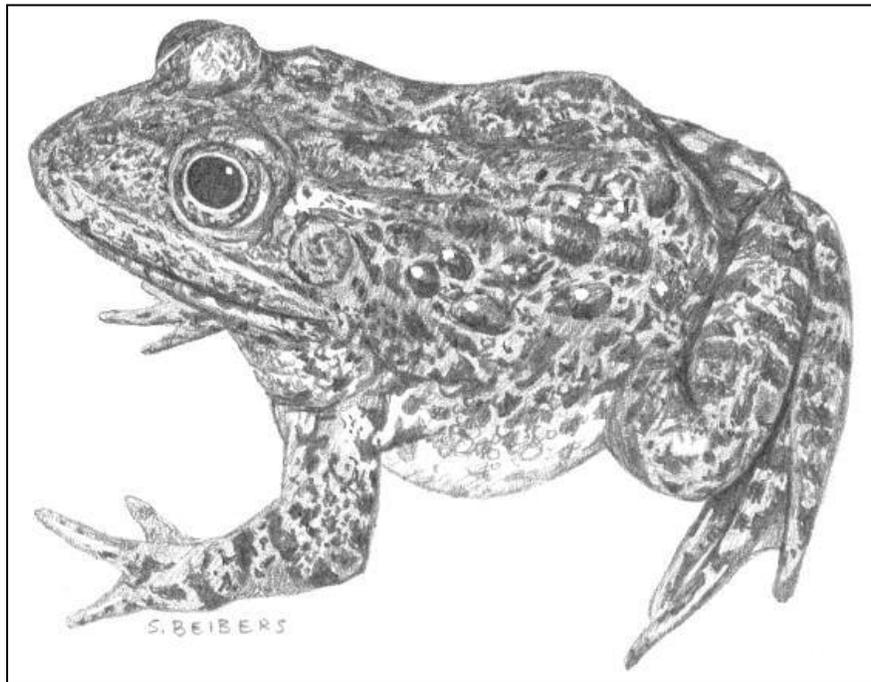


**DUSKY GOPHER FROG**  
*(Rana sevosa)*

**RECOVERY PLAN**



**DUSKY GOPHER FROG**  
**(*Rana sevosa*)**

**Recovery Plan**

Prepared by

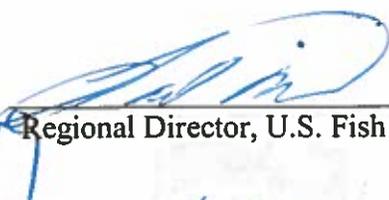
Dusky Gopher Frog Recovery Team

and

Mississippi Field Office  
U.S. Fish and Wildlife Service

for

U.S. Fish and Wildlife Service  
Southeast Region  
Atlanta, Georgia

Approved: 

Regional Director, U.S. Fish and Wildlife Service

Date: 7/23/15

Recovery plans delineate reasonable actions which are believed to be required to recover and/or protect listed species. Plans are published by the U.S. Fish and Wildlife Service (USFWS), and are sometimes prepared with the assistance of recovery teams, contractors, state agencies, and other affected and interested parties. Plans are reviewed by the public and submitted to additional peer review before they are adopted by the USFWS. Objectives of the plan will be attained and any necessary funds made available contingent upon budgetary and other constraints affecting the parties involved, as well as the need to address other priorities. Recovery plans do not obligate other parties to undertake specific tasks and may not necessarily represent the views nor the official positions or approval of any individuals or agencies involved in developing the plan other than the USFWS. Recovery plans represent the official position of the USFWS only after they have been signed by the Regional Director as approved. Approved recovery plans are subject to modification as dictated by new findings, changes in species status, and the completion of recovery tasks.

By approving this recovery plan, the Regional Director certifies that the data used in its development represent the best scientific and commercial information available at the time it was written. Copies of all documents reviewed in the development of the plan are available in the administrative record, located at the Mississippi Field Office in Jackson, Mississippi.

**Literature citation of this document should read as follows:**

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Recovery Plans can be downloaded from the U.S. Fish and Wildlife Service website:  
<http://www.fws.gov/endangered/species/recovery-plans.html>

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## EXECUTIVE SUMMARY

**Current Status:** The dusky gopher frog (*Rana sevosa*) is listed as an endangered species under the Endangered Species Act of 1973, as amended (66 FR 62993). Its current distribution is restricted to the state of Mississippi. At the time of listing, only one population of the species was known. Subsequently, two other naturally-occurring populations were discovered. One additional dusky gopher frog population has been established in Mississippi as a result of translocation experiments. Historical records exist for Alabama and Louisiana, but currently no populations are known from these two states. Presently, we estimate that a minimum of 135 individual adult frogs survive in the wild, the vast majority of which occur in the original population known at the time of listing.

Approximately 6,477 acres (ac) (2,621 hectares (ha)) in Louisiana and Mississippi have been designated as critical habitat for the dusky gopher frog (77 FR 35117). The critical habitat is located in St. Tammany Parish, Louisiana, and Forrest, Harrison, Jackson, and Perry Counties, Mississippi. The dusky gopher frog has a recovery priority number of 5 which indicates that the species faces a high degree of threat, but has a low recovery potential. The states of Louisiana and Mississippi have listed the dusky gopher frog as an endangered species that cannot be killed or removed from the wild without a permit. Although not listed as threatened or endangered by the state of Alabama, the dusky gopher frog is listed among those non-game species that cannot be killed or removed from the wild without a permit.

**Habitat Requirements and Limiting Factors:** Optimal post-larval dusky gopher frog habitat consists of uplands dominated by fire-maintained longleaf pine (*Pinus palustris*) with a grassy understory. Larval habitat consists of grassy, acidic, isolated, ephemeral, depressional wetlands that lack predaceous fish. Principal threats to the dusky gopher frog include degradation and destruction of breeding and non-breeding habitat, habitat fragmentation, and alteration of hydrological patterns due to urbanization and climate change. Additional threats include the restricted range of the dusky gopher frog, its small number of populations, and disease. All these factors act to increase the vulnerability of the species to a single catastrophic event and to the deleterious effects of genetic inbreeding.

**Recovery Strategy:** The recovery strategy for the dusky gopher frog consists of maintaining and enhancing existing populations on tracts of public and private land; monitoring the status of existing populations; identifying and securing additional dusky gopher frog populations and habitat; establishing new populations through translocations or reintroductions; and supporting research that guides land management and provides demographic and ecological data. Management plans should be developed and implemented for all sites where the dusky gopher frog occurs. Appropriate habitat management includes minimizing soil disturbance and loss of native herbaceous groundcover vegetation; conducting prescribed burning, particularly during the growing season; maintaining open-canopied, grassy wetlands; and restoring degraded upland habitat. In addition, management plans should specifically address habitat modifications (e.g.,

filling of drainage ditches and plow lines, restoring native groundcover flora) necessary to improve and maintain appropriate habitat.

Monitoring programs to track population trends and the response of this species to habitat management activities are needed for all populations. Monitoring programs should be evaluated and revised as needed. Since recovery of the dusky gopher frog will necessitate finding or creating new, currently unknown populations, assessment of potentially suitable habitat within the range of the frog and additional presence/absence surveys are needed, especially in Alabama and Louisiana. If no additional dusky gopher frog populations are found, suitable habitat for translocations/reintroductions needs to be identified, and programs developed and implemented to establish and monitor these new populations and manage the habitat that supports them. We expect to conduct a Species Status Assessment (SSA) for the dusky gopher frog in the future and will make revisions to the recovery plan accordingly.

**Recovery Objective:** The primary objective of this recovery plan is to prevent the extinction of the dusky gopher frog. The long-term recovery objective is to downlist the dusky gopher frog from endangered to threatened. Delisting is not currently foreseeable due to the extreme curtailment of range; current low number of individuals and populations; and magnitude of threats.

**Recovery Criteria:** This recovery plan sets forth criteria which, when met, will increase the range of extant dusky gopher frog populations; will increase the number of individuals and populations; and will reduce threats to the species' existence. We believe downlisting may be considered when the following criteria are met:

- 1) Six **viable metapopulations**\* are documented within blocks of recovery focus areas (described in Section II of this recovery plan) and are widely distributed across the range of the species. The six metapopulations would include a minimum of 12 breeding ponds distributed within the species historic range:
  - a) One metapopulation in Block #1 (Louisiana. Portions of St. Tammany, Tangipahoa, and Washington Parishes, west to the Tangipahoa River);
  - b) Two metapopulations each in Block #2 (South-Central Mississippi. North of State Hwy. 26, between the Pearl and Pascagoula Rivers; Forrest County and portions of George, Greene, Jones, Lamar, Marion, Pearl River, Perry, Stone, and Wayne Counties) and Block #3 (South Mississippi. South of Hwy. 26, between the Pearl and Pascagoula Rivers; Hancock and Harrison Counties, and portions of Jackson, George, Pearl River, and Stone Counties); and
  - c) One metapopulation in either Block #4 (Eastern Mississippi. East of Pascagoula/Leaf Rivers; portions of George, Greene, Jackson, Perry, and Wayne Counties) or Block #5 (Alabama. West of the Mobile River Delta; Mobile and Washington Counties, and a small portion of Choctaw County).
- 2) Long-term monitoring (at least 10 years) of each metapopulation documents population viability (viability standard to be defined through a recovery task). The 10-year timeframe will allow monitoring recruitment events and other population attributes in a

species that has been characterized by highly variable reproductive and survival rates. In each of at least two annual breeding events within a three-year period, a total of 30 egg masses per metapopulation must be documented and natural recruitment must be verified.

- 3) Breeding and adjacent upland habitats within the six metapopulations are protected long-term through management agreements, public ownership, or other means, in sufficient quantity and quality (to be determined by recovery task) to support growing populations.
- 4) Studies of the dusky gopher frog's biological and ecological requirements have been completed and measures necessary for recovery discovered during these studies are being implemented and are showing progress.

\* A **viable** population is one that is large enough to maintain sufficient genetic variation to enable it to evolve and respond to natural habitat and environmental changes, and exhibits parameters consistent with a stable or increasing reproductive rate, without the addition of frogs raised in artificial environments or introduced from other populations. Viable populations generally consist of multiple age classes of individuals, including newly recruited juveniles. In addition, a dusky gopher frog population should be supported by habitat containing breeding ponds and their surrounding uplands. To be a viable population, a dusky gopher frog population must be a **metapopulation**. Two or more breeding ponds within dispersal distance of one another function as a metapopulation; if breeding conditions for the species are not met by certain ponds in one or more years, the species may persist by breeding at other nearby ponds. For the purposes of this recovery plan, a dusky gopher frog metapopulation will be defined as two or more occupied breeding ponds, individually separated from each other by a mile (1.6 kilometer) or less, within a contiguous area of suitable habitat with no major barriers to dispersal (e.g., major highways, rivers, developed areas, etc.) between ponds.

Criteria may be reevaluated as new information becomes available.

#### **Actions Needed:**

1. Protect existing wild dusky gopher frog populations through habitat restoration, management and other conservation techniques.
2. Monitor dusky gopher frog populations and their habitat.
3. Continue searches for additional dusky gopher frog populations.
4. Conduct a population and habitat viability analysis (PHVA) and develop the necessary supporting research.
5. Formulate and implement guidelines for using translocations to establish dusky gopher frog populations.
6. Revise and implement a controlled propagation and reintroduction plan to facilitate use of captive dusky gopher frogs in translocation efforts.
7. Develop and distribute public educational and informational materials/programs to solicit and promote voluntary stewardship.
8. Review and evaluate recovery progress using the SSA framework (see <http://sites.google.com/a/fws.gov/ssa/?pli=1>).

**Estimated Cost of Recovery:** The implementation of recovery tasks, from which cost estimates can be made over a 5-year period of recovery effort, will total approximately \$3,615,000.

Year	Task 1	Task 2	Task 3	Task 4	Task 5	Task 6	Task 7	Task 8	Total Cost
FY 1	100K	260K	20K	280K	100K	50K	5K	0	815K
FY 2	100K	260K	20K	275K	100K	30K	5K	0	790K
FY 3	100K	245K	90K	280K	100K	30K	0	0	845K
FY 4	100K	280K	40K	100K	75K	0	0	0	595K
FY 5	100K	280K	40K	75K	75K	0	0	0	570K
<b>Total</b>	500K	1,325K	210K	1,010K	450K	110K	10K	0	3,615K

**Date of Recovery:** The estimated date for full recovery is undeterminable at this time.

**Recovery Milestones:**

1. Development of habitat restoration and management techniques to protect dusky gopher frog habitat.
2. Completion of surveys throughout the historical range of the dusky gopher frog for potential suitable habitat and additional populations.
3. Initiation of a translocation/reintroduction program.
4. Completion of a PHVA for the Glen’s Pond population.
5. Evaluation of the effectiveness of the translocation/reintroduction program.
6. Selection of additional reintroduction sites, if needed, and development and implementation of an updated release program.
7. Revision of the Dusky Gopher Frog Recovery Plan using the SSA framework to evaluate new information for determining updated actions, costs, and criteria for delisting.

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# I. INTRODUCTION

## STATUS

The U.S. Fish and Wildlife Service (USFWS) listed the Mississippi gopher frog (*Rana capito sevosa*) under the Endangered Species Act (Act), without critical habitat, as an endangered distinct vertebrate population segment (DPS) of the gopher frog (*Rana capito*) on December 4, 2001(66 FR 62993; USFWS 2001). At the time of listing, the frog was given a recovery priority number of 6 (48 FR 43098), indicating a high degree of threat, a low potential of recovery, and a taxonomic classification as a subspecies. In Louisiana and Mississippi, the Mississippi (dusky) gopher frog is on each state's list of Endangered Species. The Louisiana list of endangered species contains those species that cannot be killed or removed from the wild without a permit (Title 56 of the Louisiana Revised Statutes, 2013). In Mississippi, the dusky gopher frog is protected under the Nongame and Endangered Species Conservation Act of 1974 which prohibits taking, possessing, transporting, exporting, offering to sell, or offering to ship endangered species. Although not listed as threatened or endangered by the state of Alabama, the dusky gopher frog is listed among those non-game species for which it is "unlawful to take, capture, kill, or attempt to take, capture or kill; possess, sell, trade for anything of monetary value, or offer to sell or trade for anything of monetary value" (Alabama Department of Conservation and Natural Resources (ADCNR) 2015).

On June 12, 2012, USFWS published a final rule (77 FR 35118; USFWS 2012) designating critical habitat for this listed entity and changing its status to "species" and its name to the dusky gopher frog (*Rana sevosa*) based on taxonomic changes and the acceptance of these changes by the herpetological scientific community. The recovery priority number has been changed to 5 reflecting this taxonomic change; however the high degree of threat and low potential of recovery are unchanged. A total of approximately 6,477 acres (ac) (2,621 hectares (ha)) in Louisiana and Mississippi, encompassing 12 units, were designated as critical habitat for the dusky gopher frog. In Mississippi, seven critical habitat units are protected on the DeSoto National Forest; one is protected on property owned by the U.S. Army Corps of Engineers; one on property owned by the state of Mississippi; and two are on private property owned by The Nature Conservancy (TNC), a nongovernmental, nonprofit organization dedicated to land and water conservation. The remaining unit is on private property in Louisiana.

Historic records for the dusky gopher frog (as currently described) exist for sites in St. Tammany Parish, Louisiana; Forrest, Greene, Hancock, Harrison, Jackson, Pearl River, and Perry Counties in Mississippi; and Mobile County, Alabama (Allen 1932, Netting and Goin 1942, Smith and List 1955, Neill 1957, Volpe 1957, Crawford 1988, Dundee and Rossman 1989, HerpNet 2013). Researchers conducting numerous surveys have been unable to document the continued existence of the dusky gopher frog in Louisiana (Seigel and Doody 1992, Thomas 1996, Leonard *et al.* 2003, Pechmann *et al.* 2006) or in Alabama (Bailey 1994, 2009; Hart 2004). The last documented observation of a gopher frog in Louisiana was in the mid-1960s (Landry 2011). In Alabama, it was last seen in 1922 (Bailey 1994). Since 2005, only two naturally-occurring dusky gopher frog populations have been verified through documented breeding. These populations occur in Harrison and Jackson Counties, Mississippi. In addition, a population of dusky gopher frogs has been established at property owned by TNC in Jackson County,

Mississippi. We have been monitoring all three of these populations closely. Habitat loss and degradation is the primary factor in the loss of historical dusky gopher frog populations in Alabama, Louisiana, and Mississippi. Nevertheless, we continue to search for potential breeding habitat within the historically longleaf pine-dominated forests in these three states that may harbor a hidden population, or could be restored to support a translocated population.

## SPECIES DESCRIPTION AND TAXONOMY

### Description

The dusky gopher frog has a stubby appearance due to its short, plump body, comparatively large head, and relatively short legs (Conant and Collins 1991). The coloration of its back varies in individual frogs. It ranges from an almost uniform black to a pattern of reddish brown or dark brown spots on a ground color of dark gray or brown (Goin and Netting 1940). Warts densely cover the back. The belly is thickly covered with dark spots and dusky markings from chin to mid-body (Goin and Netting 1940, Conant and Collins 1991). Males are distinguished from females by their smaller size, nuptial pad (swollen area that assists grip during breeding) on their thumbs, and paired vocal sacs on either side of the throat (Goin and Netting 1940). Richter (1998) reported mean snout-vent lengths from three years of data from dusky gopher frogs at Glen's Pond. Measurements ranged from 2.5 to 2.8 inches (in) (63.2 to 70.2 millimeters (mm)) for males and 3.1 to 3.3 in (78.0 to 82.7 mm) for females. Dusky gopher frog tadpoles are similar to those of other gopher frogs and crawfish frogs (*R. areolata*) (Volpe 1957, Altig *et al.* 2001).



Figure 1. Adult male dusky gopher frog.  
Photo by John Tupy

### Taxonomy and Nomenclature

Gopher frogs (*Rana capito* and *R. sevosia*) are members of the large family, Ranidae ("true frogs"), which has a worldwide distribution. The genus *Rana* is the only North American representative of this family. The range of the dusky gopher frog includes those parts of the lower coastal plain extending from southeastern Louisiana (includes three of the "Florida Parishes"), across the southern Mississippi coastal counties, to the

Mobile River delta in Alabama. Goin and Netting (1940) originally described frogs from this geographic range as a distinct species of gopher frog (*R. sevosia*). The taxonomic history of gopher frogs is complex (summary in Altig and Lohofener 1983). Subsequent to the original description by Goin and Netting, frogs of this population segment were considered subspecies of the gopher frog, *R. capito*, (*R. c. sevosia*, common name dusky gopher frog) (Wright and Wright 1942), a distinct species (*R. sevosia*) (Wright and Wright 1949), and later, subspecies of the crawfish frog (*R. a. sevosia*) (Viosca 1949, Neill 1957). In 1991, Collins challenged the taxonomic arrangement that lumped crawfish frogs and gopher frogs together as one species and recommended their separation based on biogeographical grounds. This arrangement was followed by Conant and Collins (1991), who again recognized the name *R. c. sevosia*.

Shortly after the USFWS listed the frog, Young and Crother's (2001) paper was published describing the first comprehensive biochemical analysis of the relationships between gopher frogs and crawfish frogs and among subspecies of gopher frogs. They used allozyme electrophoresis (an assay of gene products) to examine allelic differences between and among populations. Allozyme data have been used extensively to investigate the evolution of genetic relationships among related species. Young and Crother (2001) analyzed tissue from gopher frogs across the range of the species including populations in Mississippi, Alabama, Georgia, Florida, and North Carolina, and from crayfish frogs from Arkansas, Kansas, and Missouri. They found strong support for the species designations *R. areolata* (crayfish frogs) and *R. capito* (gopher frogs). In addition, they found that the population of gopher frogs from Harrison County, Mississippi, showed a fixed difference at a single locus (site for a specific gene on a chromosome) from all other gopher frogs east of the Mobile River drainage in Alabama. This difference is considered by many taxonomists that support the phylogenetic (evolutionary) species concept (PSC) to be significant enough to warrant elevation of the frog to its own species (Young and Crother 2001). No other specific taxonomic divisions were determined among the remaining populations of gopher frogs sampled. Since Harrison County is within the range of the original specimens used to describe *R. sevosa*, Young and Crother (2001) recommended the resurrection of *R. sevosa* as a distinct species. The Standard English common name for *R. sevosa* is dusky gopher frog (Crother 2012).

