=1628 Accessed December 9, 2014.
Delaware	Species of Greatest Conservation Need	Delaware Division of Fish and Wildlife. 2015. Species of greatest conservation need wildlife action plan revision. Available at: http://www.dnrec.delaware.gov/fw/dwap/Documents/DE%20SGCN%20List%2010.01.14.*Pd*f Accessed December 9, 2014.
Florida	None	Florida Fish and Wildlife Conservation Commission. 2014. Listed mammals. Available at: http://myfwc.com/wildlifehabitats/imperiled/profiles/mammals/ Accessed December 9, 2014.
Georgia	None	Georgia Wildlife Resources Division. 2014. Rare species profiles. Available at: http://www.georgiawildlife.org/rare_species_profiles Accessed December 9, 2014.
Illinois	None	Illinois Department of Natural Resources. 2011. Illinois endangered species protection board. Available at: http://www.dnr.illinois.gov/ESPB/Documents/ETChecklist2011.*Pd*f Accessed December 9, 2014.
Indiana	Species of Special Concern	Indiana Department of Natural Resources. 2013. Mammals of Indiana. Available at: http://www.in.gov/dnr/fishwild/files/fw-Mammals_Of_Indiana.*Pd*f Accessed December 9, 2014.
Iowa	None	Iowa Department of Natural Resources. 2012. Iowa species of greatest conservation need. Available at: http://www.iowadnr.gov/Environment/WildlifeStewardship/IowaWildlifeActionPlan.aspx Accessed December 9, 2014.
Kansas	None	Kansas Department of Wildlife, Parks, and Tourism. 2005. Comprehensive Wildlife Conservation Plan. Available at: http://kdwpt.state.ks.us/Services/Kansas-CWCP/Kansas-CWCP Accessed December 9, 2014.
Kentucky	None	Kentucky Department of Fish and Wildlife Resources. 2014. Species of greatest conservation need. Available at: http://fw.ky.gov/Wildlife/Documents/1.1%20ListSpeciesForTable.*Pd*f Accessed December 9, 2014.
Louisiana	None	Louisiana Department of Wildlife and Fisheries. 2005. Species of Conservation Concern. Available at: http://www.wlf.louisiana.gov/sites/default/files/*Pd*f/document/32917-appendix-f-species-conservation-concern-louisiana/24_appendix_f--species_of_conservation_concern.*Pd*f Accessed December 9, 2014.
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Maryland	None	Maryland Department of Natural Resources. 2005. Wildlife Diversity Conservation Plan. Available at: http://www.dnr.maryland.gov/wildlife/Plants_Wildlife/WLDP/*Pd*fs/WCDP_Chapter3_20050926.*Pd*f Accessed December 9, 2014.
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Michigan	Species of Special Concern	Michigan Department of Natural Resources. 2014. Wildlife action plan. Available at: http://www.michigan.gov/documents/dnr/mammals_326070_7.*Pd*f Accessed December 10, 2014.
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Missouri	None	Missouri Department of Conservation. 2014. Missouri animals of conservation concern. Available at: http://mdc.mo.gov/sites/default/files/resources/2010/04/4068_1693.*Pdf* Accessed December 10, 2014.
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New Hampshire	None	New Hampshire Fish and Game. 2011. Species profile Eastern pipistrelle. Available at: http://www.wildnh.com/Wildlife/Wildlife_Plan/WAP_species_*PD*Fs/Mammals/EasternPipistre.*Pdf* Accessed December 9, 2014.
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New York	None	New York State Department of Environmental Conservation. 2014. List of endangered, threatened and special concern fish and wildlife species of New York state. Available at: http://www.dec.ny.gov/animals/7494.html Accessed December 10, 2014.
North Carolina	None	North Carolina Wildlife Resources Commission. 2014. Protected wildlife species of North Carolina. Available at: http://www.ncwildlife.org/Portals/0/Conserving/documents/protected_species.*Pdf* Accessed December 10, 2014.
Ohio	Species of Concern	Ohio Department of Natural Resources. 2012. Species guide index. Available at: http://wildlife.ohiodnr.gov/species-and-habitats/species-guide-index/mammals/tricolored-bat Accessed December 10, 2014.
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Pennsylvania	Proposed Endangered	Pennsylvania Game Commission. 2013. Annual report. Available at: http://www.portal.state.pa.us/portal/server.pt/document/1396620/2013_pgc_annual_legislative_report_*Pdf*?qid=96114191&rank=1 Accessed December 10, 2014.
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Tennessee	None	Tennessee Wildlife Resources Agency. 2005. State wildlife action plan. Available at: http://www.tn.gov/twra/cwcs/tncwcs2005app.*Pdf* Accessed December 10, 2014.
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Virginia	None	Virginia Department of Game and Inland Fisheries. 2010. Virginia wildlife action plan. Available at: http://www.bewildvirginia.org/species/mammals.*Pdf* Accessed December 10, 2014.
West Virginia	None	West Virginia Division of Natural Resources. 2012. Rare, threatened, and endangered species in West Virginia. Available at: http://www.wvdnr.gov/Wildlife/*PD*FFiles/RTE_Animals_2012.*Pdf* Accessed December 10, 2014.

Wisconsin	Threatened	Wisconsin Department of Natural Resources. 2011. Wisconsin natural heritage working list rare mammals. Available at: http://dnr.wi.gov/topic/endangeredresources/animals.asp?mode=list&grp=17 Accessed December 10, 2014.
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