

Recovery Plan for Desert Pupfish (*Cyprinodon macularius*)

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DRAFT AMENDMENT 1

We have evaluated the best available information generated since the Desert Pupfish Recovery Plan (Recovery Plan) was completed. In this proposed modification, we synthesize the adequacy of the existing recovery criteria, show new recovery criteria, and provide the rationale supporting the proposed recovery plan modification. The proposed modification is shown as an appendix that updates the Recovery Plan, superseding only the Section II. Recovery, Downlisting Criteria (pp. 13-14) of the Recovery Plan (U.S. Fish and Wildlife Service [Service] 1993: 13-14).

**For
U.S. Fish and Wildlife Service
Southwest Region
Albuquerque, New Mexico**

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Approved: _____ DRAFT _____ Date: _____
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BACKGROUND INFORMATION

Recovery plans should be consulted frequently, used to initiate recovery activities, and updated as needed. A review of the recovery plan and its implementation may show that the plan is out of date or its usefulness is limited, and, therefore, warrants modification. Keeping recovery plans current ensures that the species benefits through timely, partner-coordinated implementation based on the best available information. The need for, and extent of, plan modifications will vary considerably among plans. Maintaining a useful and current recovery plan depends on the scope and complexity of the initial plan, the structure of the document, and the involvement of stakeholders.

An amendment involves a substantial rewrite of a portion of a recovery plan that changes any of the statutory elements. The need for an amendment may be triggered when, among other possibilities: (1) the current recovery plan is out of compliance with regard to statutory requirements; (2) new information has been identified, such as population-level threats to the species or previously unknown life history traits, that necessitates new or refined recovery actions and/or criteria; or (3) the current recovery plan is not achieving its objectives. The amendment replaces only that specific portion of the recovery plan, supplementing the existing recovery plan, but not completely replacing it. An amendment may be most appropriate if

significant plan improvements are needed, but resources are too scarce to accomplish a full recovery plan revision in a short time.

Although it would be inappropriate for an amendment to include changes in the recovery program that contradict the approved recovery plan, it could incorporate study findings that enhance the scientific basis of the plan, or that reduce uncertainties as to the life history, threats, or species' response to management. An amendment could serve a critical function while awaiting a revised recovery plan by: (1) refining and/or prioritizing recovery actions that need to be emphasized, (2) refining recovery criteria, or (3) adding a species to a multispecies or ecosystem plan. An amendment can, therefore, efficiently balance resources spent on modifying a plan against those spent on managing implementation of ongoing recovery actions.

METHODOLOGY USED TO COMPLETE THE RECOVERY PLAN AMENDMENT

The process of review and modification of the existing recovery criteria for the Desert Pupfish was initiated using the Desert Pupfish 2010 status review (5-year review) as a foundation document because it was more recent than the Recovery Plan (Service 1993: entire). This status review was also comprehensive with regard to all information known about this species through 2010. After 2010, when new information became available from external partners regarding field work, surveys, research projects, or other types of efforts, we reviewed, analyzed, and catalogued the information and we have considered that information in this proposed recovery criteria modification. In addition, we sought informal review of the draft Recovery Plan amendment from the States of Arizona and California, the Service's Carlsbad Fish and Wildlife Office, and partners in Mexico. Input on the draft amendment was received from the Arizona Game and Fish Department.

ADEQUACY OF RECOVERY CRITERIA

Section 4(f)(1)(B)(ii) of the Endangered Species Act (Act) requires that each recovery plan shall incorporate, to the maximum extent practicable, "objective, measurable criteria which, when met, would result in a determination...that the species be removed from the list." Legal challenges to recovery plans [see Fund for Animals v. Babbitt, 903 F. Supp. 96 (D.D.C. 1995)] and a Government Accountability Audit (GAO 2006) also have affirmed the need to frame recovery criteria in terms of threats assessed under the five delisting factors.

Recovery Criteria

The original Recovery Plan defined criteria for downlisting the Desert Pupfish subspecies, *C. m. macularius*. Delisting criteria were not established in the Recovery Plan because development of delisting criteria was not considered feasible due to insoluble threats and limited habitat for this subspecies. No recovery criteria were established for the Quitobaquito pupfish subspecies (*C. m. eremus*) in the original Recovery Plan because down- and delisting was not expected due to its limited range, continuing threats to its survival, and lack of historical range in which the subspecies could be recovered.