Frost *et al.* (2006) proposed removing the genus name *Rana* from a group of North American frogs (one of which is the dusky gopher frog) and replacing it with the genus *Lithobates*. There is still disagreement within the scientific community about accepting this change (Hillis 2007, Pauly *et al.* 2009, Wiens *et al.* 2009, Pyron and Wiens 2011, Crother 2012). For that reason, we will continue to use the scientific name of *Rana sevosa* for the dusky gopher frog.

## **POPULATION TRENDS AND DISTRIBUTION**

### **Alabama**

The only dusky gopher frog (as currently described) record from Alabama was an observation by Löding in 1922, and summarized in Wright and Wright (1949). No breeding sites for the dusky gopher frog are known from Alabama. The Löding record was of three gopher frogs under drift logs on the beach of Mobile Bay just south of the mouth of Dog River, Mobile County, Alabama. Bailey (1994) visited this area in 1993, and found it to be a residential development, although large longleaf pine trees in lawns and vacant lots indicated the area could have formerly been suitable upland habitat for gopher frogs. Neither Löding nor Bailey located a possible breeding site in the vicinity of the record. Researchers have conducted two studies in southwestern Alabama to look for habitat that could support dusky gopher frogs. Hart (2004) initiated a remote sensing study using aerial photography of Mobile and Washington Counties, Alabama, to find open, isolated ponds in proximity to forested terrain. This technique was used to identify sites with the potential for supporting dusky gopher frog populations. Hart (2004) conducted field assessments of 41 ponds in Mobile County, Alabama, but habitat quality at these ponds was limited. Ponds were overgrown with woody vegetation and lacked the emergent vegetation necessary for dusky gopher frog egg attachment (Hart 2004). Additional ponds were identified remotely in Washington County, Alabama, but were not visited, and their habitat quality is unknown. Bailey (2009) used a similar remote sensing technique to locate a total of 21 ponds in

Choctaw, Mobile, and Washington Counties, Alabama. However, this was a coarse filter approach, and field assessments were not possible due to drought conditions and inaccessibility resulting from site isolation. Thus, no areas suitable for conservation of the dusky gopher frog were positively identified in either of the remote sensing studies. No dusky gopher frog populations in Alabama were discovered during field assessments associated with Hart's (2004) study. Shaver (2013) has begun a GIS habitat assessment of the localities in private ownership identified by Hart (2004) and Bailey (2009). He is contacting landowners with potential habitat on their property and identifying those willing to allow access for habitat assessments and surveys, but thus far no dusky gopher frogs have been found. In fall 2012, habitat assessments were initiated by dusky gopher frog researchers and biologists with the USFWS and Alabama Department of Conservation and Natural Resources on three longleaf pine tracts owned and actively managed by Mobile County Environmental Division and Mobile County Board of Education. Automated frog call recording devices were deployed at three wetland sites on these properties during the 2012-2013 breeding season and data collection has continued in 2015; however no dusky gopher frog calls have yet been detected. Further site evaluations to assess habitat suitability at these and other sites will continue.

### **Louisiana**

In the 1950s, the dusky gopher frog was observed and studied at several localities in St. Tammany Parish (Volpe 1957). The last observation of the dusky gopher frog in Louisiana was 1965. However, by the late 1980s, Dundee and Rossman (1989) believed the future of the frog in Louisiana was bleak due to habitat conversion of native habitat to agriculture, estates, and tree farms. In March 2011, dusky gopher frog researchers, biologists with the Louisiana Department of Wildlife and Fisheries, and a biologist with the USFWS visited the area of historic dusky gopher frog occurrence in St. Tammany Parish and conducted a habitat assessment. The area is managed for timber by a company conducting industrial forestry. This same area was surveyed for gopher frogs in the 1990s and 2000s. During those visits, several ephemeral ponds were considered similar in appearance (water clarity, depth, vegetation) to ponds in Mississippi used for breeding by the dusky gopher frog (Thomas and Ballew 1997, Leonard *et al.* 2003, Pechmann *et al.* 2006). Our observations in 2011 indicated the Louisiana ponds were little changed from the descriptions provided by the previous surveyors. In addition, five of the high quality ponds are in close proximity to each other, which would allow movement of adult gopher frogs between them. In fact, no group of five ponds such as these was found in any of the areas of historical occurrence that have been searched in Mississippi. Maintaining the five ponds as suitable habitat into which dusky gopher frogs could be translocated is essential to decrease the risk of extinction of the species resulting from stochastic events and to provide for the species' eventual recovery. These five ponds were included in the final designation of critical habitat for the species (USFWS 2012).

### **Mississippi**

Allen (1932) found gopher frogs to be common in the coastal counties of Mississippi early in the 20<sup>th</sup> century; however, between this time and the early 1980s, very little information is available on the status of the species. In 1987 and 1988, Crawford (1988) surveyed 42 ponds in six Mississippi counties for the dusky gopher frog. During his attempts to find all of the state's historical localities for the gopher frog, he found that habitat in the vicinity of most localities had been altered by conversion of natural forest to agriculture, pine plantations, and urban areas. In

addition, the character of remaining historical breeding ponds had been changed from open-canopy, temporary ponds with clear water and hard bottoms to muddy, more permanent closed canopy ponds (G. Johnson, U.S. Forest Service, pers. comm. 1999). No appropriate habitat for the dusky gopher frog could be found near any of the historical localities (G. Johnson, pers. comm. 1999). Nevertheless, during his study Crawford discovered a new breeding pond on the DeSoto National Forest (DNF), Harrison County, Mississippi. In the period between this discovery in 1988 and 2004, this site, named Glen's Pond, supported the only known population of dusky gopher frogs. Glen's Pond has been monitored continuously since its discovery. Between 1996 and 2007, years of natural recruitment at this site were limited to 1997, 1998, and 2003 (only three metamorphs) due to inadequate rainfall or pond drying prior to tadpole metamorphosis. However, in the ensuing years, natural recruitment has occurred in 2008, 2010, 2012, 2013, and 2014. At each breeding event since 2002, a portion of the available egg masses have been collected and the hatchlings reared to metamorphosis in outdoor tanks for release at Glen's Pond. In addition, in 2001 and 2005, water from an onsite well was added to Glen's Pond to prevent it from drying. These interventions resulted in metamorphic recruitment during the 9-year period between 1998 and 2007 when there was virtually no natural recruitment, and supplemented natural recruitment in other years (Sisson 2003, 2005; Pechmann *et al.* 2012). Prior to their release into the wild, metamorphic frogs raised in cattle tanks received dye marks for several years, but currently receive visible implanted alphanumeric (VIA) tags. The predominance of adult frogs with dye marks that entered Glen's Pond to breed in 2007, at the end of the "drought period" of extremely limited natural recruitment, indicated that raising frogs in tanks is a successful technique (Baxley and Qualls 2007). Mark-recapture and demographic analyses suggest that human recruitment assistance rescued the population from likely extinction and helped maintain the population size (Pechmann *et al.* 2012). This assessment is supported by an examination of the average number of breeding adults per breeding year during the two periods before and after the drought period which was, respectively, 91 and 96 (Richter *et al.* 2003, Pechmann *et al.* 2012). On-going monitoring at Glen's Pond indicates this population is continuing to increase as natural recruitment has increased in conjunction with wetter years and a longer pond hydroperiod. In 2014, 135 adult dusky gopher frogs were captured at Glen's Pond (Pechmann and Tupy 2014). As previous monitoring has indicated that all adult dusky gopher frogs breed each year that the pond fills to a sufficient depth (Pechmann *et al.* 2012), this number provides an approximate estimate of the current size of the Glen's Pond population.

In 2013, dusky gopher frogs dispersed 0.8 miles (mi) (1.3 kilometers (km)) from Glen's Pond to a restored pond named Pony Ranch Pond where they had never been observed previously (See further discussion under: **Management through Partnerships**). Seven individual adult frogs were captured using a temporary drift fence, three egg masses were found, and 18 metamorphosed juvenile dusky gopher frogs were documented (Pechmann and Tupy 2013). During the 2014 breeding season, 16 adult dusky gopher frogs were captured at Pony Ranch Pond and three egg masses were found. Unfortunately, survival of natural metamorphic dusky frogs was low at Pony Ranch Pond in 2014 and may have been influenced by the documented continued presence of an unnamed disease in the population (Pechmann and Tupy 2014) (See further discussion under: **REASONS FOR LISTING AND ONGOING THREATS ASSESSMENT: Disease or Predation**).



Figure 2. Cattle watering tanks used to raise dusky gopher frog tadpoles to metamorphosis. Photo by John Tupy

In 2004, dusky gopher frogs were found at two additional sites, Mike's Pond and McCoy's Pond, in Jackson County, Mississippi. Mike's Pond is approximately 20 mi (32 km) east of Glen's Pond and separated from it by the Tchoutacabouffa River drainage. Mike's Pond supports a very small breeding population. Breeding at Mike's Pond has been verified in only four years (2004, 2005, 2010, 2012) since it was discovered, although several male dusky gopher frogs have been heard calling as recently as 2013 (Lee 2013). The breeding in 2010 was the result of two Glen's Pond females being introduced into Mike's Pond to breed with the two males heard calling there; two egg masses resulted from this event (See further discussion below under: **Management through Partnerships**). McCoy's Pond is east of Mike's Pond by approximately 16 mi (25 km) and separated from it by the Pascagoula River drainage. No dusky gopher frogs have been observed at this site since a frog was heard calling there in 2004; the pond has not held water long enough in most years to support natural population recruitment.

Efforts to locate new dusky gopher frog populations continue within the historical distribution of the frog in Alabama, Louisiana, and Mississippi. However, available habitat is limited, and the USFWS and partners have shifted focus to finding habitat that can be restored and used as translocation sites to establish populations. Since 2004, eggs have been removed from the Glen's Pond population, and tadpoles and metamorphic dusky gopher frogs have been raised in cattle tanks and released in Jackson County, Mississippi, at a pond (TNC Pond 1) on a site managed by TNC (Old Fort Bayou Mitigation Bank). Survival to adulthood of at least some of the released frogs has been documented; calling male dusky gopher frogs have been heard, a

single dusky gopher frog egg mass was observed in two different years, and five egg masses were found in 2014 (Lee 2014). The number of breeding adults in this population is unknown.

To summarize, since the dusky gopher frog was listed as an endangered species in 2001, three naturally-occurring populations supported by four breeding ponds have been documented. The four ponds are Glen's Pond, Pony Ranch Pond, Mike's Pond, and McCoy's Pond [See Figure 3]. A fourth population, breeding at TNC Pond 1 [See Figure 3], has been established through translocation of Glen's Pond frogs. The Glen's Pond population, supported by the Glen's Pond and Pony Ranch Pond breeding sites, is the only population that is considered stable at this time. We have restored an additional 11 ponds on the DNF, Ward Bayou Wildlife Management Area (WBWMA) (owned by the U.S. Army Corps of Engineers), and TNC property. Two additional ponds have been created; one on DNF and one on WBWMA. We hope these 13 ponds (all designated as critical habitat) may eventually be used as translocation sites. In addition, we designated critical habitat at a site in Louisiana which contains two historical dusky gopher frog breeding ponds. The frog does not currently exist at this privately-owned site. We continue to survey areas within the historical range of the frog and hope to discover currently unknown populations or at least habitat that could be restored and used to establish populations.

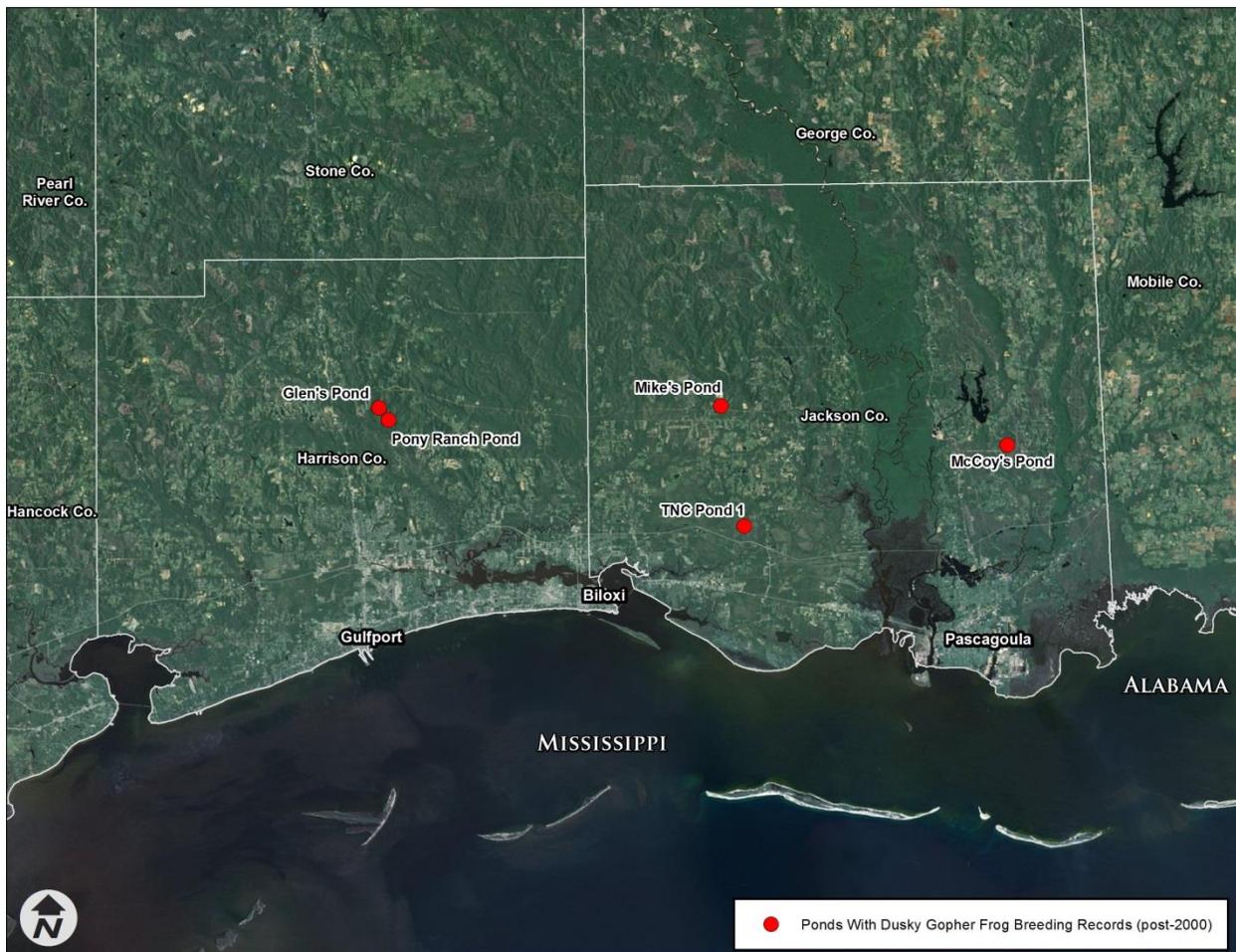


Figure 3. Ponds with dusky gopher frog breeding records, post-2000. [Glen's Pond, Pony Ranch Pond, Mike's Pond, TNC Pond 1, and McCoy's Pond]

## **LIFE HISTORY AND ECOLOGY**

Dusky gopher frogs are amphibians with a complex life cycle that consists of aquatic eggs/larvae and terrestrial adults. Adult dusky gopher frogs spend most of their lives underground in forested habitat consisting of fire-maintained, open-canopied woodlands historically dominated by longleaf pine (*Pinus palustris*) with an understory of grasses such as little bluestem (*Schizachyrium scoparium*). During the breeding season, dusky gopher frogs leave their subterranean retreats in the uplands and migrate to their breeding sites during rains associated with passing cold fronts (Young 1997). Both forested uplands and isolated wetland breeding sites are needed to provide space for normal behavior and both individual and population growth.

Although breeding typically occurs from December through March, reproduction has been documented in all months except May, June and July. Late summer and autumn breeding has occurred after heavy rains from tropical depressions and hurricanes in August, September and October (Seigel and Kennedy 1999, Thurgate and Pechmann 2007, Pechmann and Tupy 2012). Male dusky gopher frogs move to breeding ponds before females and begin calling (Richter and Seigel 2002); however, males may call below water and calls may be difficult to detect (Dundee and Rossman 1989, Jensen *et al.* 1995). Females typically arrive at the pond, breed, deposit their eggs as a single clutch on emergent herbaceous vegetation (Goin and Netting 1940, Dundee and Rossman 1989, Young *et al.* 1995, Richter and Seigel 2002, Richter *et al.* 2003), and leave the pond; males generally remain at the pond longer. Using data collected from 2002 through 2007 from 113 marked frogs, Jones (2008) determined that the mean time spent in the pond basin was 8.97 days for females and 16.88 days for males. Egg masses can be distinguished from the very similar leopard frog egg masses due to their attachment to emergent vegetation; their firm, globular nature; and darker color. The number of eggs per egg mass ranges from 500 to 2,800 in Mississippi (Richter and Seigel 1997, 1998; Young 1997, Richter 1998), to 3,000 to 7,000 in Louisiana (Volpe 1957, Dundee and Rossman 1989). As clutch size is related to body size in most amphibians, first time breeders likely lay smaller egg masses due to their smaller body size.



Figure 4. Dusky gopher frog egg mass, Harrison County, Mississippi. Photo by Shauna Ginger, USFWS

Dusky gopher frog egg masses take 9 to 21 days to complete hatching; the hatching rate is driven by water temperature (Richter and Seigel, unpublished data, Baxley and Qualls 2007). Metamorphosis occurs from mid-May to early August at Glen's Pond (Richter *et al.* 2003, Sisson *et al.* 2008). Tadpoles develop in the pond and may metamorphose as early as 94 days after hatching (Pechmann pers. comm. 2014); however, if the breeding pond continues to hold water, tadpoles may gain mass and metamorphose after a longer period. The date that metamorphosis begins appears to be unaffected by oviposition date and over-wintering of dusky gopher frog tadpoles has been documented (Sisson 2003, Pechmann and Tupy 2010). For example, during the 2009/2010 breeding season, juvenile dusky gopher frogs were first observed on June 2, 2010, 250 days after the first eggs of the season were laid in September of 2009 (Pechmann and Tupy 2010). In contrast, during the 2012/2013 breeding season, the first metamorphosed juveniles were observed on June 2, 2013, only 94 days after the first oviposition on February 28, 2013 (Pechmann and Tupy 2013). Monitoring of the Glen's Pond population has provided evidence that the period of metamorphosis is often truncated by pond drying (Richter *et al.* 2003, Sisson *et al.* 2008, Pechmann and Tupy 2013).

Richter and Seigel (2002) found that metamorphic body size differed considerably between years. Size (measured as snout-vent length/mass) of pond-reared dusky gopher frogs ranged from 1 in/0.05 ounces (oz) to 1.7 in/0.24 oz (24.8 mm/1.5 grams (gm) to 42 mm/6.8 g) (Richter and Seigel 2002). The proportion of juveniles resulting from a breeding event compared to the number of eggs deposited is highly variable. It can range from 0 percent in years when the breeding site dries before metamorphosis is possible, to 5.4 percent (Richter *et al.* 2003). Richter and Jensen (2005) surveyed the literature and noted that estimates for this measure of reproductive success, when there was no reproductive failure, ranged from 4.3 to 5 percent in other ranid frogs with similar life histories.



Figure 5. Dusky gopher frog burrow and basking site; dusky gopher frog basking outside burrow. Photos by John Tupy

After breeding, adult dusky gopher frogs leave pond sites during rainfall events and move to terrestrial belowground refugia. Metamorphic frogs follow, once their development is complete. Limited data are available on the distance between the wetland breeding and upland terrestrial habitats of post-larval and adult dusky gopher frogs. Richter *et al.* (2001) used radio transmitters to track a total of 13 adult frogs from Glen's Pond to their primary upland retreats. The farthest movement recorded was 981 feet (ft) (299 meters (m)) by a frog tracked for 63 days from the time of its exit from the breeding site (Richter *et al.* 2001). Tupy (2012) conducted a more recent radio telemetry study of 17 dusky gopher frogs captured at Glen's Pond. The maximum distance traveled by one of these frogs to its underground refuge was 787 ft (240 m). In 2013, dusky gopher frogs from the Glen's Pond population moved 0.8 mi (1.3 km) to Pony Ranch Pond where they bred (Pechmann and Tupy 2013) (See discussion below: **Management through Partnerships**). Apparently, dusky gopher frogs are able to move considerable distances, and movements may be tied to abundance and distribution of below-ground refugia and available breeding habitat. Distances moved between breeding and upland habitat by other gopher frogs have been reviewed by the USFWS (2012).

In the wild, male dusky gopher frogs attain adult size and become reproductively mature at age 1 to 5 years and females at 2 to 5 years (Richter and Seigel 2002, Pechmann *et al.* 2012). Results from field enclosure experiments indicate timing to maturity can take up to 5 years depending on habitat quality (J. Tupy, Western Carolina University, pers. comm. 2013). Adult body size ranges from 2.2 in to 4.1 in (56 to 105 mm) and varies between the sexes with females being larger than males (Goin and Netting 1940, Wright and Wright 1949, Richter and Seigel 2002). The estimated maximum longevity, based on mark-recapture data, for male dusky gopher frogs is 9 years and 12 years for females (Pechmann *et al.* 2012). However, only an estimated one quarter of males live longer than 3 years, and only one third of females live longer than 5 years (Richter and Seigel 2002, Pechmann *et al.* 2012). Frogs breed, on average, only one to two seasons during their lifetime (Richter and Seigel 2002, Pechmann *et al.* 2012).

Little information is available regarding the food habits of dusky gopher frogs. Dusky gopher frog larvae are likely filter-feeders in their pond's water column and also grazers on periphyton

and epiphytic algae, as is typical of most tadpoles (Duellman and Trueb 1986, Alford 1999, Hoff *et al.* 1999). Netting and Goin (1942) provide the only published account for the diet of an adult dusky gopher frog and described finding carabid (*Pasimachus* sp.) and scarabaeid (genera *Canthon* sp. and *Ligryus* sp.) beetles in the gut of one specimen. Adult dusky gopher frogs are carnivorous and likely have a diet similar to that reported for other species of gopher frogs which includes frogs, toads, small mammals, beetles, hemipterans, grasshoppers, spiders, roaches, and earthworms (Deckert 1920, Carr 1940, Dickerson 1969, Blihovde, USFWS, pers. comm. 2005).

## **HABITAT**

The dusky gopher frog is an endemic of the longleaf pine ecosystem. Optimal habitat is maintained when management includes the use of seasonally-appropriate prescribed fire to support a diverse ground cover of herbaceous plants, both in the uplands and in the breeding ponds (Hedman *et al.* 2000, Kirkman *et al.* 2000, Roznik *et al.* 2009). The use of prescribed fire as a management tool has been reduced as longleaf pine dominated uplands have been converted to pine plantations (often loblolly (*P. taeda*) or slash pine (*P. elliotii*)). Outside of occupied habitat and those areas managed as potential translocation sites, many remaining parts of the longleaf pine ecosystem within the historical range of the frog continue to decline through fragmentation and destruction, primarily as a result of urbanization from residential and commercial development (Wear and Greis 2013).

Dusky gopher frog habitat includes both upland sandy and sandy loam habitats—historically forest dominated by longleaf pine—and wetland breeding sites embedded within the forested landscape. Breeding sites are ephemeral (seasonally flooded) ponds not connected to other water bodies (isolated) (Kirkman *et al.* 2007) with an open canopy (Thurgate and Pechmann 2007). Prescribed fire is being used at those sites occupied by the dusky gopher frog to maintain optimal habitat, and at sites being managed as potential translocation sites, to create the open canopy and native groundcover vegetation of the frog’s aquatic and terrestrial habitats (Roznik *et al.* 2009). Additional active management to improve habitat quality is occurring at both occupied and potential translocation sites including tree thinning, planting longleaf pine, restoring native groundcover vegetation, and controlling invasive vegetation.

Adult and subadult dusky gopher frogs spend the majority of their lives underground, generally in stump holes or small mammal burrows within their forested habitat (Richter *et al.* 2001, Tupy 2012). Historically, they were frequently found in active and abandoned gopher tortoise (*Gopherus polyphemus*) burrows (Allen 1932). Thurgate (2006) conducted a choice experiment with two different sets of artificial burrows, those containing chemical cues of gopher tortoises or cotton mice (*Peromyscus gossypinus*) and those without. She found that dusky gopher frogs spent significantly more time in the treatment burrows than control burrows. This suggests that the species has an innate response to the chemical cues of these species, and that this response may help them locate burrows in the environment. The gopher tortoise, whose burrows are considered good terrestrial habitat for gopher frogs, is listed as a threatened species under the Act within the range of the dusky gopher frog and is in decline. Thus, the specialized microhabitat which they create is rare in occupied dusky gopher frog habitat. Because fossorial (underground) habitat represents the primary upland habitat for the dusky gopher frog, their survival is dependent on the quality and quantity of appropriate underground refugia (Roznik and

Johnson 2009b). High winds, generated during Hurricane Katrina in August of 2005, pushed over many pine trees in the vicinity of Glen's Pond and created a large number of belowground habitats for the dusky gopher frog.



Figure 6. Glen's Pond, the primary breeding site for the dusky gopher frog. Photo by: Linda LaClaire, USFWS

Connectivity of dusky gopher frog breeding and nonbreeding habitat within the geographic area occupied by the species must be maintained to support the species' survival (Semlitsch 2002, Rothermel 2004, Harper *et al.* 2008, Richter *et al.* 2009, Richter and Nunziata 2013). This connectivity allows for gene flow among local populations within a metapopulation, which enhances the likelihood of metapopulation persistence and allows for recolonization of sites that are lost due to drought, disease, or other factors (Hanski and Gilpin 1991).



Figure 7. Longleaf pine upland habitat occupied by dusky gopher frogs during most of their adult lives.  
Photo by John Tupy

Published studies of population dynamics in gopher frogs (*R. capito*) indicate that their populations are naturally (but often only historically) distributed across the landscape among multiple breeding ponds interconnected by suitable upland habitat; they may have small local/pond subpopulation sizes, which cumulatively can form large populations (Semlitsch *et al.* 1995, Greenberg 2001, Richter *et al.* 2009). When multiple breeding ponds were present in the landscape, there was greater potential for recruitment in a given year. It is likely that, historically, dusky gopher frogs were similarly distributed. As subpopulations of dusky gopher frogs became fragmented and isolated, overall population sizes and genetic variation rapidly diminished (Richter *et al.* 2009). The result is that only three small, isolated, naturally-occurring populations have been documented since 2001 and their distribution is limited from what was once likely a larger, connected complex of subpopulations and breeding ponds.

### **Critical Habitat**

Critical habitat was designated for the dusky gopher frog on June 12, 2012 (77 FR 35118). Based on our current knowledge of the life history, biology, and ecology of the species, and the habitat requirements for sustaining the essential life-history functions of the species, we determined that the primary constituent elements (PCEs) for the dusky gopher frog are:

(1) *Ephemeral wetland habitat*. Breeding ponds, geographically isolated from other waterbodies and embedded in forests historically dominated by longleaf pine communities, that are small (generally < 1 to 10 ac (0.4 to 4.0 ha)), ephemeral, and acidic. Specific conditions necessary in breeding ponds to allow for successful reproduction of dusky gopher frogs are:

- (a) An open canopy with emergent herbaceous vegetation for egg attachment;
- (b) An absence of large, predatory fish that prey on frog larvae;
- (c) Water quality such that frogs, their eggs, or larvae are not exposed to pesticides or chemicals and sediment associated with road runoff; and
- (d) Surface water that lasts for a minimum of 195 days during the breeding season to allow a sufficient period for larvae to hatch, mature, and metamorphose.

(2) *Upland forested nonbreeding habitat*. Forests historically dominated by longleaf pine, adjacent to and accessible to and from breeding ponds, that are maintained by fires frequent enough to support an open canopy and abundant herbaceous ground cover and gopher tortoise burrows, small mammal burrows, stump holes, or other underground habitat that the dusky gopher frog depends upon for food, shelter, and protection from the elements and predation.

(3) *Upland connectivity habitat*. Accessible upland habitat between breeding and nonbreeding habitats to allow for dusky gopher frog movements between and among such sites. This habitat is characterized by an open canopy, abundant native herbaceous species, and a subsurface structure that provides shelter for dusky gopher frogs during seasonal movements, such as that created by deep litter cover, clumps of grass, or burrows.

We designated critical habitat on lands that have been determined to be essential to the conservation of the dusky gopher frog.

Twelve critical habitat units, three of which are divided into two subunits each, were included in the designation. Of these, a total of 1,544 ac (625 ha) are located in St. Tammany Parish, Louisiana, and 4,933 ac (1,996 ha) are located in Forrest, Harrison, Jackson, and Perry Counties, Mississippi. Fifty-four percent of these acres are in Federal ownership, 4 percent in state (Mississippi) ownership, and 42 percent in private ownership. Twenty-seven percent of the critical habitat acreage is occupied and 73 percent is unoccupied. Seven critical habitat units are primarily in the DNF in Harrison, Forrest, and Perry Counties, Mississippi; one critical habitat unit is on Ward Bayou Management Area owned by the U.S. Army Corps of Engineers, Jackson County, Mississippi; one critical habitat unit is on a site owned by the state of Mississippi in Jackson County, Mississippi; two critical habitat units are on property owned by TNC, Jackson County, Mississippi; and one critical habitat unit is located on private property, St. Tammany Parish, Louisiana.

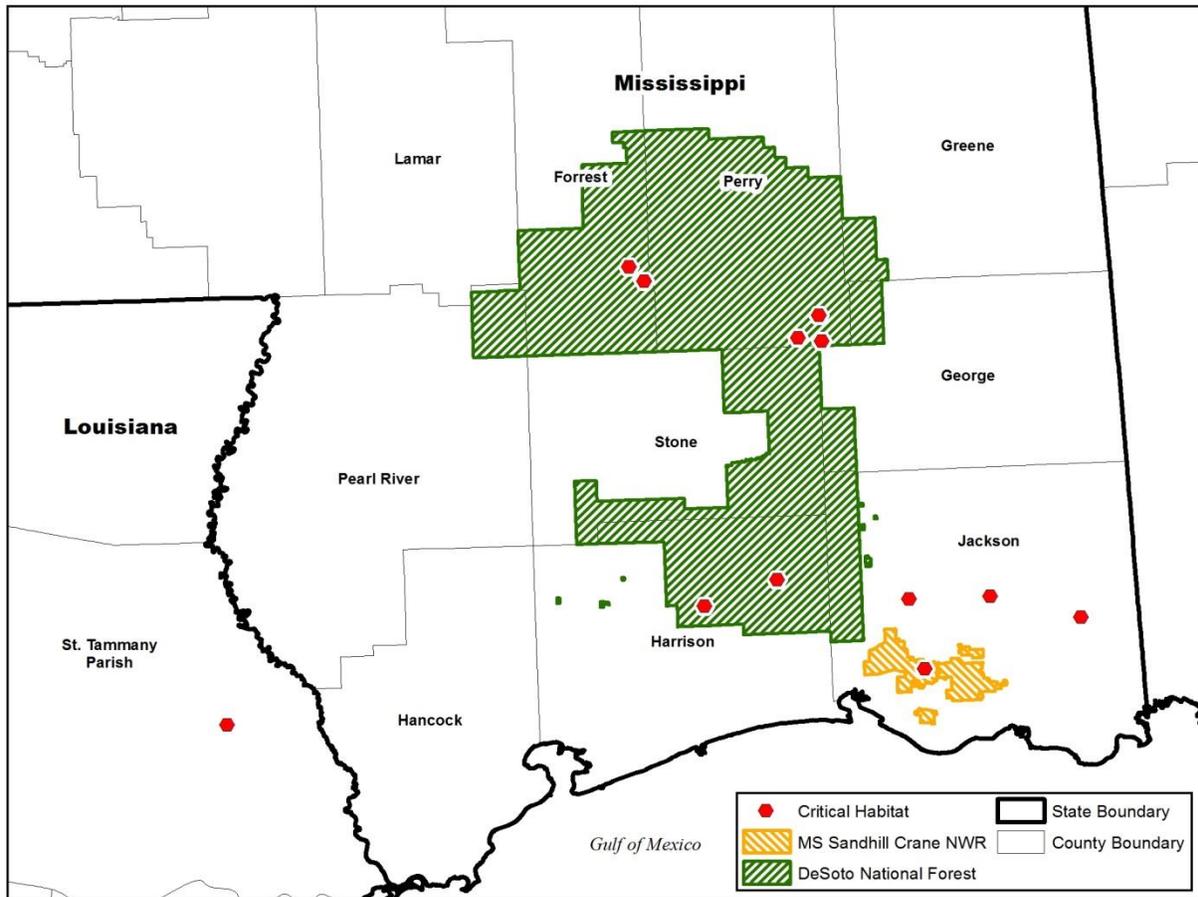


Figure 8. Critical Habitat Units for the Dusky Gopher Frog (USFWS 2012).

Table 1. Designated critical habitat units for dusky gopher frog by land ownership. Shaded unit names represent those units that are currently occupied. [Area estimates (hectares (ha) and acres (ac)) reflect all land within critical habitat unit boundaries.] (USFWS 2012)

Unit	Parish/County	Ownership			Total Area
		Federal	State	Private	
<b>LOUISIANA</b>					
1	St. Tammany			625 ha (1,544 ac)	625 ha (1,544 ac)
<b>MISSISSIPPI</b>					
2, Subunit A	Harrison	100 ha (247 ac)		21 ha (52 ac)	121 ha (299 ac)
2, Subunit B	Harrison	425 ha (1,050 ac)		3 ha (7 ac)	428 ha (1,057 ac)
3	Harrison	121 ha (299 ac)			121 ha (299 ac)
4, Subunit A	Jackson			121 ha (299 ac)	121 ha (299 ac)
4, Subunit B	Jackson	48 ha (119 ac)		109 ha (269 ac)	157 ha (388 ac)

5, Subunit A	Jackson			121 ha (299 ac)	121 ha (299 ac)
5, Subunit B	Jackson			54 ha (133 ac)	54 ha (133 ac)
6	Jackson	121 ha (299 ac)			121 ha (299 ac)
7	Jackson		107 ha (264 ac)	14 ha (35 ac)	121 ha (299 ac)
8	Forrest	121 ha (299 ac)			121 ha (299 ac)
9	Forrest	120 ha (297 ac)		1 ha (2.5 ac)	121 ha (299 ac)
10	Perry	127 ha (314 ac)		20 ha (49 ac)	147 ha (363 ac)
11	Perry	119 ha (294 ac)		2 ha (5 ac)	121 ha (299 ac)
12	Perry	115 ha (284 ac)		6 ha (15 ac)	121 ha (299 ac)
Total		1,417 ha (3,501 ac)	107 ha (264 ac)	1,097 ha (2,711 ac)	2,621 ha (6,477 ac)

Note: Area sizes may not sum due to rounding.

## REASONS FOR LISTING AND ONGOING THREATS ASSESSMENT

The USFWS listed the dusky gopher frog as endangered primarily due to threats of habitat loss and degradation, and small population size. The factors leading to the listing of the species continue to affect its long-term viability. The following discussion of threats below addresses each of the five listing factors outlined in section 4(a)(1) of the Act, and incorporates new information obtained since the final rule to list the dusky gopher frog (USFWS 2001) and final critical habitat rule (USFWS 2012).

### **Destruction, modification, or curtailment of the dusky gopher frog's habitat or range.**

The dusky gopher frog is an endemic of the longleaf pine ecosystem. Outside of occupied habitat and those areas managed as potential translocation sites, the remaining parts of this ecosystem within the historical range of the frog continue to decline through fragmentation and destruction, primarily as a result of urbanization from residential and commercial development. In addition, management of remaining natural areas of the longleaf pine ecosystem is inadequate (e.g., limited use of prescribed fire as a management tool) (See discussion below: **Other natural or manmade factors affecting the dusky gopher frog's continued existence**). Plant community changes as a result of invasive species such as cogon grass (*Imperata cylindrica*) and tallow tree (*Triadica sebifera*) represent an additional threat to the frog's habitat. Optimal terrestrial microhabitat, within burrows of the threatened gopher tortoise, continues to decline as gopher tortoise populations are diminished (Hinderliter 2015). Historically, the dusky gopher frog was found in the coastal counties of Mississippi as well as in the Florida parishes of Louisiana (8 parishes in southeast Louisiana that include East Baton Rouge, East Feliciana, Livingston, St. Helena, St. Tammany, Tangipahoa, Washington, and West Feliciana) and in Alabama west of Mobile Bay (Allen 1932, Dickerson 1969, Neill 1957, Dundee and Rossman 1989). Populations in Alabama and Louisiana appear to be extirpated (Pechmann *et al.* 2006, Bailey 2009). Searches for potentially suitable gopher frog habitat in Alabama are on-going in 2015 and some likely candidate ponds will be studied further. Recent visits to localities

historically occupied by the frog in Louisiana have verified the continued presence of suitable breeding sites there. In Mississippi, only three naturally-occurring dusky gopher frog populations have been documented since 2001; an additional population has been established through translocation.

Connectivity of dusky gopher frog breeding and nonbreeding habitat within the geographic area occupied by the species, and gene flow among local populations within a metapopulation, are important for the species' survival (See discussion above: **HABITAT**). Additionally, connectivity of these sites with other areas outside the geographic area occupied currently by the dusky gopher frog is essential for the conservation of the species (Semlitsch 2002, Harper *et al.* 2008) (See discussion of habitat fragmentation below: **Other natural or manmade factors affecting the dusky gopher frog's continued existence**). Metapopulation dynamics is also important for the dusky gopher frog because ponds with slightly different drainage basins and hydrologies will respond differently to variations in local rainfall and provide different breeding opportunities. This variability can make the difference between whether or not tadpoles survive to metamorphosis in a population. Until recently, there were no dusky gopher frog metapopulations. However, longleaf pine restoration efforts and work conducted by the Forest Service to improve the hydrology of a pond (Pony Ranch Pond; see discussion below under: **Management through Partnerships**) near Glen's Pond, the primary breeding pond for dusky gopher frogs, has resulted in a new breeding site and creation of a metapopulation for the species on the DNF.

A site slated for residential development is located immediately north of the Glen's Pond breeding site on the DNF. After the frog's listing under the Act, the USFWS began working with the developers of the site to restore and protect habitat immediately adjacent to the DNF property boundary (See discussion under **Conservation Measures** for the positive actions that have been taken). Nevertheless, large scale development in the vicinity of this habitat, including ongoing highway expansion, will fragment the remaining longleaf pine habitat in the area (See discussion below: **Other natural or manmade factors affecting the dusky gopher frog's continued existence**). Urbanization will expand along these highway corridors and further reduce potential habitat for the frog.

The Mike's Pond dusky gopher frog breeding population is located primarily on a site owned by TNC. Unfortunately, part of the Mike's Pond drainage basin occupies private property outside of TNC ownership. A semi-truck repair shop was developed on this area after the owner's original shop was destroyed during Hurricane Katrina. Efforts have been made to work with the shop owner and encourage him to eliminate possible sources of toxic chemical inflow into Mike's Pond. Runoff of oils, gasoline, or other toxic substances from this shop represents a very real threat to the future of the Mike's Pond population.