Synthesis

The Desert Pupfish (*Cyprinodon macularius*) was described by Baird and Girard (1853) from specimens collected in the San Pedro River, Arizona. In the 1980s, pupfish experts realized that the Desert Pupfish in Quitobaquito Springs and the Rio Sonoyta were different from each other

and from the rest of the pupfish within the species historical range (McMahon and Miller 1985). In 1987, Miller and Fuiman named the pupfish at Quitobaquito Springs the Quitobaquito pupfish (*C. m. eremus*). In 2000, Echelle et al. named pupfish in the Rio Sonoyta and Quitobaquito as the Quitobaquito pupfish *C. eremus*. The common name is now the Sonoyta pupfish (Miller et al. 2005, Nelson et al. 2006, Page et al. 2013). The 1993 Recovery Plan included separate criteria for *C. m. macularius* and *C. m. eremus*; this is incorrect because the listed entity is the species, and not the subspecies. Thus the revised and new criteria below covers all of the individuals and populations considered *C. macularius* at the time of listing. This includes what are now considered *C. macularius*, *C. eremus*, and *C. arcuatus*.

Historical collections of pupfish from the Santa Cruz River basin were made in the Tucson Basin, Sonoita Creek, and Monkey Spring (Minckley and Marsh 2009). In 1973, Minckley (pg. 192) considered the Monkey Spring pupfish an extinct and undescribed *Cyprinodon* species. Subsequently, the Monkey Spring pupfish and the other pupfish in the entire Santa Cruz River basin were described and named the Santa Cruz pupfish, *C. arcuatus* (Minckley et al. 2002); and are extinct.

At the time of listing (Service 1986), the historical range of the Desert Pupfish included the Lower Colorado River Basin, the Gila River Basin, Laguna Salada, and the Rio Sonoyta Basin. This geographic area now includes all three species, *C. macularius*, *C. eremus*, and *C. arcuatus* (Echelle et al. 2000, Minckley et al. 2002, Minckley and Marsh 2009, Service 2010). However, the listed entity remains *C. macularius*. What was the Desert Pupfish in 1986 is now recognized as three separate species (Page et al. 2013); the ESA listing should be changed to reflect the taxonomic changes. If a taxonomic name change is made under the ESA, any sub-group split off from the listed entity (*C. macularius*) would also be listed (*C. eremus*).

Collectively, there are 11 extant natural populations of Desert Pupfish known in the wild in the United States and Mexico (California = 5, Arizona = 1, and Mexico = 5; Tier 1 populations in the Recovery Plan). Many reestablishments have been attempted. Approximately 25 transplanted populations of the Desert Pupfish exist in the wild at present, although this number fluctuates due to the ongoing establishment (and failure) of populations (Moyle 2002) (Tier 2 populations in the Recovery Plan) (Service 1993, Voeltz and Bettaso 2003, Robinson and Mosher 2018, Service files). Approximately 47 captive or refuge Desert Pupfish populations (that do not qualify as Tier 3 under the Recovery Plan) exist, comprised of 34 in Arizona, 8 in California, and 5 in Sonora, Mexico. The range-wide status of Desert Pupfish is poor but stable, although the population trend recently has been increasing in Arizona due to an active recovery program (Duncan and Clarkson 2013; Robinson and Crowder 2015; Robinson and Mosher 2018).

The two main threats to the Desert Pupfish have continued mostly unabated since listing of the species, have increased in parts, and are predicted to increase into the future (Minckley and Marsh 2009, Garfin et al. 2013). The two main threats are loss and degradation of aquatic habitats (Factor A), and the continual spread and introduction of non-native aquatic species (Factor C), which prey upon and compete with all pupfish species. These threats of habitat loss, predation, and competition continue to be exacerbated by increasing human development and demand for water, as well as interactions with predicted trends for warmer, drier, and more

extreme hydrological conditions associated with climate change (Flieshman et al. 2013, Gershunov et al. 2013). The 2010 Desert Pupfish 5-year Review (Service 2010) described these same threats and their significant impacts, and challenges. The ongoing long-term drought, in addition to climate change, continues to be synergistic with the threats of habitat loss and degradation and impacts from non-native fish species. The warming and drying caused by global climate change are predicted to continue through the 21st century, which will likely reduce suitable habitat and further concentrate interactions with non-natives. Additionally, because existing regulatory mechanisms do not effectively control the movement and spread of non-native aquatic species, prevent loss and degradation of aquatic habitats, or affect climate conditions and their causes, those regulations are inadequate (Factor D). Definitions and explanations of various terms and concepts and a full explanation of the recovery tasks can be found in the recovery plan.