Dusky gopher frog wetland breeding sites have also been degraded and destroyed. The number and diversity of these small wetlands have been reduced by alterations in hydrology, agricultural and urban development, incompatible silvicultural practices, shrub encroachment, dumping into or filling ponds, conversion of wetlands to fish ponds or farm ponds for domestic animal grazing, soil disturbance, and highway construction (Richter and Jensen 2005). Fire suppression and hydrological alterations represent serious threats to dusky gopher frog breeding sites. Fire

suppression at some sites has led to tree and shrub encroachment into ponds and destruction of the herbaceous groundcover needed for egg attachment. Lowered water levels and shortened hydroperiods, even at sites with herbaceous groundcover, limit opportunities for successful dusky gopher frog reproduction.

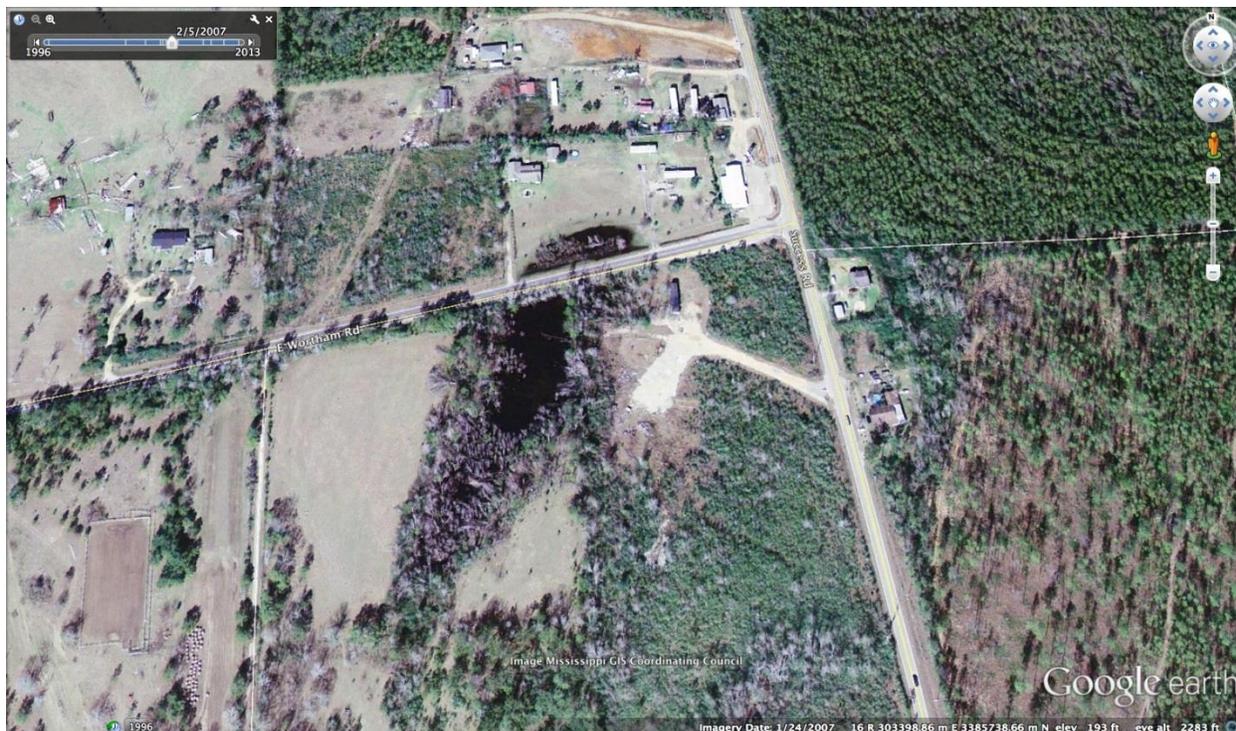


Figure 9. Aerial photo of an historical dusky gopher frog locality. Breeding site destroyed by road construction; uplands fragmented and converted to pasture and homes (Google Earth, imagery: 1/27/2007, Mississippi GIS Coordinating Council).

### **Overutilization for commercial, recreational, scientific, or educational purposes.**

Direct take of dusky gopher frogs for commercial, recreational, scientific, or educational purposes was not considered a threat at the time of listing and is not currently known to be a threat to the species.

### **Disease or predation.**

A lethal disease killed most gopher frog tadpoles at the Glen's Pond site in 2003 (Overstreet and Lotz 2004). Recent monitoring indicates this disease, an unnamed protist (*Dermomycooides* sp., also known as "Perkinsus-like" disease (Green *et al.* 2003, Jones *et al.* 2012)) is still present at the site, but mortality is sporadic and has never been as high as that which occurred during the first episode. The disease has also recently caused mortality of dusky gopher frog tadpoles at Pony Ranch Pond (Pechmann and Tupy 2014), the site where the disease was originally observed in Mississippi in 2001. Fortunately, this disease does not appear to negatively affect adult dusky gopher frogs. Portions of egg masses are collected at each dusky gopher frog breeding event, the eggs are hatched in the lab, and tadpoles are raised in cattle tanks adjacent to the pond. This strategy helps to ameliorate any threat from disease. Metamorphic frogs from the cattle tanks are then released back at the breeding site. In addition to the above disease, a disease caused by the pathogen *Batrachochytrium dendrobatidis*, commonly referred to as chytrid fungus,

has been found in two other species of amphibians at Glen's Pond, although not yet on dusky gopher frogs (Sisson 2003). Dusky gopher frogs may not be susceptible to the disease due to their basking behavior. Thermal environment has been shown to affect the progress of this disease. Experimentally housing frogs at 98.6 degrees Fahrenheit (F) (37 degrees Celsius (C)) for less than 16 hours can clear them of the pathogen (Woodhams *et al.* 2003).

Ranaviruses in the family Iridoviridae may be potential threats. Ranaviruses have been responsible for die-offs in eight different species of frogs, especially those with an aquatic larval stage. Studies of the susceptibility of the dusky gopher frog to infection by ranaviruses have been conducted at the University of Tennessee. Initial results indicate that dusky gopher frogs are highly susceptible to ranavirus and when infected in laboratory experiments, have high mortality rates approaching 100 percent (Gray 2013; frogs for this experiment were individuals that resulted from an earlier captive breeding experiment and were excess to those needed to maintain genetic diversity in captivity). For the most part, diseases of amphibians in the southeastern United States have not been well studied, and they may represent a bigger threat to the dusky gopher frog than is currently understood.

Predation may be a threat to the dusky gopher frog. Predation is expected to be high as survivorship from the egg stage to adulthood is typically low for ranid frogs (reviewed in Richter *et al.* 2003). No published records of predation on adults or juvenile dusky gopher frogs exist, but predators would be similar to those of other gopher frog and ranid species (e.g., snakes, birds, and mammals; Jensen and Richter 2005, Pechmann and Tupy 2010). Richter (2000) reported an undetermined amount of the egg mortality due to predation by caddisfly larvae (Order Trichoptera, Family Phryganeidae) on the egg masses. Caddisfly infestations of dusky gopher frog egg masses have been variable since the time of listing (Baxley and Qualls 2007); however, they do not currently pose a threat to the species. No other direct documentation of egg or larval predation on dusky gopher frogs exists, but potential predators include those observed feeding on southern leopard frog eggs (*Rana sphenoccephala*) and larvae in Glen's Pond and those of other gopher frog species. These potential predators include dragonfly naiads (Odonata), backswimmers (Hemiptera), giant water bugs (Hemiptera), predaceous diving beetles (Coleoptera), fish, salamanders, snakes, turtles, and birds (Jensen and Richter 2005, Richter pers. comm. 2013).

Predation from fishes likely contributed to the loss of historic populations. Studies of other amphibians, which breed in temporary wetlands, have demonstrated a decline in larval survival in the presence of predatory fish (Semlitsch 1987, 1988). Gregoire and Gunzburger (2008) studied the effects of predatory fish on survival and behavior of larval gopher frogs (*R. capito*) and southern leopard frogs (*Rana sphenoccephala*) in Florida. Their results suggested that the presence of fish predators had a greater effect on survival and behavior of gopher frog tadpoles than those of the southern leopard frogs which are habitat generalists. Thurgate (2006) conducted experiments with dusky gopher frogs in which she recorded their lack of a behavioral response to the presence of the green sunfish (*Lepomis cyanellus*). This lack of a response suggests that the species may lack inducible defenses against fish predation and may be more vulnerable to fish introductions than other species (Thurgate 2006). Exposure to increased predation by fish may be a threat to current dusky gopher frog populations when isolated, seasonally-ponded wetland breeding sites are changed to, or connected with, more permanent

wetlands inhabited by fishes. In addition, ponds may be modified specifically to serve as fish ponds, sites may be altered due to the construction of drainage ditches or firebreaks which allow fish to enter the wetlands, or fisherman may purposefully stock fish at sites.

Predation on amphibians by the red imported fire ant (*Solenopsis invicta*) has been reported in the literature (Allen *et al.* 2004) and these ants have been observed at Glen's Pond and caused the death of at least one gopher frog (Pechmann and Thurgate 2001). Although this invasive species appears to be tied to disturbance and does not invade undisturbed forest habitats of native ants (King and Tschinkel 2008), increases in habitat alterations near occupied habitat is a concern (Todd *et al.* 2008). At Glen's Pond, control of this species is necessary in the disturbed area of the drift fence surrounding the pond. The threat of predation by red imported fire ants is likely tied to the increase in urbanization occurring through-out the longleaf pine ecosystem (See discussion under: **Destruction, modification, or curtailment of the dusky gopher frog's habitat or range.**).

Although the magnitude of the threat from disease and predation is unknown, a significant increase in mortality resulting from these factors is a concern due to the extremely small size, low levels of natural recruitment, and isolation of dusky gopher frog populations.

#### **Inadequacy of existing regulatory mechanisms.**

At the state and local levels, regulatory mechanisms are limited to restrictions on direct take of dusky gopher frogs and do not protect the habitat required for their survival. Although not listed as threatened or endangered in Alabama, the dusky gopher frog is listed among those non-game species for which it is "unlawful to take, capture, kill, or attempt to take, capture or kill; possess, sell, trade for anything of monetary value, or offer to sell or trade for anything of monetary value" (ADCNR 2013). In January 2013, the Louisiana Wildlife and Fish Commission voted to add the Mississippi (dusky) gopher frog to the Louisiana list of endangered species so that it cannot be killed or removed from the wild without a permit and this change was formalized in 2014 (Louisiana Administrative Code 2014). In Mississippi, the dusky gopher frog is listed as endangered and is protected under the Nongame and Endangered Species Conservation Act of 1974 which prohibits taking, possessing, transporting, exporting, offering to sell, or offering to ship endangered species. There are no Alabama, Louisiana, or Mississippi state regulations that protect dusky gopher frog habitat.

Several Federal regulations exist that provide measures to protect habitat of the dusky gopher frog. For example, on June 12, 2012, critical habitat, as defined under the Act, was designated for the dusky gopher frog (77 FR 35118; USFWS 2012). This designation includes 1,196 ac (484 ha) of habitat occupied by the frog, as well as 5,281 ac (2,137 ha) of unoccupied habitat. The protections afforded under the Act to unoccupied critical habitat will help secure essential habitat features for the frog. However, critical habitat protection would only apply in cases where a Federal action, such as Federal funding or a Federal permit, is associated with the potential destruction of dusky gopher frog critical habitat.

The dusky gopher frog's habitat is afforded some protection under the National Forest Management Act (NFMA) when it occurs on lands managed by the Forest Service. Forest Service rules and guidelines implementing NFMA require land management plans include

provisions supporting recovery of endangered and threatened species. Land management plans must contain components to maintain or restore ecosystem integrity, ecosystem diversity, and provide additional components where needed to contribute to the recovery of listed species and conservation of proposed and candidate species. In addition, Section 7(a)(1) of the Act requires Federal agencies to carry out programs for the conservation of endangered and threatened species. Land managers on the DNF, where the primary population of the dusky gopher frog occurs, have conducted management actions in both occupied and unoccupied habitat to benefit the dusky gopher frog. These actions have included prescribed burning, pond restoration, and upland habitat improvements; improvements at one particular site have resulted in dusky gopher frog breeding at a pond near the primary breeding site, Glen's Pond.

Section 404 of the Clean Water Act (CWA) is the primary Federal law that has the potential to provide some protection for the wetland breeding sites of the dusky gopher frog. However, due to recent case law (*Solid Waste Agency of Northern Cook County v. U.S. Army Corps of Engineers (SWANCC)* 2001, *Rapanos v. United States (Rapanos)* 2006), isolated wetlands, such as dusky gopher frog breeding sites, are no longer considered to be under Federal jurisdiction. Nevertheless, in the case of large development sites where dusky gopher habitat occurs and jurisdictional wetlands would be affected, CWA may come into play when a section 404 permit is required.

**Other natural or manmade factors affecting the dusky gopher frog's continued existence.**

Fire is the preferred habitat management tool used to maintain the natural longleaf pine community. Fire suppression of naturally-occurring fire and low fire frequencies have the potential of reducing the quality of terrestrial and aquatic habitat for the dusky gopher frog. Urban areas are being developed around dusky gopher frog habitat and, as a result, it is becoming more challenging to conduct prescribed burns. Drought has also contributed to a reduction in the number of days available to conduct prescribed burns (See discussion of annual variability of rainfall below, under this factor). Although prescribed burning is an important management tool, timing of introducing fire into dusky gopher frog habitat should be carefully assessed in order to prevent mortality to the species during its migrations to and from breeding sites (Humphries and Sisson 2012).

Pesticides and herbicides commonly used in habitat management pose a threat to amphibians such as the dusky gopher frog, because their permeable eggs and skin readily absorb substances from the surrounding aquatic or terrestrial environment (Duellman and Trueb 1986). Negative effects of commonly used pesticides and herbicides on amphibian larvae include delayed metamorphosis, paralysis, reduced growth rates, and mortality (Bishop 1992, Berrill *et al.* 1997, Bridges 1999). Sublethal levels of chemical contamination can alter juvenile recruitment in amphibian populations (Bridges and Semlitsch 2000, Rohr *et al.* 2013). Herbicides may alter the density and species composition of vegetation surrounding a breeding site and reduce the number of potential sites for egg deposition, larval development, or shelter for migrating frogs. For the reasons described above, the USFWS and our private and Federal partners who own property occupied by the dusky gopher frog are vigilant in the approval and use of any pesticides and/or herbicides on these sites. Through cooperation, we are working to keep this threat extremely low.

Habitat fragmentation of the longleaf pine ecosystem, resulting from habitat conversion, threatens the survival of the remaining dusky gopher frog populations. Even large tracts of intact longleaf pine habitat are fragmented by roads and pine plantations. Roads contribute to habitat fragmentation by isolating blocks of remaining contiguous habitat. This fragmentation may disrupt migration routes and dispersal of individuals to and from breeding sites and result in the death of dusky gopher frogs when they are attempting to cross roads. Extant dusky gopher frog populations are widely separated from each other by unsuitable habitat. Studies have shown that the loss of small, fragmented populations is common, and recolonization is critical for their regional survival (Fahrig and Merriam 1994, Burkey 1995, Marsh and Trenham 2001). As patches of available habitat become separated beyond the dispersal range of a species, disruption of metapopulation dynamics occurs and populations become more sensitive to genetic, demographic, and environmental variability and may be unable to sustain themselves (Gilpin 1987, Sjogren 1991, Blaustein *et al.* 1994). Dusky gopher frogs, not existing as part of a metapopulation, may be unable to recolonize areas after local extinctions due to their physiological constraints, relatively low mobility, and site fidelity (Blaustein *et al.* 1994). The isolation of dusky gopher frog populations eliminates the possibility of reestablishment occurring naturally and brings into question the long-term viability of the species. The genetic and population ecology data available for the dusky gopher frog illustrate the consequences of geographic range collapse and geographic isolation of populations: reduced overall population sizes, increased negative effects of variation in reproductive success, inbreeding-related mortality, low genetic diversity, and elevated probability of extinction (Richter *et al.* 2009, Richter and Nunziata 2013). Small populations are at increased threat from natural processes and random events (genetic isolation, inbreeding, and drought) as well as the threats listed above. Inbreeding depression and loss of genetic diversity may also occur in small populations and reduce the fitness of individuals and the ability of the population to adapt to change (Frankel and Soule 1981), as well as increase their vulnerability to environmental stressors (Weyrauch and Grubb 2006).

Low reproductive potential may also present a threat to the dusky gopher frog's continued existence. Studies at the Mississippi breeding site suggest that female dusky gopher frogs do not breed until at least 2 to 3 years of age and only average one to two lifetime breeding events (Richter *et al.* 2003, Pechmann *et al.* 2012). In addition, larval survival at Glen's Pond is extremely low (Richter *et al.* 2003, Pechmann *et al.* 2012).

Annual variability in rainfall influences how frequently, and how long, a pond remains appropriate breeding habitat for the dusky gopher frog. The amount of rainfall has been shown to have a positive effect on the number of egg masses produced in closely related *R. capito* (Jensen *et al.* 2003). Breeding events can be unpredictable (and may become more so with climate change), and the likelihood that recruitment will occur in a given year cannot be predicted. Higher temperatures that may result from climate change could reduce the hydroperiod of breeding ponds. A pond must hold water long enough for metamorphosis of dusky gopher frog tadpoles to occur, typically in late May or June (Richter *et al.* 2003, Sisson *et al.* 2008, Pechmann and Tupy 2010, 2013). Since the frog was listed in 2001, natural recruitment has occurred at Glen's Pond in only 5 of 13 (38 percent) breeding seasons at Glen's Pond due either to inadequate rainfall needed for filling the breeding pond or keeping it filled until metamorphosis was possible (Sisson *et al.* 2008, Pechmann and Tupy 2010, 2012, 2013,

2014). In 2 of the last 12 (17 percent) breeding seasons, Glen's Pond remained dry and the frogs did not breed; in 6 of the last 12 (50 percent) breeding seasons the frogs bred but the ponds dried before their tadpoles could complete development (Pechmann *et al.* 2012). Although rainfall variability is a result of natural processes, extreme weather events such as drought may increase as a result of global climate change. When rainfall variability is combined with other threats such as population isolation, small population size, and low reproductive potential, dusky gopher frog populations may be threatened to the point that they cannot recover.

## **CONSERVATION MEASURES**

### **Federal Regulatory Protection**

A number of tools are available to benefit the conservation of the dusky gopher frog due to its placement on the list of threatened and endangered species authorized by the Act. Section 6 of the Act directs the USFWS to cooperate with state agencies to conserve listed species and to provide funding for recovery projects. Through section 7 of the Act, Federal agencies are required to use their authorities to carry out programs for the conservation of listed species, and to consult with the USFWS when a Federal action may have an effect on listed species or its critical habitat. The primary breeding pond for the dusky gopher frog is Glen's Pond located on Federal lands managed by the DNF, and thus any actions taken by the Forest Service there that may impact this species would require consultation. Examples of Federal actions that may affect the dusky gopher frog include logging and other vegetation removal activities, management of recreation, road construction, prescribed fire and fire suppression. The outcome of the section 7 consultation often involves inclusion of reasonable and prudent measures into project plans to minimize "take" of listed species or otherwise reduce potential adverse effects to the species and its habitat. In biological opinions, the USFWS also provides conservation measures that Federal agencies can implement on a voluntary basis. Development on private or state lands requiring permits from Federal agencies, such as permits from the U.S. Army Corps of Engineers (COE) under Section 404 of the CWA, are also subject to the section 7 consultation process.

During the period when the frog was originally proposed to be listed as an endangered species (2000-2001), the COE consulted with the USFWS on issuance of a dredge and fill permit under the CWA for a new residential and commercial development on private land 656 feet (200 meters) immediately north of Glen's Pond. This consultation resulted in a biological opinion for the threatened gopher tortoise, and a conference opinion for the dusky gopher frog. The biological opinion written by the USFWS established measures that must be undertaken before each section of the development can proceed, for the life of the permit. In 2015, 170 ac (69 ha) of the development property were acquired by the Land Trust for the Mississippi Coastal Plain with the intent of protecting and managing the property in perpetuity for the dusky gopher frog.

Two formal section 7 consultations on actions affecting the species have been completed since the dusky gopher frog was listed as an endangered species. The first was an internal (USFWS) section 7 consultation written in 2002 on the effects of the action of issuance of 10(a)(1)(A) (recovery) permits on the dusky gopher frog. A number of research and monitoring activities were identified that had the potential to cause "take" of the species. These activities were analyzed and limits to them were established that could be used when issuing permits. The

second was a programmatic biological opinion, finalized in 2007, resulting from consultation with the Natural Resources Conservation Service (NRCS) on their pilot Healthy Forests Initiative in Mississippi (Mississippi Healthy Forest Reserve Program). This program offers funding to private landowners to assist them in managing their property in Mississippi when their actions provide conservation benefits. The dusky gopher frog was one of many species considered under this biological opinion; it was determined that the recovery of the frog would be positively affected by the program (See discussion below under: **Management through Partnerships**).

Federal actions not affecting the species or its habitat, as well as actions that are not Federally-funded or permitted, do not require section 7 consultation, however, prohibitions under section 9 of the Act apply. Section 9 of the Act prohibits “taking” (i.e., harassing, harming, pursuing, hunting, shooting, wounding, killing, trapping, capturing, or collecting) listed species. Regulations implementing the Act define “harm” to include significant habitat modification or degradation that results in the killing or injury of wildlife, and intentional or negligent “harassment” as acts that significantly impair essential behavioral patterns (i.e., breeding, feeding). Section 9 also prohibits import or export, transport in interstate or foreign commerce, or sale in interstate or foreign commerce. It is also illegal to possess, sell, deliver, carry, transport, or ship any such wildlife that has been taken illegally. Section 10 of the Act permits exemptions to the protections granted under section 9 (e.g., incidental take, scientific permits). Incidental take is take that is “incidental to, and not the purpose of, carrying out an otherwise lawful activity.” To receive an exemption from this prohibition, an applicant must prepare a habitat conservation plan (HCP) that specifies the impacts of the proposed project and the steps the applicant will take to minimize and mitigate the impacts.

### **Management through Partnerships**

Silviculture, including timber sales with associated longleaf pine restoration and pine thinnings, is the primary activity on the DNF, the location of Glen’s Pond. DNF continues to work with the USFWS, and our state and non-governmental partners, to improve habitat for the frog in the area of Glen’s Pond and elsewhere on the Forest. They have been leading the effort to restore other ponds on the DNF to make them appropriate breeding sites for the dusky gopher frog so they may be used in future translocations. Pond restoration efforts on the DNF have resulted in dusky gopher frogs colonizing a new breeding pond near Glen’s Pond and thus creating metapopulation. Additional actions conducted by managers on the DNF to improve habitat for the dusky gopher frog translocations have included improved prescribed burning, deepening of existing ponds, construction of water retention berms at existing ponds, shrub and tree removal, and prescribed fire. Ponds on DNF have been altered to increase water depth and hydroperiod, and to create a more open canopy which will support herbaceous growth. Restoration of the surrounding upland habitat through thinning and re-establishment of longleaf pine has also been implemented by DNF.

In 2002, a pond (New Pond) was constructed at a site on the DNF where one had not previously existed. The Harrison County Soil Conservation Service and the Natural Resources Conservation Service (NRCS) worked with USFWS, MDWFP, DNF, and gopher frog researchers to develop a plan for creating a pond that would provide an additional breeding site near Glen’s Pond. DNF provided personnel and equipment for the construction. The initially-

completed pond required years of alterations to improve its hydroperiod and size. In addition, considerable effort was required in order to establish herbaceous groundcover and wetland vegetation in the basin. In 2012, 10 years after the pond was first completed, it achieved the point where it was considered appropriate dusky gopher frog breeding habitat, and the first dusky gopher frog tadpoles were released there. Given that female dusky gopher frogs become reproductively mature at 2 to 5 years, the 2014-2015 breeding season would be the earliest that it could be possible to document frogs returning to the pond and successfully breeding.

The USFWS, DNF, and our non-governmental partners began working with the developers of a site immediately adjacent to Glen's Pond and the DNF property boundary to restore and protect habitat, even prior to the listing of the species. Coordinated management efforts have included control of invasive vegetation; removal of beds used to plant off-site pine species; and re-vegetation with longleaf pine trees. Representatives of the development have also permitted DNF to burn this area as a part of the adjacent forest burn unit surrounding Glen's Pond. By burning the whole area as a single unit, the need for a permanent firebreak was avoided, along with potential threats to the frog and its belowground habitat.

The Nature Conservancy has worked with the USFWS and NRCS to develop a management plan that will improve the longleaf pine habitat at the naturally-occurring dusky gopher frog population supported by Mike's Pond. TNC received funding from NRCS through the Healthy Forest Reserve Program to implement the management plan which includes prescribed burning, restoring an additional pond for potential gopher frog breeding, and planting longleaf pine on the site. Unfortunately, part of the Mike's Pond drainage basin occupies private property outside of TNC ownership. A semi-truck repair shop was developed on this area after the owner's original shop was destroyed during Hurricane Katrina. Efforts have been made to work with the shop owner and encourage him to eliminate possible sources of toxic chemical inflow into Mike's Pond.

Populations that become completely isolated, such as those of the dusky gopher frog, have high probabilities of extinction (Richter *et al.* 2009, Richter and Nunziata 2013). Due to the paucity of available suitable habitat for the dusky gopher frog, we have worked with our state, Federal, and nongovernmental partners to identify and restore additional upland and wetland habitats to create appropriate translocation sites for the species, in close proximity to each other when possible. Thus far, we have focused our efforts on areas in the state of Mississippi because of the proximity of the primary breeding site to nearby restorable habitat and the availability of willing partners. We have identified more than 15 ponds and associated forested uplands, which we considered to have restoration potential, and have worked to improve these sites as potential translocation areas. After restoration efforts were completed, suitable sites were included in the designation of critical habitat for the dusky gopher frog. After completing habitat assessments of available restored habitat, a site on TNC property, managed as Old Fort Bayou Mitigation Bank, was considered to be in the best condition to support an initial translocation attempt. Tadpoles and metamorphic frogs were released at the site and two breeding events have been verified there (See discussion below: **Translocation**). This site is currently considered to have an extant population. The potential for population establishment using translocation is limited because only the Glen's Pond population is large enough to have a sufficient number of egg masses to supply the frogs needed.

Several years of limited recruitment at the Glen's Pond site due to pond drying inspired the initial transfer of dusky gopher frogs into captivity at the Detroit Zoo and the Audubon Zoo (Seigel *et al.* 2002). After an outbreak of disease killed thousands of tadpoles at Glen's Pond in 2003 and reduced that year's recruitment to three individuals, it was apparent that establishment of a captive population was necessary to prevent the possibility that the dusky gopher frog would become extinct in the wild. The Memphis Zoo (MZ) volunteered to organize and lead this effort. Working with the USFWS, Mississippi Department of Wildlife, Fisheries, and Parks (MDWFP), and partners within the Association of Zoos and Aquariums (AZA), MZ accepted 64 diseased tadpoles from the Glen's Pond population and conducted research to look for a method to clear them of the infection. These individuals were the foundation for what became a Species Survival Plan® (Reichling and Schad 2012) managed to preserve genetic and demographic diversity under the guidelines of the AZA and to support dusky gopher frog recovery efforts. For a decade, numerous unsuccessful efforts in captive reproduction were made and the potential founder population was periodically augmented from Glen's Pond. A breakthrough using *in vitro* fertilization was achieved in 2008, and captive breeding efforts have subsequently occurred at two facilities. Results from the most recent census of dusky gopher frogs in captivity (March, 2014) indicate there are 554 individuals distributed among 16 AZA institutions. The maintenance of initial founder genetic diversity is being achieved through selected pairings to avoid inbreeding. Founder representation is maximized and balanced through breeding and transfer recommendations that are reanalyzed, revised, and disseminated every two years through use of a population management plan and studbook (Lance 2011). Given that the dusky gopher frog occurs primarily at one locality and the threats to this population are high, the USFWS appreciates the valuable efforts of MZ and other AZA members to hold and protect the genetic diversity of the species. In the future, captive-bred dusky gopher frogs may be used to establish additional wild populations; presently, only head-started individuals from the wild Glen's Pond population are used (See Task 6 under **Narrative Outline**).

The COE owns the Ward Bayou Wildlife Management Area (WBWMA) in Jackson County, Mississippi, a property managed by the MDWFP. The COE, MDWFP, USFWS, and our non-governmental partners are cooperating on efforts to establish two potential dusky gopher frog breeding ponds on WBWMA. Beginning in 2006, efforts were begun to restore one pond and create an additional pond nearby. Over time, alterations to both ponds have been necessary to improve their hydrology. Monitoring of the two ponds will continue until such time that the wetlands are determined to be appropriate breeding habitat for dusky gopher frogs and translocations can begin. In conjunction with the work on the two ponds, improvements have been made to the uplands surrounding them.

The MDWFP has used Section 6 funding provided under the Act in collaboration with the USFWS to benefit the dusky gopher frog by conducting surveys; monitoring the Glen's Pond and Mike's Pond population, as well as other sites; and head-starting tadpoles for, and monitoring, translocation efforts. The USFWS has supported a number of surveys for dusky gopher frogs in Alabama, Louisiana, and Mississippi, and funded research projects including radio-tracking of juveniles and adult frogs to determine upland habitat use; laboratory experiments to study possible innate cues that drive terrestrial habitat selection; experiments to study the unnamed disease; field studies of tadpole growth and survival in ponds of varying

canopy cover; studies to assess survival and timing to maturity in different terrestrial habitat types; and experiments to determine the best age class to use for translocation efforts. The USFWS is currently funding the monitoring of the Glen's Pond population; head-start and translocation efforts to nearby ponds along with their monitoring; and associated research on demography and optimizing translocation success.

In 2012, through a partnership between Ecological Services and Refuges, the USFWS acquired funding through our own Cooperative Recovery Initiative to work towards establishing dusky gopher frogs on the Mississippi Sandhill Crane National Wildlife Refuge (MSCNWR). This project was begun by identifying potential breeding sites to improve or restore, with the goal of implementing a translocation effort in close proximity to TNC Pond 1 which would have the potential of establishing a metapopulation (See discussion below: **Translocation Efforts**).

Gopher tortoises, whose burrows are frequently occupied by gopher frogs of other species, are absent from most of the areas currently occupied by the dusky gopher frogs. As a result, efforts to reestablish gopher tortoises to these areas have been made to improve available belowground habitat for the frogs. In 2000, three gopher tortoises were relocated to an area adjacent to Pony Ranch Pond and penned in place through the winter for a period of approximately 6 months; however, all tortoises left the area when released from their pen (Harrison County Board of Supervisors 2000). A project to reintroduce gopher tortoises to the Glen's Pond area was initiated in 2002. Ten gopher tortoises were penned near the breeding site for over a year; however, the soils in the area were poorly drained and all but one tortoise immediately left the area when the containment fence was removed (Mississippi Department of Transportation 2004). In 2012, active gopher tortoise burrows were seen approximately 1,437 ft (438 m) from Glen's Pond and may represent remnant tortoises from this project (Tupy, pers. comm. 2013). A third gopher tortoise relocation project occurred in the vicinity of New Pond. During this project, a total of 24 tortoises were placed in four pens for three to 12 months but most tortoises left the area and the project was not considered a success (Seigel 2004), although active tortoise burrows were observed less than 1000 ft (300 m) from New Pond in 2012 (Tupy, pers. comm. 2014). Although we still believe gopher tortoise burrows represent an important potential habitat for dusky gopher frogs, we are not focusing on additional tortoise relocations until we have a better understanding of why our prior efforts have had such little success.

### **Surveys**

The Glen's Pond dusky gopher frog breeding site was discovered during surveys conducted in 1988. Ever since that time, searches for additional populations of the frog have been on-going. Many partners have been involved including amateur and professional herpetologists, USFWS, MDWFP, and others. Nevertheless, it was not until 2004 that any additional breeding sites were found. In that year, calling dusky gopher frogs were discovered at two additional sites, Mike's Pond and McCoy's Pond, both in Jackson County, Mississippi. Minimal, sporadic breeding has occurred at Mike's Pond (See discussion below under: **Monitoring**) since it was discovered. No additional gopher frog activity has been documented at McCoy's Pond. Surveys in Mississippi continue. Unfortunately, no new dusky gopher frog populations have been discovered recently.

Surveys in the Florida Parishes of southeastern Louisiana, conducted by researchers and Louisiana Department of Wildlife and Fisheries personnel, have not confirmed a single

remaining gopher frog population in that state. Surveys included visits to historical sites where gopher frogs were once documented. Similarly, surveys in southwestern Alabama have failed to document any dusky gopher frog populations there. For more details, see discussion above:

## **POPULATION TRENDS AND DISTRIBUTION.**

Currently, work continues in Alabama, Louisiana, and Mississippi searching for additional dusky gopher frog populations and possible restoration/translocation sites.

### **Monitoring**

#### Glen's Pond

Glen's Pond was discovered to be a gopher frog breeding site on February 3, 1988 (Young *et al.* 1995). Egg mass and breeding call surveys were conducted at the pond from 1987 through 1996 as the primary means of monitoring the population (Young *et al.* 1995). A drift fence completely enclosing Glen's Pond was established in late 1995 and continues to be used to monitor the population. The fence is equipped with 5 gallon (gal) (18.9 liter (L)) pitfall traps (n = 128) at intervals of approximately 26 ft (8 m) on either side of the fence in order to capture adult dusky gopher frogs during breeding migrations and emigrations, and metamorphic frogs as they emerge from the pond basin. Currently, metamorphic dusky gopher frogs captured at the drift fence are marked below the knee with fluorescent VIA tags and all adult gopher frogs are implanted with a Passive Integrated Transponder (PIT) tag (Sisson *et al.* 2008). Egg mass and call surveys are used in addition to the data collected at the drift fence to monitor the population and collect demographic information.

Rainfall is frequently insufficient to establish or maintain the necessary hydroperiod for larval dusky gopher frog development and metamorphosis. Maintaining the pond water level after a dusky gopher frog breeding event was achieved in 2001 by supplementing the pond with 96,899 gal (366,805 L) of water from water tanker trucks and  $\leq 7,133$  gal/day (27,000 L/day) of water pumped from underground for 23 days (Seigel *et al.* 2006). This was attempted again in 2005 for 8 days of 5,831 gal/day (22,073 L/day) using only pumped ground water. Both events resulted in the maintenance of the pond level and allowed larval dusky gopher frogs to reach metamorphosis. In 2002, the use of cattle watering tanks (350 gal (1,325 L)) (Behlen Manufacturing Company, Columbus, Nebraska) was initiated as a technique to raise tadpoles for experiments; since then, the tanks have been used continuously in order to raise tadpoles to metamorphosis, without the threat of pond drying, and to supplement the natural recruitment at Glen's Pond (Thurgate *et al.* 2002). At each breeding event, we have removed a portion of each egg mass (to a maximum of one-third of each egg mass) and raised tadpoles through metamorphosis for release back into breeding ponds using a technique modified by Sisson (2008). Without this intervention, dusky gopher frog recruitment would be severely limited. The practice of supplementing Glen's Pond with ground water was discontinued following a die-off of dusky gopher frog tadpoles due to disease (See discussion under: **Disease or predation.**) and cattle tanks became the primary means of supplementing recruitment to the population (Tupy *et al.* 2010). The die-off is thought to have been a result of the pond water's increase in pH after ground water was added; the disease zoospores hatch most readily at a pH of 6.5 (Cook 2008). Unfortunately, the acidic water of Glen's Pond tends to move towards a neutral pH after a large influx of basic ground water (Seigel *et al.* 2001).

Demographic data collected at the drift fence by researchers supported by USFWS indicate a slight increase in the adult breeding population of dusky gopher frogs at Glen's Pond between 1996 and 2012 (Tupy and Pechmann 2013). The data also demonstrate that supplemental recruitment, through the use of cattle watering tanks (as discussed above), provided the individuals that allowed the breeding adult population to increase (Tupy and Pechmann 2013). Due to the effects of drought and disease, natural metamorphic recruitment alone would not have been sufficient to maintain the Glen's Pond population in the absence of this supplemental recruitment.

### Mike's Pond

Breeding activity at Mike's Pond has been sporadic and only small numbers of egg masses and tadpoles have been documented in comparison to the numbers recorded during Glen's Pond breeding events. Additionally, only 1 or 2 males are heard calling during breeding seasons which suggests a very small population occupies the area. In 2010, dusky gopher frogs were heard calling at Mike's Pond for three consecutive months, but egg masses were never discovered (Lee 2010). Due to fear that the population lacked mature female dusky gopher frogs, two females from the Glen's Pond population were brought to Mike's Pond and released. The day after the second female was released, two dusky gopher frog egg masses were found in the pond (Lee 2010). One-third of the egg masses were collected and hatched in a nearby laboratory. Tadpoles from the eggs were either released back into Mike's Pond (295 tadpoles/approximately 80 days post-hatching) or raised in cattle watering tanks and then released at Mike's Pond (138 metamorphs) or Glen's Pond (389 metamorphs) after metamorphosis (Lee 2010, Pechmann and Tupy 2010). Additionally, progeny were also sent to the Memphis Zoo and the Audubon Zoo (Pechmann and Tupy 2010). The Mike's Pond hydroperiod after the 2010 breeding event was long enough to allow those tadpoles not collected for head-starting sufficient time to develop to metamorphosis. In 2012, an additional 400 head-started tadpoles from Glen's Pond were released at Mike's Pond in an attempt to bolster the population (Lee 2012). The hydroperiod at the pond was likely sufficient to allow these tadpoles to undergo metamorphosis (Lee 2012). Calling males were documented at Mike's Pond in 2013, but not in 2014 (Lee 2013, Lee 2014).

### Genetics

When breeding has occurred at Glen's Pond and/or Mike's Pond, eggs have been collected from individual clutches for genetic sampling. Five eggs from each clutch are collected and stored in separate vials of 95 percent non-denatured ethanol. These samples have been sent to Dr. Stephen Richter at Eastern Kentucky University for genetic analysis. Richter *et al.* (2009) found evidence of past population bottlenecks and that population genetic variation was low overall. Richter and Nunziata (2013) discovered inbreeding depression in the Glen's Pond population; however, individuals with greater genetic variability had higher survival to metamorphosis, which should help prolong population persistence.

Dr. Richter and the Memphis Zoo have collaborated on genetic analysis of the captive dusky gopher frog population. Initial results indicate the genetics of the captive population reflect the wild population and many captive individuals are unrelated based on relatedness measures (Richter 2012).

### **Translocation Efforts**

Breeding dusky gopher frogs are primarily limited to one breeding population (See: **POPULATION TRENDS AND DISTRIBUTION**). Therefore, to achieve recovery, translocation efforts will be necessary to establish additional populations elsewhere. The first attempt at a translocation was conducted in 2001 at a pond near Glen's Pond (Reserve Pond). This attempt was unsuccessful due to drying of the pond before the tadpoles could metamorphose (Seigel *et al.* 2002). In 2004, Old Fort Bayou Mitigation Bank, Jackson County, Mississippi, was chosen as a translocation site because it will be managed appropriately in perpetuity to reestablish and maintain a longleaf pine community there, and three isolated wetlands were present on site. After some pond restoration work was completed, one of these ponds, designated TNC 1, was selected as our next translocation recipient site.

#### TNC Pond 1

Attempts to translocate dusky gopher frogs from Glen's Pond to TNC Pond 1 began in 2004. This action was intended to establish a new dusky gopher frog population at a currently unoccupied site beyond the distance the frogs would be expected to migrate. TNC Pond 1 is approximately 22 mi (35 km) east of Glen's Pond. Dusky gopher frog eggs from Glen's Pond were hatched in a nearby laboratory and then tadpoles were either released into TNC Pond 1 after 15 days (newly-hatched), 2 to 3 months (head-started) or once metamorphosed (metamorphs). Head-started tadpoles and metamorphs were raised in cattle watering tanks. From 2004 through 2014, a total of 4,867 newly-hatched tadpoles, 3,771 head-started tadpoles and 4,196 recently metamorphosed frogs have been released at TNC Pond 1 during periods when the pond held water for a sufficient duration to allow metamorphosis (Lee 2014). In all but 2 years, the hydroperiod at TNC Pond 1 was long enough to allow translocated dusky gopher frog tadpoles sufficient time to metamorphose. The first signs of adult recruitment were documented in late 2007; one male dusky gopher frog was heard calling and one dusky gopher frog egg mass was found at the pond. Multiple male dusky gopher frogs were heard calling and one egg mass was discovered in spring 2010. Four to five male dusky gopher frogs were heard calling in both 2012 and 2013; however, although multiple late stage dusky gopher frog tadpoles were dip-netted from TNC Pond 1 in spring 2012, no tadpoles were detected in 2013 (Lee 2013). At least five dusky gopher frog egg masses were deposited in 2014 (Lee, pers. comm. 2014). Future efforts at TNC Pond 1 will concentrate on maximizing survival of dusky gopher frogs to sexual maturity.

In accordance with a Cooperative Agreement (CA) between the MDWFP and TNC, gopher frog monitoring and recovery efforts in 2012 consisted of: 1) Monitoring breeding activities at the translocation site [TNC Pond 1; Old Fort Bayou Mitigation Bank (OFBMB)], Mike's and Powerline Ponds; 2) Collecting portions of egg masses (or acquiring recently hatched tadpoles from Glen's Pond researchers) and raising tadpoles to metamorphosis for release into TNC Pond 1, a new translocation site (Mayhaw Pond; Ward Bayou WMA), or at Mike's Pond (if breeding took place there); and 3) Compiling and evaluating data for the year-end report (Lee 2012). An additional component of the CA was to implement habitat management for the dusky gopher frog at TNC Pond 1, Powerline Pond, and Mike's Pond (which are all on TNC property), or on the private property near Glen's Pond. Management activities have included manually cutting

shrubs and small trees, treating cogongrass, and prescribed burning at OFBMB; and treating cogongrass, and prescribed burning at the Mike's Pond property (Lee 2012).

### New Pond

New Pond is a completely man-made pond constructed on DNF approximately 1.5 mi (2.3 km) southeast of Glen's Pond in Harrison County, Mississippi. Partners, including personnel with DNF, NRCS, Harrison County Soil and Water Conservation District, the USFWS, MDWFP, researchers, and non-governmental organizations, designed and built this pond primarily for use as a dusky gopher frog breeding site. It is situated in restored longleaf pine forest and has an active gopher tortoise population nearby. Pony Ranch Pond (See discussion below: **Natural Colonization**) is located between Glen's Pond and New Pond. Translocations of gopher frog tadpoles from Glen's Pond to New Pond have occurred in Spring 2012, Fall 2012, and Spring 2013 (Pechmann and Tupy 2012, 2013). More than 800 dusky gopher frog tadpoles (the majority newly-hatched, some 1 month old, some 6 months old) have been released thus far at New Pond. Frog call surveys and egg mass surveys are being employed to monitor the site. As of the date of this plan, no signs of dusky gopher frog breeding have been detected.

### Sawdust Pond

Sawdust Pond is located on the MSCNWR where we have begun a translocation project using funding from the Cooperative Recovery Initiative (See discussion above: **Management through Partnerships**). In 2015, cattle tanks were setup on the refuge and dusky gopher frog tadpoles from the Glen's Pond population were raised to metamorphosis. By mid-May 2015, more than 250 metamorphic frogs were released at the pond with hundreds more likely to follow. This release effort will be continued over the next several years and Sawdust Pond will be monitored for future dusky gopher frog breeding and adult survival.

## **Natural Colonization**

### Pony Ranch Pond

Pony Ranch Pond is a naturally-occurring ephemeral, hill-top pond located approximately 0.8 mi (1.3 km) southeast of Glen's Pond within DNF. No gopher frog activity had ever been documented at the pond prior to DNF personnel completing a series of restoration actions to improve the pond and surrounding uplands. These actions included the construction of a water retention berm on the south side of the pond to improve the hydroperiod, clearing of shrubs and root mounds in the pond basin, restoration of longleaf pine in surrounding uplands, and prescribed fire in and around the pond. In March 2013, dusky gopher frogs were heard calling at the pond and subsequently three egg masses were discovered during surveys. Seven gopher frogs were captured (two female, five male) exiting the pond by means of a temporary drift fence and box traps (Pechmann and Tupy 2012). Upon examination, identification marks indicated that the two females originated from Glen's Pond. None of the five males bore any previously given mark. Because no dusky gopher frogs have ever been translocated to Pony Ranch Pond, it is believed that these individuals migrated and found the pond on their own.

## **BIOLOGICAL CONSTRAINTS AND NEEDS**

The number of individuals in amphibian populations fluctuates widely because of their susceptibility to biological constraints, especially their dependence on seasonal aquatic habitat. Dusky gopher frogs are no different (Richter *et al.* 2003, Pechmann *et al.* 2012). The stability of populations may depend in part on the species' ability to colonize new sites and maintain connections among extant populations. If natural re-colonization is insufficient, reintroductions may be necessary to maintain natural populations and may require the use of captive-bred stock due to the lack of a large primary population. Suitable habitat for maintenance of existing populations and establishment of new ones must contain certain characteristics if the dusky gopher frog is to survive. Breeding sites must be available in sufficient quality and quantity long enough (greater than 3 months) for metamorphosis to occur. Ponds must be shallow, open, and contain emergent vegetation for egg attachment. Ponds must also dry periodically in order to prevent establishment of aquatic predators and prevent regeneration of hardwood plant species. Upland pine habitat (including habitat necessary to disperse between ponds in a metapopulation) must be adjacent to the breeding ponds. Below-ground habitat such as stumpholes or gopher tortoise burrows must be present for long-term survival of adult dusky gopher frogs. Prescribed fire is an important management tool to ensure long-term optimal habitat quality for both terrestrial and aquatic habitats.

## II. RECOVERY

### Recovery Strategy

The recovery strategy for the dusky gopher frog consists of maintaining and enhancing existing populations on tracts of public and private land; monitoring the status of existing populations; identifying and securing additional dusky gopher frog populations and habitat; establishing new populations through translocations or reintroductions; and supporting research that guides land management and provides demographic and ecological data. Management plans should be developed and implemented for all sites where the dusky gopher frog occurs. Appropriate habitat management includes minimizing soil disturbance and loss of native herbaceous groundcover vegetation; conducting prescribed burning, particularly during the growing season; maintaining open-canopied, grassy ephemeral wetlands; and restoring degraded upland habitat. In addition, management plans should specifically address habitat modifications (e.g., filling of drainage ditches and plow lines, restoring native groundcover flora) necessary to improve and maintain appropriate habitat.

Monitoring programs to track population trends and the response of this species to habitat management activities are needed for all populations. Monitoring programs should be critically evaluated and revised as needed. Since recovery of the dusky gopher frog will necessitate finding or creating new, currently unknown populations, assessment of potentially suitable habitat within the range of the frog and additional presence/absence surveys are needed, especially in Alabama and Louisiana. If no additional dusky gopher frog populations are found, suitable habitat for translocations/reintroductions needs to be identified, and programs developed and implemented to establish and monitor these new populations and manage the habitat that supports them. We expect to conduct a Species Status Assessment (SSA) for the dusky gopher frog in the future and will make revisions to the recovery plan accordingly.

### Recovery Objective

The primary objective of this recovery plan is to prevent the extinction of the dusky gopher frog. The long-term recovery objective is to downlist the dusky gopher frog from endangered to threatened.

### Recovery Criteria

This recovery plan sets forth criteria which, when met, will increase the range of extant dusky gopher frog populations; will increase the number of individuals and populations; and will reduce threats to the species' existence.

#### **We believe downlisting may be considered when the following criteria are met:**

- 1) Six **viable metapopulations\*** are documented within blocks of recovery focus areas (see Figure 10 below) and are widely distributed across the range of the species. The six metapopulations would include a minimum of 12 breeding ponds distributed within the species historic range:
  - a) One metapopulation in Block #1 (Louisiana. Portions of St. Tammany, Tangipahoa, and Washington Parishes, west to the Tangipahoa River);

- b) Two metapopulations each in Block #2 (South-Central Mississippi. North of State Hwy. 26, between the Pearl and Pascagoula Rivers; Forrest County and portions of George, Greene, Jones, Lamar, Marion, Pearl River, Perry, Stone, and Wayne Counties) and Block #3 (South Mississippi. South of Hwy. 26, between the Pearl and Pascagoula Rivers; Hancock and Harrison Counties, and portions of Jackson, George, Pearl River, and Stone Counties); and
  - c) One metapopulation in either Block #4 (Eastern Mississippi. East of Pascagoula/Leaf Rivers; portions of George, Greene, Jackson, Perry, and Wayne Counties) or Block #5 (Alabama. West of the Mobile River Delta; Mobile and Washington Counties, and a small portion of Choctaw County).
- 2) Long-term monitoring (at least 10 years) of each metapopulation documents population viability (viability standard to be defined through a recovery task). The 10-year timeframe will allow monitoring recruitment events and other population attributes in a species that has been characterized by highly variable reproductive and survival rates. In each of at least two annual breeding events within a three-year period, a total of 30 egg masses per metapopulation must be documented and natural recruitment must be verified to document viability.
  - 3) Breeding and adjacent upland habitats within the six metapopulations are protected long-term through management agreements, public ownership, or other means, in sufficient quantity and quality (to be determined by recovery task) to support growing populations.
  - 4) Studies of the dusky gopher frog's biological and ecological requirements have been completed and measures necessary for recovery discovered during these studies are being implemented and are showing progress.

\* A **viable** population is one that is large enough to maintain sufficient genetic variation to enable it to evolve and respond to natural habitat and environmental changes, and exhibits parameters consistent with a stable or increasing reproductive rate, without the addition of frogs raised in artificial environments or introduced from other populations. Viable populations should consist of multiple age classes of individuals, including newly recruited juveniles. In addition, a dusky gopher frog population should be supported by habitat containing breeding ponds and their surrounding uplands. To be a viable population, a dusky gopher frog population must be a **metapopulation**. Breeding ponds within dispersal distance of one another function as a metapopulation; if breeding conditions for the species are not met by certain ponds, the species may persist by breeding at other nearby ponds. For the purposes of this recovery plan, a dusky gopher frog metapopulation will be defined as two or more occupied breeding ponds, individually separated from each other by a mile (1.6 kilometer) or less, within a contiguous area of suitable habitat with no major barriers to dispersal (e.g., major highways, rivers, developed areas, etc.) between ponds.

Defining reasonable delisting criteria is not possible at this time given the current low number of individuals and populations, limited information about the species' biology, and magnitude of threats. Therefore, this recovery plan establishes only downlisting criteria. Criteria will be reevaluated as new information becomes available.

## Recovery Focus Areas

In order to provide our partners with focus areas for implementing recovery efforts, we divided up the range of the dusky gopher frog into blocks. We began this effort by overlaying the historical distribution of the dusky gopher frog compiled from Mount (1975) (Alabama), Lohofener and Altig (1983) (Mississippi), and Dundee and Rossman (1989) (Louisiana) over: historical maps of longleaf pine distribution throughout southern Louisiana, Mississippi, and Alabama; a watershed boundary dataset (Natural Resources Conservation Service HUC12); the National Hydrography dataset; and the range of the listed gopher tortoise (See **HABITAT** discussion, above). Then, we broke up the area into five different blocks to provide a geographical context for recovery efforts and to provide the state boundaries by which recovery efforts will be implemented in many cases (See Figure 10). The geographical area where dusky gopher frogs currently survive encompasses only a small portion of their historical range. Recovery of the species will require their distribution across a broader area. Most of the dusky gopher frog locations currently receiving protection and management are on public lands. However, public lands alone are insufficient to ensure long-term survival of the species.

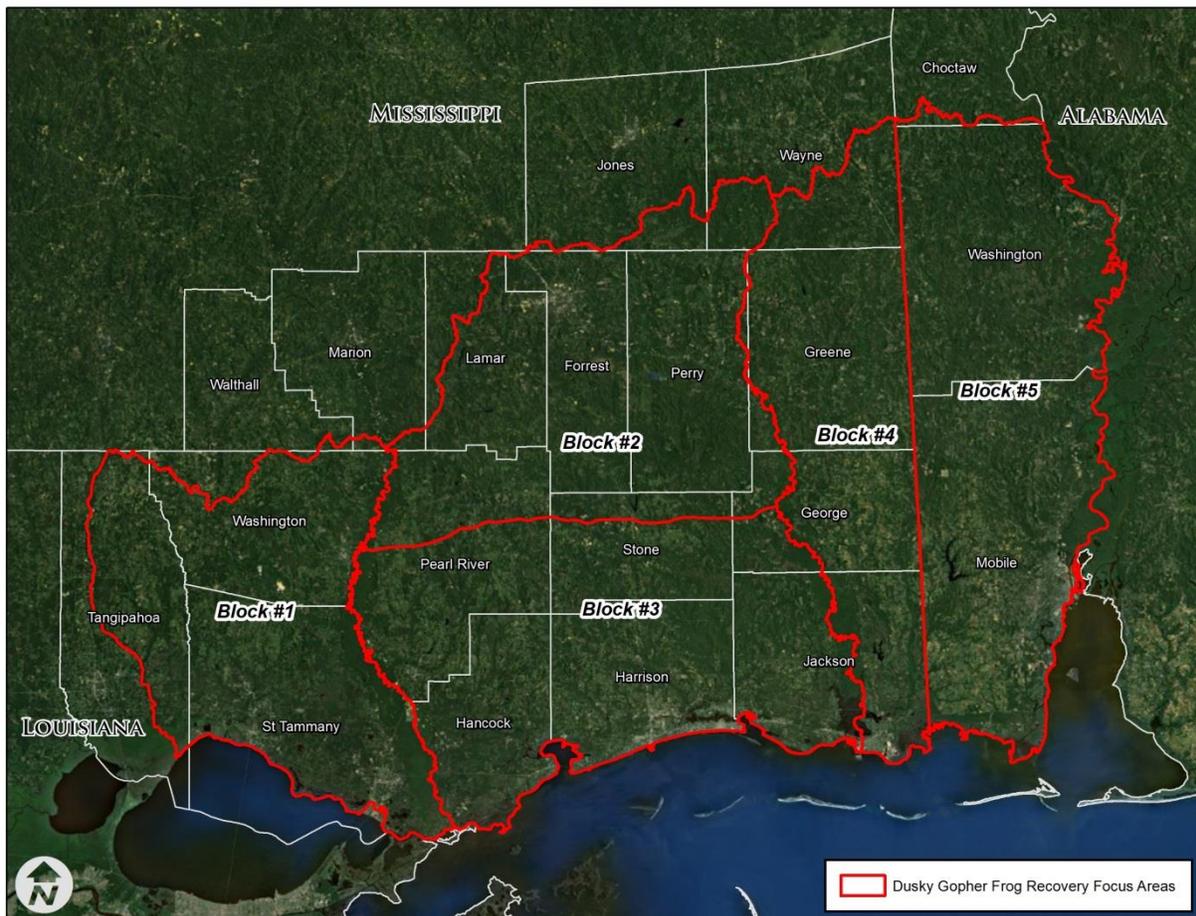


Figure 10. Dusky Gopher Frog Recovery Focus Areas.

**Listing/Recovery Factors Addressed by Recovery Tasks:** Tasks listed below with each listing/recovery factor are examples of actions that may reduce or remove the identified threats. These tasks are described in more detail in the Narrative Outline section that follows.

**Listing/Recovery Factor A: The Present or Threatened Destruction, Modification, or Curtailment of a Species Habitat or Range.** To ensure the long-term recovery needs of the dusky gopher frog and provide adequate assurance of population stability/viability, threats to the species' habitat must be removed or minimized (see Reasons for Listing and Ongoing Threats for a discussion of applicable threats). This can be accomplished by the following actions:

- a) Protect the habitat of existing dusky gopher frog populations on public lands (Task 1.1.1, 1.1.2, and 1.1.3);
- b) Protect the habitat of existing dusky gopher frog populations on private lands (Task 1.2.1, 1.2.2, 1.2.3, 1.2.4, 1.3.1, 1.3.2, 1.4, and 1.5.1 );
- c) Gather baseline data on physical characteristics of both terrestrial and aquatic habitat and monitor habitat conditions for dusky gopher frog populations (Task 2.2);
- d) Evaluate habitat parameters (Task 2.3.1, 2.5, 3.1, 4.1.1, 4.1.2, 4.1.3, 4.1.4, 4.2.2, 4.2.3); and
- e) Study the fire ecology of dusky gopher frog habitat (Task 4.6.1, and 4.6.2).

**Listing/Recovery Factor B: Overutilization for Commercial, Recreational, Scientific, or Educational Purposes.** This is not currently known to be a threat to this frog species. However, we work closely with our partners to protect existing dusky gopher frog populations and Tasks like 1.4, 2.0, 5.0, and 6.0 which call for enforcement of existing regulations, general monitoring and close security of all populations, and maintenance of the captive frog population will help us protect against any possibility of an incident arising.

**Listing/Recovery Factor C: Disease or Predation.** Disease is a threat to this animal that must be monitored for closely. Predation is likely to exist on dusky gopher frogs but has not been fully documented and should also be closely monitored. We will monitor the severity of this listing factor through Tasks 2.1.1, 2.1.2, 2.1.3, 2.2., 2.3.1, 2.3.2, 2.5, 4.5.1, and 4.5.2.

**Listing/Recovery Factor D: The Inadequacy of Existing Regulatory Mechanisms.** Existing regulatory mechanisms protect the species from take but are limited to some extent to protect habitat from threats like conversion to other uses such as development. The following actions can help to overcome these inadequacies and lead to recovery:

- a) Work with public landowners to develop habitat management plans that support dusky gopher frog recovery (Task 1.1.1);
- b) Establish partnerships with private landowners owning dusky gopher frog habitat and develop protective agreements and management plans (Task 1.2.2); and
- c) Work with other government agencies to protect the species (Task 1.4).

**Listing/Recovery Factor E: Other Natural or Manmade Factors Affecting Its Continued Existence.** Multiple factors influencing recovery of the dusky gopher frog are identified under this threat – fire suppression; contaminants like pesticides and/or herbicides; habitat fragmentation; small population size and low reproductive potential; and variability in rainfall. Most of the recovery tasks in the narrative outline will aid in working towards protecting this amphibian from these threats.

## Narrative Outline

1. Protect existing wild dusky gopher frog populations by using appropriate habitat management and other conservation techniques. Management should focus on maintaining, enhancing, and restoring essential components of both the terrestrial and aquatic habitat supporting dusky gopher frogs. Protection of existing populations will be achieved when habitat restoration has been accomplished and long-term commitments for management required for habitat maintenance have been formalized.
  - 1.1 Protect the habitat of existing dusky gopher frog populations on public lands. Eliminate impacts to dusky gopher frogs and their habitats on public lands where the species occurs.
    - 1.1.1 Work with public landowners to develop habitat management plans that support dusky gopher frog recovery. Within plans, include specifics on forestry practices, such as prescribed burning and techniques to maintain an open canopy forest, that when used result in quality dusky gopher frog habitat. Incorporate any beneficial management and restoration actions discovered during research (under 4.0).
    - 1.1.2 Implement habitat management plans to benefit dusky gopher frog populations on public land.
    - 1.1.3 Revise public lands habitat management plans as new information is obtained from monitoring and research (see 2.5).
  - 1.2 Protect dusky gopher frog habitat on private lands. Develop mechanisms and incentives to help facilitate habitat protection on private lands. Public ownership of dusky gopher frog habitat is insufficient to recover the species, especially in Alabama and Louisiana. Cooperation of private landowners is essential for recovery.
    - 1.2.1 Adopt and implement habitat management practices, developed under 1.1.1, on private land where dusky gopher frog populations exist, whenever possible.

- 1.2.2 Establish partnerships with private landowners owning dusky gopher frog habitat and develop protective agreements and management plans.
- 1.2.3 Identify programs, financial incentives, and other opportunities to assist private landowners with dusky gopher frog habitat protection and management.
- 1.2.4 Revise private lands habitat management plans as new information is obtained from monitoring and research (see 2.5).
- 1.3 Connect dusky gopher frog habitat protection efforts with initiatives that protect or reestablish other species endemic to the longleaf pine ecosystem (e.g., gopher tortoises).
  - 1.3.1 Identify initiatives that protect or reestablish other longleaf pine endemic species within the range of the dusky gopher frog and evaluate cooperative opportunities that benefit its recovery.
  - 1.3.2 Implement efforts that connect dusky gopher frog recovery with other efforts for longleaf pine endemic species.
- 1.4 Utilize existing regulations to promote dusky gopher frog recovery. Using these regulations, minimize adverse effects of proposed actions to the frog and its habitat, and maximize proposed actions to enhance recovery of the species. Implement by working with our Federal partners through use of the Act's Section 7 consultation process and by expanding other existing programs to further support recovery of the dusky gopher frog such as those utilized by the USDA/Forest Service and Natural Resources Conservation Service.
- 1.5 Minimize impact of development. The location of known dusky gopher frog populations should be considered when development planning occurs.
  - 1.5.1 Protect known and potential dusky gopher frog breeding sites, and their surrounding uplands, from the impacts of development.
- 2. Monitor known dusky gopher frog populations and the habitat that supports them.
  - 2.1 Gather baseline population data. Baseline population data are needed to assess the effectiveness of management efforts. Trapping frogs at drift fences that completely enclose breeding sites is currently the most effective means of tracking population fluctuations and monitoring recruitment. Automated recorders are tools that can document presence/absence of calling males. Egg mass surveys can be conducted to verify reproduction. Dipnets and funnel traps can be used to monitor the presence or absence of larvae at sites. Survey work should be conducted according to established biosecurity protocols to avoid transmission of pathogens between sampling sites.

- 2.1.1 Monitor the Glen's Pond population annually using egg mass surveys and a drift fence until such time as it is determined this level of effort is no longer needed. Monitoring will generate demographic data including population size, number and survivorship of breeding adults, breeding success, and juvenile recruitment.
- 2.1.2 Monitor the Mike's Pond population annually and determine if and when translocations of frogs are required to maintain this population or if the population should be considered extinct. Automated frog call recording devices, egg mass surveys, and dipnetting will be used to provide data on calling adult males, breeding success (including an estimate of adult females), and hatching success. Use of a drift fence will provide information on recruitment; if cost prohibitive, another technique will be needed to measure recruitment (see 4.7).
- 2.1.3 Initiate long-term monitoring of translocated recovery populations across the geographic range. Automated frog call recording devices and egg mass surveys can be used to verify adult survival and identify successful breeding events. Use of a drift fence will provide information on recruitment; if cost prohibitive, another technique will be needed to measure recruitment (see 4.7).
- 2.2 Gather baseline data on physical characteristics of both terrestrial and aquatic habitats and monitor habitat conditions for all dusky gopher frog populations. Initiate data collection describing known breeding sites and their surrounding uplands. Collect data relative to annual hydroperiod (include water depth gauge at pond center); pH of pond water; vegetation in ponds and uplands (document increase/decrease in native grasses and forbs, and increase/decrease in shrub cover); available belowground habitat (e.g. abundance of stump holes; presence of gopher tortoises); soils; fire history and other past land management practices; and upland and wetland habitat disturbance. Collect data on the response of vegetation to various management regimes or techniques. Look for relationships between population viability and environmental factors (natural or human-induced).
- 2.3 Assess and reduce threats to recovery populations and their habitats.
  - 2.3.1 Assess existing threats to populations using known techniques. Conduct species evaluation by assessing habitat quality and other factors affecting population growth and recovery.
  - 2.3.2 Develop and implement a strategy to assess dusky gopher frog population health and implement appropriate measures to remove health threats. Monitor the level of parasites and disease in recovery populations over time to ensure that threats have been removed or are under control.

- 2.4 Coordinate population monitoring. Monitoring programs should be periodically evaluated and revised as needed.
  - 2.4.1 Establish a centralized database for range-wide population data storage, analysis, and recovery review. Base maps and baseline data should be maintained for all populations.
- 2.5 Monitor the results of population-specific dusky gopher frog management plans at specified intervals to determine if management goals are being met and threats have been addressed. Amend population management practices as needed; amending habitat management plans is addressed under 1.1.3 and 1.2.4.
3. Continue searches for additional dusky gopher frog populations throughout their historical range. Although there have been intensive searches for this species, potentially suitable habitat remains to be investigated. Future survey efforts of these areas, especially those in Alabama and Louisiana, should be conducted on public lands and on private land whenever landowner permission is granted. Surveys of private lands are especially critical in recovery focus areas where public land is insufficient to meet recovery goals. Survey work should be conducted according to established biosecurity protocols, such as those developed by the U.S. Geological Survey (<http://fl.biology.usgs.gov/armi/Biosecurity/biosecurity.html>), to avoid transmission of pathogens between sampling sites.
  - 3.1 Develop and implement a standardized protocol for identifying potentially suitable dusky gopher frog habitat. Identify ownership of potentially suitable sites and request permission to conduct surveys.
  - 3.2 Survey for dusky gopher frog populations in potentially suitable habitat. Tracts of habitat on public land, or lands under some sort of conservation management should be prioritized for surveys. If no new dusky gopher frog populations are found, identify localities where the species could be translocated/reintroduced.
  - 3.3 Use GIS to collect and store data on available dusky gopher frog habitat.
    - 3.3.1 Develop a GIS database of actual/potential dusky gopher frog habitat by state and recovery focus area. Build on established databases in each state and make the GIS database compatible with the centralized recovery monitoring database (Task 2.4.1.).
    - 3.3.2 Ground-truth the GIS database.
4. Conduct research. Much remains to be learned about the basic ecology and demography of the dusky gopher frog. Elements critical to the perpetuation of populations need to be determined to ensure long-term survival for the species. In order to address most of the

following research topics, long-term studies will be necessary. Variation in life history characteristics resulting from factors such as failure to breed every year, low recruitment in years with breeding, drought that interferes with breeding, etc., insure that meaningful data can be obtained only by conducting research over multiple years with varying environmental conditions.

- 4.1 Conduct dusky gopher frog studies to provide data for a population and habitat viability analysis (PHVA).
  - 4.1.1 Conduct demographic studies. Determine the population structure, estimate the proportion of females breeding in a given year, and determine the probability of an egg becoming a metamorphosed adult.
  - 4.1.2 Develop techniques and conduct studies to determine the dispersal patterns of adults and metamorphic frogs to, and from, breeding sites and to describe preferred upland microhabitat within underground refugia.
  - 4.1.3 Using data on preferred upland microhabitat obtained through 4.1.2, explore and implement ways to provide more subterranean refugia within habitat of dusky gopher frog recovery populations.
  - 4.1.4 Determine the landscape configuration that would support a viable metapopulation. Identify the distribution of ponds such that dusky gopher frogs can move between them and create multiple breeding sites.
- 4.2 Use the results of the population and habitat studies, described under 4.1.1 through 4.1.3, to conduct a PHVA analysis and amend population-specific management plans, as necessary.
  - 4.2.1 Estimate the minimum number of individuals necessary to sustain a viable metapopulation using PHVA.
  - 4.2.2 Estimate the amount of aquatic habitat needed to maintain a viable metapopulation using PHVA.
  - 4.2.3 Estimate the amount of terrestrial habitat needed to maintain a viable metapopulation using PHVA.
  - 4.2.4 Revise population-specific management plans using PHVA results, as necessary.
- 4.3 Employ cattle tanks for raising dusky gopher frog tadpoles to metamorphosis as a method to protect against catastrophic loss and to provide frogs for translocation. Continue to assess the results of using this technique and modify as necessary.

- 4.4 Assess and preserve genetic variability in wild dusky gopher frog populations.
  - 4.4.1 Conduct population genetic analysis using 5 eggs removed from each clutch per dusky gopher frog breeding event.
  - 4.4.2 Preserve genetic variability in wild populations by crossing Glen's Pond and Mike's Pond individuals, when possible and determined to be necessary. Introduce offspring from these crosses into both parental populations and at translocation sites.
  - 4.4.3 Monitor genetic variability of newly translocated populations, and of current populations, following expansion from single to multiple breeding ponds (i.e., as metapopulations are established).
- 4.5 Study the interaction of dusky gopher frogs with other animals that may be predators and/or competitors.
  - 4.5.1 Identify the significant invertebrate and/or vertebrate predators of larval and adult dusky gopher frogs; determine if any predator control actions are needed.
  - 4.5.2 Identify the significant competitors of larval and adult dusky gopher frogs; determine if any competitor control actions are needed.
- 4.6 Study the fire ecology of dusky gopher frog habitat.
  - 4.6.1 Determine the importance of fire seasonality in maintaining physical and chemical aspects of both upland and pond habitat. Investigate the effect of fire on the water chemistry of breeding ponds and how this might impact the dusky gopher frog.
  - 4.6.2 Determine the direct effects of fire on migrating dusky gopher frogs.
- 4.7 Identify what adult and larval dusky gopher frogs eat and how these dietary needs affect population growth and survival.
- 4.8 Investigate the development of a potentially less disruptive and more cost effective technique than using a drift fence to document and measure recruitment in dusky gopher frog populations.
- 5. Revise and implement guidelines for using translocations to establish dusky gopher frog populations within recovery focus areas, as needed to meet recovery goals.
  - 5.1 Determine the appropriate life stage to use in translocation/reintroduction.
  - 5.2 Identify essential habitat conditions for translocation/reintroduction sites.

- 5.3 Identify sites with the appropriate habitat conditions for translocation/reintroduction. Initial efforts should focus on areas designated as critical habitat for the species.
- 5.4 Identify methods that the Service and its partners can take to protect sites selected for translocation. Initiate translocations only once sites are protected through use of these methods; some potential methods include public ownership, conservation easements, and interagency agreements.
- 5.5 Translocate/reintroduce dusky gopher frogs as needed to meet population recovery goals.
6. Develop and implement a controlled propagation and reintroduction plan (CPRP) to facilitate maintenance and use of captive dusky gopher frog populations in support of recovery goals. Use the existing Species Survival Plan (SSP) as a model.
  - 6.1 Maintain and manage captive populations to reflect the genetics of wild populations and provide protection against extinction and loss of genetic diversity. Use the protocol established by the Dusky Gopher Frog Recovery Team (See Appendix D) until such time that a more extensive manual can be produced (See Task 6.3), and the AZA dusky gopher frog studbook. Complete a DNA analysis of captive dusky gopher frog populations.
  - 6.2 Use experimental trials of different environmental manipulations to develop a technique to induce captive dusky gopher frog reproduction without in vitro fertilization.
  - 6.3 Develop a comprehensive captive husbandry and breeding manual for the maintenance of genetic diversity in the captive population. Include techniques to reduce mortality and loss of founders, and to maximize reproductive success. This manual can be included as an Appendix within the CPRP.
  - 6.4 Conduct a pilot project testing the feasibility of using captive-bred progeny to establish recovery populations using the protocol established by the Dusky Gopher Frog Recovery Team (See Appendix B).
  - 6.5 Revise the “Use of Captive-bred Progeny in Translocation/Reintroduction Efforts” protocol (Appendix B), as necessary, after analyzing the results of the pilot project.
  - 6.6 Finalize a CPRP for the dusky gopher frog as necessary based on new information.
7. Develop and distribute public educational and informational materials/programs to solicit and promote voluntary stewardship to support dusky gopher frog recovery.

- 7.1 Develop fact sheets and other tools with our recovery partners and distribute them to the public to provide important habitat and management information. Include information on prescribed burning, protection of soil and groundcover, and the importance of restoring longleaf pine. Incorporate dusky gopher frog information into DeSoto National Forest educational materials and other agency and conservation group materials as appropriate.
  - 7.2 Institute and maintain proactive public outreach. Pursue outreach opportunities, especially in Alabama and Louisiana where private landowners may play an important role in achieving dusky gopher frog recovery goals.
  - 7.3 Involve the local community in dusky gopher frog recovery projects, whenever possible. Contact local experts in herpetology, hydrology, and forestry and invite them to participate in recovery activities. Expand relationships with local environmental friends groups.
8. Review/evaluate recovery progress.
    - 8.1 Review results of monitoring, survey efforts, and research to determine success of recovery efforts. Conduct annual recovery meetings to review recovery progress.
    - 8.2 Adapt population-specific management plans as necessary based on revised recovery goals. Management plans need to have built-in flexibility in order to incorporate findings of ongoing monitoring and research.
    - 8.3 Revise recovery criteria, as needed, utilizing the SSA framework.

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### III. RECOVERY IMPLEMENTATION SCHEDULE:

The following implementation schedule outlines recovery tasks and their estimated costs. It is a guide for meeting the objectives described in Part II of this plan. This schedule indicates task priorities, task numbers, task descriptions, duration of tasks, the responsibility of agencies, and estimated costs. It should be noted that these are minimum estimates of the costs associated with recovery of the dusky gopher frog.

Priorities in column one of the following Implementation Schedule are assigned as follows:

1. An action that must be taken to prevent extinction or to prevent the species from declining irreversibly in the foreseeable future.
2. An action that must be taken to prevent a significant decline in the species' population, habitat quality, or some other significant negative impact short of extinction.
3. All other actions necessary to meet the recovery objective.

#### Key to acronyms used in the Implementation Schedule

ADCNR	-Alabama Department of Conservation and Natural Resources
COE	- U.S. Army Corps of Engineers
DOD	- Camp Shelby
LAND	- Other private landowners, land trusts, etc.
LDWF	- Louisiana Department of Wildlife and Fisheries
MDWFP	- Mississippi Department of Wildlife, Fisheries, and Parks
MFC	- Mississippi Forestry Commission
MZ	- Memphis Zoo and other partners in Association of Zoos and Aquariums
NRCS	- Natural Resources Conservation Service
RESEARCH	- Independent researchers
TNC	- The Nature Conservancy
USFWS	- U.S. Fish and Wildlife Service
USFS	- U.S. Forest Service
USGS	- U.S. Geological Survey

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	1.1.1	Work with public landowners to develop habitat management plans that support dusky gopher frog recovery.	2 years	COE DOD MDWFP MFC USFS USFWS						Cost included in 1.1.2
1	1.1.2	Implement habitat management plans to benefit dusky gopher frog populations on public lands.	Continuous	COE DOD MDWFP MFC USFS USFWS	50	50	50	50	50	Additional costs will be covered under existing programs.
1	1.2.1	Adopt and implement habitat management practices, developed under 1.1.1, on private land where dusky gopher frog populations exist whenever possible.	Continuous	ADCNR LAND LDWF MDWFP TNC USFWS	50	50	50	50	50	Additional costs will be covered under existing programs.
1	1.2.2	Establish partnerships with private landowners owning dusky gopher frog habitat and develop protective agreements and management plans.	At least 5 years.	ADCNR LAND LDWF MDWFP NRCS TNC USFWS						Expenditures covered under existing program costs.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	1.2.3	Identify programs, financial incentives, and other opportunities to assist private landowners with dusky gopher frog habitat protection and management.	Continuous	ADCNR LAND LDWF MDWFP NRCS TNC USFWS						Expenditures covered under existing program costs.
1	1.4	Utilize existing regulations to promote dusky gopher frog recovery.	Continuous	ADCNR LDWF MDWFP NRCS USFS USFWS						Expenditures covered under existing program costs.
1	1.5.1	Protect known and potential dusky gopher frog breeding sites, and their surrounding uplands, from development.	Continuous	ADCNR LDWF MDWFP USFS USFWS						Expenditures covered under existing program costs.
1	2.1.1	Monitor the Glen's Pond population annually using egg mass surveys and a drift fence until such time as it is determined this level of effort is no longer needed.	Continuous	MDWFP RESEARCH USFS USFWS	80	80	80	80	80	

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

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TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	2.1.2	Monitor the Mike's Pond population annually and determine if and when translocations of frogs are required to maintain this population or if the population should be considered extinct.	Continuous	MDWFP RESEARCH TNC USFWS	45	45	45	45	45	
1	2.1.3	Initiate long-term monitoring of translocated recovery populations across the geographic range.	Continuous	ADCNR COE LDWF MDWFP TNC USFS USFWS USGS	80	80	80	120	120	Costs increase as more populations are established and monitoring increases.
1	2.2	Gather baseline data on physical characteristics of both terrestrial and aquatic habitats and monitor habitat conditions for all dusky gopher frog populations.	Continuous	ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS USGS	25	25	25	25	25	Most of cost included in 2.1.1, 2.1.2, and 2.1.3.
1	2.3.1	Assess existing threats to populations using known techniques.	Continuous	MDWFP RESEARCH TNC USFS USFWS						Cost covered under 2.1.1, 2.1.2, and 2.1.3.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	2.3.2	Develop and implement a strategy to assess dusky gopher frog population health and implement appropriate measures to remove health threats.	Continuous	ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS	25	25	10	5	5	
1	2.5	Monitor the results of population-specific management plans at specified intervals to determine if management goals are being met and threats have been addressed.	Continuous	ADCNR LAND LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
1	3.1	Develop and implement a standardized protocol for identifying potentially suitable dusky gopher frog habitat.	2 years	ADCNR LDWF MDWFP RESEARCH USFS USFWS USGS	20	20				Incorporate into GIS database (Task 3.3.1).
1	3.2	Survey for dusky gopher frog populations in potentially suitable habitat.	3 years	ADCNR LDWF MDWFP RESEARCH USFWS USGS			40	40	40	Incorporate into GIS database (Task 3.3.1).



**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	4.1.1	Conduct demographic studies.	4 years	ADCNR LDWF MDWFP RESEARCH USFWS USGS	80	80	80	80		Coordinate effort with 2.1.1, 2.1.2; 2.1.3; part of cost could be shared.
1	4.1.2	Develop techniques and conduct studies to determine the dispersal patterns of adults and metamorphic frogs to, and from, breeding sites and to describe preferred upland microhabitat within underground refugia.	3 years	MDWFP RESEARCH USFWS	30	30	30			Coordinate effort with 2.1.1, 2.1.2; 2.1.3; part of cost could be shared.
1	4.1.3	Using data on preferred upland microhabitat obtained through 4.1.2, explore and implement ways to provide more subterranean refugia within habitat of dusky gopher frog recovery populations.	2 years	MDWFP RESEARCH TNC USFWS						Expenditures covered under existing program costs.

<b>IMPLEMENTATION SCHEDULE</b>										
<b>Dusky Gopher Frog Recovery Plan</b>					<b>U.S. Fish and Wildlife Service</b>					
TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	4.1.4	Determine the landscape configuration that would support a viable metapopulation.	1 year	MDWFP RESEARCH USFWS						Expenditures covered under existing program costs.
1	4.2.1	Estimate the minimum number of individuals necessary to sustain a viable metapopulation using PHVA.	1 year	MDWFP RESEARCH USFWS					50	PHVA analysis is dependent on completion of demographic studies (Task 4.1.1).
1	4.2.2	Estimate the amount of aquatic habitat needed to maintain a viable metapopulation using PHVA.	1 year	MDWFP RESEARCH USFWS						Cost coincident with 4.2.1.
1	4.2.3	Estimate the amount terrestrial habitat needed to maintain a viable metapopulation using PHVA.	1 year	MDWFP RESEARCH USFWS						Cost coincident with 4.2.1.
1	4.2.4	Revise population-specific management plans using PHVA results, as necessary.	As necessary	ADCNR LAND LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	4.3	Employ, and continue to assess the results of using, cattle tanks for raising dusky gopher frog tadpoles to metamorphosis as a method to protect against catastrophic loss and to provide frogs for translocation.	Continuous	MDWFP RESEARCH USFWS	20	20	20	20	20	Cost covered under 2.1.1.
1	4.4.1	Conduct population genetic analysis using 5 eggs removed from each clutch per dusky gopher frog breeding event.	Continuous	MDWFP RESEARCH USFWS	5		5		5	
1	4.4.2	Preserve genetic variability in wild populations by crossing Glen's Pond and Mike's Pond individuals, when possible and determined to be necessary.	As necessary	MDWFP RESEARCH USFWS						Cost covered under 2.1.1 and 2.1.2.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
1	4.4.3	Monitor genetic variability of newly translocated populations, and of current populations, following expansion from single to multiple breeding ponds (i.e., as metapopulations are established).	As necessary	ADCNR LDWF MDWFP RESEARCH USFWS						Cost covered under 4.4.1.
1	6.1	Maintain and manage captive populations to reflect the genetics of wild populations and provide protection against extinction and loss of genetic diversity.	2 years	MZ RESEARCH USFWS						Expenditures covered under existing program costs.
2	1.1.3	Revise public lands habitat management plans as new information is obtained from monitoring and research.	As necessary	ADCNR LDWF MDWFP RESEARCH USFS USFWS						Cost covered under 1.1.2.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
2	1.2.4	Revise private lands habitat management plans as new information is obtained from monitoring and research.	As necessary	ADCNR LAND LDWF MDWFP RESEARCH TNC USFWS						Cost covered under 1.2.2.
2	1.3.1	Identify initiatives that protect or reestablish other longleaf pine endemic species within the range of the dusky gopher frog and evaluate cooperative opportunities that benefit its recovery.	Continuous	ADCNR LDWF MDWFP NRCS RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
2	1.3.2	Implement efforts that connect dusky gopher frog recovery with other efforts for longleaf pine endemic species.	Continuous	ADCNR LDWF MDWFP NRCS RESEARCH TNC USFS USFWS						Cost unknown at this time.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

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TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
2	3.3.1	Develop a GIS database of actual/potential dusky gopher frog habitat by state and recovery focus area.	2 years	ADCNR LDWF MDWFP RESEARCH USFS USFWS						Expenditures covered under existing program costs.
2	3.3.2	Ground-truth the GIS database.	1 year	ADCNR LDWF MDWFP RESEARCH USFS USFWS			50			
2	4.6.1	Determine the importance of fire seasonality in maintaining physical and chemical aspects of both upland and pond habitat.	3 years	MDWFP RESEARCH USFS USFWS	50	50	50			
2	4.6.2	Determine the direct effects of fire on migrating dusky gopher frogs.	Continuous	MDWFP RESEARCH USFS USFWS						Costs covered under 2.1.1, 2.1.2, 2.1.3.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

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TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
2	4.7	Identify what adult and larval dusky gopher frogs eat and how these dietary needs affect population growth and survival.	3 years	MDWFP RESEARCH USFWS	30	30	30			
2	4.8	Investigate the development of a potentially less disruptive and more cost effective technique than using a drift fence to document and measure recruitment in dusky gopher frog populations.	3 years	MDWFP RESEARCH USFWS	50	50	50			
2	5.1	Determine the appropriate life stage to use in translocation /reintroductions.	3 years	MDWFP RESEARCH TNC USFWS USGS	50	50	50			
2	5.2	Identify essential habitat conditions for translocation /reintroduction sites.	3 years	ADCNR LDWF MDWFP RESEARCH USFS USFWS USGS						5.1 and 5.2 can be conducted concurrently and costs shared.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
2	5.3	Identify sites with the appropriate habitat conditions for translocation /reintroduction.	2 years	ADCNR LDWF MDWFP RESEARCH USFS USFWS USGS				25	25	Use GIS database developed under 3.3.1.
2	5.4	Identify methods that the Service and its partners can take to protect sites selected for translocation.	As needed	ADCNR LDWF MDWFP RESEARCH USFS USFWS USGS						Expenditures covered under existing program costs.
2	5.5	Translocate/reintroduce dusky gopher frogs as needed to meet population recovery goals.	As necessary	ADCNR LDWF MDWFP RESEARCH USFS USFWS	50	50	50	50	50	
2	6.2	Use experimental trials of different environmental manipulations to develop a technique to induce captive dusky gopher frog reproduction without in vitro fertilization.	2 years	MZ RESEARCH USFWS						Expenditures covered under existing program costs.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
2	6.3	Develop a comprehensive captive husbandry and breeding manual for the maintenance of genetic diversity in the captive population.	2 years	MZ						Expenditures covered under existing program costs.
2	6.4	Conduct a pilot project testing the feasibility of using captive-bred progeny to establish recovery populations using the protocol established by the Dusky Gopher Frog Recovery Team (See Appendix B).	3 years	ADCNR LDWF MDWFP MZ RESEARCH USFWS	50	30	30			
2	6.5	Revise the “Use of Captive-bred Progeny in Translocation/ Reintroduction Efforts” protocol (Appendix B), as necessary, after analyzing results of the pilot project.	1 year	ADCNR LDWF MDWFP MZ RESEARCH USFWS						Expenditures covered under existing program costs.
2	6.6	Finalize a CPRP for the dusky gopher frog as necessary based on new information.	1 year	ADCNR LDWF MDWFP MZ RESEARCH USFWS						Expenditures covered under existing program costs.

**IMPLEMENTATION SCHEDULE**

**Dusky Gopher Frog Recovery Plan**

**U.S. Fish and Wildlife Service**

TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
3	2.4.1	Establish a centralized database for range-wide population data storage, analysis, and recovery review.	Continuous	ADCNR LDWF MDWFP RESEARCH USFS USFWS	5	5	5	5	5	
3	4.5.1	Identify the significant invertebrate and/or vertebrate predators of larval and adult dusky gopher frogs; determine if any predator control actions are needed.	2 years	RESEARCH USFWS	15	15	15			
3	4.5.2	Identify the significant competitors of larval and adult dusky gopher frogs; determine if any competitor control actions are needed.	2 years	RESEARCH USFWS						Cost covered under 4.5.1.
3	7.1	Develop Fact Sheets and other tools with our recovery partners and distribute them to the public to provide important habitat and management information.	2 years	ADCNR LDWF MDWFP MZ RESEARCH TNC USFS USFWS USGS	5	5				

IMPLEMENTATION SCHEDULE										
Dusky Gopher Frog Recovery Plan						U.S. Fish and Wildlife Service				
TASK PRIORITY	TASK NUMBER	TASK DESCRIPTION	TASK DURATION	PARTICIPANTS	COST ESTIMATES (\$K)					COMMENTS/NOTES
					FY 1	FY 2	FY 3	FY 4	FY 5	
3	7.2	Institute and maintain proactive public outreach.		ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
3	7.3	Involve the local community in dusky gopher frog recovery projects, whenever possible.		ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
3	8.1	Review results of monitoring, survey efforts, and research to determine success of recovery efforts.	Every 3 years	ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
3	8.2	Adapt population-specific management plans as necessary based on revised recovery goals.	As necessary	ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.
3	8.3	Revise recovery criteria, as needed, utilizing the SSA framework.	As necessary	ADCNR LDWF MDWFP RESEARCH TNC USFS USFWS						Expenditures covered under existing program costs.

## V. APPENDICES

### Appendix A. List of Stakeholders/Recovery Team Members (\*\*)/Invited Peer Reviewers (<sup>PR</sup>)

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## Appendix B. Summary of Peer Review

Certain minor editorial suggestions were incorporated into the recovery plan and are not addressed here.

### Section: **EXECUTIVE SUMMARY AND RECOVERY**

Comment: One reviewer suggested that we add “natural” before recruitment in our second recovery criterion to clarify that we were not suggesting augmenting populations using captive animals or frogs from other populations.

Response: We have made this change in the text of the recovery plan.

Comment: One reviewer stated that our fourth recovery criterion where we state that “(S)udies of the dusky gopher frog’s biological and ecological requirements have been completed” is contradicted by Task 4 in the Narrative Outline where we state that “(M)uch remains to be learned about the basic ecology and demography of the dusky gopher frog.”

Response: The recovery criteria, as described in the Executive Summary, are those actions that when met will prevent the extinction of the dusky gopher frog. The Narrative Outline describes in detail those tasks necessary to achieve the recovery criteria defined in the Executive Summary. In other words, the recovery criteria define what is needed for recovery and the recovery tasks describe what is needed to accomplish these recovery goals. Thus, the two statements complement each other and are not contradictory.

Comment: Two reviewers were uncertain that a metapopulation supported by two breeding ponds could be considered viable long-term. One of these reviewers suggested that using 30 as the number of egg masses for a metapopulation seemed low.

Response: We agree that a metapopulation with three ponds and more than 30 egg masses over the period we defined would likely provide additional protection from threats to a population. However, our current recovery criteria are for downlisting the dusky gopher frog to threatened and we believe the viable population as currently defined will provide long-term stability to the extent that the threat of extinction is removed.

### Section: **STATUS**

Comment: One reviewer suggested we clarify that The Nature Conservancy is a nonprofit, non-governmental organization dedicated to land and water conservation.

Response: We added this language to the recovery plan.

### Section: **LIFE HISTORY AND ECOLOGY**

Comment: One reviewer suggested that we add “during their lifetime” to statement that frogs breed, on average, only one to two seasons.

Response: We added this text to the recovery plan.

### Section: **HABITAT**

Comment: One reviewer suggested that we change “created” to “maintained” in our discussion of the use of seasonally-appropriate prescribed fire and optimal habitat and add “native” to groundcover description.

Response: These changes have been made in the recovery plan. We use “created” when referring to fire used to improve habitat conditions at potential translocation sites.

Comment: One reviewer pointed out that we were using the incorrect common name for *Peromyscus gossypinus*.

Response: We have corrected this error and changed Oldfield mouse to cotton mouse.

## Section: **RECOVERY**

### **Narrative Outline**

Comment: One reviewer asked if public ownership of dusky gopher frog habitat is insufficient to recover the species in Louisiana (as stated in Task 1.2: “Protect the habitat of existing dusky gopher frog populations on private land”), is it feasible to expect a metapopulation can be established there, and if not, does this mean the species will never be considered recovered?

Response: We have changed the wording of Task 1.2 by removing the word “existing”. We do believe that cooperation of private landowners is essential for recovery of the dusky gopher frog in Alabama and Louisiana. Our intention is to establish partnerships with private landowners in these two states and use existing programs to protect and manage suitable habitat for the purpose of establishing dusky gopher frog recovery populations.

Comment: One reviewer noted that within Task 2.1.1 we did not provide an endpoint for monitoring of Glen’s Pond and suggested that we add text to indicate monitoring could be curtailed when it was no longer needed.

Response: We modified Task 2.1.1 to reflect this addition.

Comment: One reviewer noted that within Task 2.1.2 we did not address the rarity of frogs at Mike’s Pond and asked at what point monitoring should be discontinued if no additional dusky gopher frogs are introduced to the pond.

Response: Maintaining a dusky gopher frog population at the Mike’s Pond site is an important aspect of achieving recovery goals for the dusky gopher frog. However, we acknowledge that we may reach a point where it is determined that this population cannot be saved. We amended Task 2.1.2 to include determining if and when additional frogs need to be translocated to this site for population maintenance or if the population should be considered extinct.

Comment: Two reviewers suggested we add a task to the recovery plan that would identify ways to provide more subterranean refugia within dusky gopher frog habitat.

Response: We added Task 4.1.3 to reflect this addition to the recovery plan.

Comment: One reviewer asked about Task 4.4.2 and whether it was still possible to preserve the genetic variability from the Mike’s Pond population, and if there is any way to genetically differentiate between individuals from Glen’s Pond and Mike’s Pond.

Response: We added “when possible” to this task to reflect the uncertainty of pure Mike’s Pond individuals remaining in that population due to past crosses with females from Glen’s Pond. Genetic data from each population has been collected previously so it would be possible to determine the extent of hybridization between the two populations, if tissue samples from Mike’s Pond frogs are collected. Our goal is to preserve as much dusky gopher frog genetic diversity as possible.

Comment: One reviewer questioned whether predator and/or competitor control actions will be taken to reduce their numbers if they are documented through implementing Task 4.5 in the Narrative Outline.

Response: We modified Task 4.5.1 and Task 4.5.2 to include determining if predator and/or competitor control actions are needed.

Comment: One reviewer suggested that language be added to Task 4.8 to indicate that a technique be investigated that is less disruptive, as well as more cost effective, than a drift fence for documenting and measuring recruitment in dusky gopher frog populations.

Response: We modified Task 4.8 to include this change.

Comment: One reviewer questioned why local experts would be limited to academics in Task 7.3 which concerns involving the local community in dusky gopher frog recovery projects.

Response: We have amended Task 7.3 by using the phrase “local experts in herpetology, hydrology, and forestry”.



## **Appendix C. Protocol for Maintenance of Captive Dusky Gopher Populations and Use of Captive-bred Progeny in Translocation/Reintroduction Efforts**

Captive dusky gopher frog populations must be managed to reflect the genetic diversity of wild populations. The genetic relationships between all individual wild-caught frogs in captivity have been determined. The current Species Survival Plan (SSP) (Reichling and Schad 2012) assigns pairings based on an assumption of no relatedness between wild caught specimens. A more accurate means of preserving existing genetic diversity will be enabled by revising the SSP breeding recommendations according to the genetic relatedness indicated by DNA analysis (see Recovery Tasks 4.4.1 and 6.1).

To manage genetic diversity, individual frogs must be permanently and individually marked. A requirement that all SSP participants mark all frogs with implanted passive-integrated transponders will be a component of the revised SSP.

To monitor declines in captive genetic diversity due to genetic drift, loss of founders, or imbalances in founder representation, periodic reanalysis of the population must be done. A genetic reanalysis of the captive population should be funded and executed on a 5-year schedule. When genetic diversity has declined by 10% from the last assessment, the acquisition of additional individuals from the in situ population should proceed under the direction of the U.S. Fish and Wildlife Service.

Assisted Reproductive Technologies (ART) currently provide the only means of breeding specific frogs that are chosen for their genetic pairing. A written protocol for ART is currently included as an Appendix to the SSP. The effective application of ART at all holding institutions requires funded on-site instruction and assistance from personnel who are applying the methodology successfully.

Attempts to establish in situ populations using captive-bred dusky gopher frogs should only be made if the following criteria are met:

- 1) The reintroduction area is within the historical range of the dusky gopher frog;
- 2) The habitat appears to be suitable and is under long-term protection;
- 3) The specific habitat alterations and threats that led to the dusky gopher frog's extirpation have been determined and eliminated or minimized;
- 4) No reproducing populations of dusky gopher frog remain in the area.
- 5) The effort should commit to:
  - a) Multiple consecutive-year releases at each site, preferably at several sites within the area. As sufficient data are compiled, assessment of progress will be made and recommendations made for continuation of the project.
  - b) Monitoring annually for at least 10 years after the last release (see strategy described at Recovery Task 2.1.3).
- 6) Captive-bred dusky gopher frogs to be used for release into the wild are free of disease so there is no potential for a detrimental effect on in situ amphibian populations, other vertebrate or invertebrate populations, or those in the surrounding geographical region.

A pilot project must be conducted to assess the feasibility of using captive-bred dusky gopher frogs to establish recovery populations. We recommend a research project conducted under natural conditions at the release site. A list of research tasks to be conducted to improve the success of the reintroduction effort includes, but is not limited to, the following topics:

- 1) Determination of life stage best suited for release.
- 2) Advantages and disadvantages of releasing head-started individuals versus naive individuals.
- 3) Important elements of head-starting that enhance subsequent survival after release.
- 4) Release site fidelity of released frogs, comparing captive-bred individuals to translocated wild-bred individuals.
- 5) Comparisons of survival, growth, health, and behavior between released individuals (both translocated wild specimens and captive-bred specimens) with in situ wild specimens.

General guidelines for transportation:

- 1) Eggs:
  - a) Eggs should be collected from masses that are freshly laid (<5 days) or show little signs of development. If hatching has already begun, or if embryos can easily dislodge from the mass during handling, the egg mass should be left alone.
  - b) If transport will take less than 24 hours, eggs may be placed in small, square, plastic containers with lids that have been bleached and rinsed or in new 1 gallon, self-closing plastic bags, rinsed. Fill containers or bags with as much water as possible to prevent sloshing that could damage eggs.
  - c) Containers should be kept shaded and cool. For long trips (those taking over 24 hours), containers should be filled with just enough water to cover the eggs and then fill the rest of the container or bag with air and seal it. Make sure the eggs are kept upright. Eggs can be kept cool in a Styrofoam box or cooler with ice packs, provided that frozen material does not come in direct contact with egg masses.
- 2) Tadpoles:
  - a) Tadpoles to be transported must be moving and have fully-absorbed the yolk. Tadpoles that only move when stimulated have higher mortality during transportation than older, more active tadpoles.
  - b) Tadpoles may be transported in the same method as for eggs (See egg guidelines b) and c), above) or in 5-gallon buckets provided that the containers are filled as much as possible with water to prevent sloshing and injury to tadpoles.
- 3) Metamorphs and Adults:
  - a) Do not use pre- metamorphs with full tails. They do not typically do as well with transport and release at this stage. Only remove metamorphs if tails are approximately 25% or less of their original length.
  - b) Place metamorphs or adults in a well-ventilated plastic container (make sure the ventilation holes have no sharp edges pointing inward that might injure the frog). Place moist substrate in the container. Keep containers shaded and cool until such time as all are ready for transport.

General guidelines for release:

- 1) Eggs: Currently, we are not recommending stocking translocation sites using eggs unless the majority of the release is of another age class and the eggs are considered excess to the needs of the captive propagation facility.
- 2) Tadpoles:
  - a) Fill an appropriate number of 5-gallon buckets to 50% capacity with source water. As tadpoles are captured place them in the bucket. When all tadpoles are captured, fill buckets with source water. Buckets should be tightly sealed for transport to release site.
  - b) At release site, tadpoles should to be acclimatized to their new aquatic environment prior to release, via a series of partial water changes from the release site.
- 3) Metamorphic frogs and adults
  - a) Metamorphic or adult dusky gopher frogs should be released at the edge of the receiving pond, even if it is dry, in the evening.
  - b) Additional studies need to be conducted on the effects of head-starting (enclosing in suitable habitat for a period of time) metamorphic or adult gopher frogs when releasing them to new environments.

Because emerging infectious diseases represent a potential major threat for some amphibian species and populations, monitoring and mitigation of key pathogens must be part of any reintroduction or translocation program. It must be acknowledged that it is nearly impossible to design programs that are completely free of disease risk, but risk can be greatly reduced by careful planning and adherence to recommendations for risk mitigation. One of the greatest risks for introductions of detrimental infectious agents into wild amphibian populations via a reintroduction or translocation program is exposure of reintroduction candidates to other amphibians from outside their native range.

To minimize risk of transfer of pathogens from a captive facility to the reintroduction site, candidate individuals should be maintained in isolation from other amphibians. This point is of special importance in the case of institutions such as zoos, aquariums, nature centers, fish hatcheries where amphibians of a variety of species and sources (a “cosmopolitan collection”) are commonly found (captive and/or wild). Nevertheless, bio-secure isolated colonies of dusky gopher frogs should be established and maintained at such institutions with appropriate attention to physical location, water management, and protocols for husbandry personnel. Conceptually simple, but time-consuming steps enable the rederivation of an isolated colony from founders of most amphibian species that may have not been held in strict isolation.

Prior to reintroduction of individuals from the captive facility, pre-release disease testing for pathogens and parasites of special concern is recommended. Similar screening should be done for all species of amphibians already native at the reintroduction site. Ideally, the individuals will be free of target pathogens (e.g., ranavirus, “chytrid fungus”, etc.). If presence of pathogens is detected among the individuals to be reintroduced, then the identity (including identification of specific genetic strains) should be compared with pathogens already documented from the reintroduction site. The presence of subclinical levels of infection among the individuals to be

reintroduced does not necessarily preclude their reintroduction if the same pathogen or parasites already exist at the reintroduction site. In general, if an identical pathogen is present in both the captive and wild populations, the presence of that pathogen should not impact the decision on whether to reintroduce captive animals to the wild.

Clinically ill individuals with any disease among the captive population should not be released under any circumstances. Similarly, if the captive population has had any significant disease outbreaks in the recent past, then stakeholders should proceed with a focused risk analysis that may include reestablishing a new isolated colony.

In addition to using larval, metamorphic, or postmetamorphic dusky gopher frogs, reintroductions from captive stock may include transfer of egg masses from a captive facility directly to the site of reintroduction. No pathogens are known to be transferred in or upon rinsed amphibian eggs, so vertical pathogen transmission has not been documented and therefore can be assumed to be very unlikely.