AMENDED RECOVERY CRITERIA

Recovery criteria serve as objective, measurable guidelines to assist in determining when an endangered species has recovered to the point that it may be downlisted to threatened, or that the species may be delisted. Delisting is the removal of a species from the Federal Lists of Endangered and Threatened Wildlife and Plants. Downlisting is the reclassification of a species from endangered to threatened. The term “endangered species” means any species (species, subspecies, or DPS) which is in danger of extinction throughout all or a significant portion of its range. The term “threatened species” means any species which is likely to become an endangered species within the foreseeable future throughout all or a significant portion of its range.

This amendment applies to the full species as listed (*C. macularius*), which includes both the Desert Pupfish and Quitobaquito pupfish subspecies of the Recovery Plan. We provide downlisting criteria for the Desert Pupfish (*C. macularius*), which will supersede those included in the original Recovery Plan (Service 1993: 13-14) and introduce delisting criteria for the species as follows:

Downlisting Recovery Criteria

Existing recovery criteria (Verbatim)

Desert pupfish (*Cyprinodon macularius macularius*) will be considered for downlisting when:

- (1) **Naturally occurring populations** in the United States and Mexico are established and secure. These include five metapopulations at 12 known locations:
 - (a) Salton Sink (San Felipe Creek/San Sebastian Marsh, upper Salt Creek, and shoreline pools and irrigation drains of Salton Sea, California);
 - (b) Rio Sonoyta, Sonora;
 - (c) El Doctor (3 localities) and Santa Clara Slough (2 localities), Sonora;
 - (d) Laguna Salada, Baja California; and
 - (e) Cerro Prieto (2 localities), Baja California, Mexico;
- (2) Populations of Desert Pupfish are **reestablished** and secure within probable historical range according to specifications detailed in task 2 of this plan (which

include the persistence of a reestablished, secure population for a minimum of 10 years; see Table 3, pg. 19);

- (3) A protocol for exchange of genetic material among reestablished populations is developed and implemented to ensure maintenance of natural levels of allelic genetic diversity; and
- (4) Population and genetic monitoring plans as outlined below in the stepdown of this plan are devised and implemented to routinely assess status of all populations.

Security is herein defined as formal protection of habitat and water rights by methods such as land and water rights acquisition, legislation, or management agreement, and maintenance of a genetically pure, self-sustaining, stable or increasing (viable) population. Until additional information becomes available, a viable population (Lacy 1987, Ryman and Utter 1987, Soule 1987, Templeton 1990) will include not fewer than 500 overwintering adults or existing numbers, whichever is greater, in a normal sex ratio with in-situ reproduction and recruitment sufficient to maintain that number¹.

1 Natural populations may be unable to meet this criterion.

In the United States, formal protection of water and land will be considered to occur when one of the following criteria is met:

- (1) Water rights and habitat associated with each naturally occurring population are in the legal possession of an agency, or organization, or entity whose goals include protection and recovery of endangered species, which possess adequate statutory authority to protect those populations against other land and water uses which may adversely affect Desert Pupfish, which has adequate regulations in place to enforce such authority, and which has demonstrated over a period of not less than 10 years adequate capability to protect and manage a viable population of Desert Pupfish.
- (2) A legally-binding, long-term (>25 years) agreement is in place between the land and water rights owner(s) and an agency, organization, or entity such as described above, which provides sufficient legal rights to the agency or organization to manage a viable population of Desert Pupfish. The efficacy of this agreement should be demonstrated over a period greater than (if not equal to) 10 years.

In Mexico, formal protection of land and water will be considered to occur when security comparable to that defined for the United States is achieved (See Table 3, below).

Locally adjacent Desert Pupfish populations are considered separate only if a discrete catastrophic event (e.g., invasion by exotic fishes, habitat destruction, etc.) is likely to

impact only one population. Unless demonstrated otherwise on a case-by-case basis, the presence of non-native fishes is considered a threat to Desert Pupfish population viability.

Once this plan is finalized and approved, downlisting of *C. m. macularius* is expected to take 15 years. Total recovery (delisting) is not expected in the foreseeable future.

FROM DESERT PUPFISH RECOVERY PLAN, PAGE 19.

Table 3. Re-establishment specifications for *Cyprinodon macularius macularius* populations. There are 130 replicates required.

Area	Natural Populations	Re-established Populations	
	Tier 1	Tier 2	Tier 3
Arizona	0	10	45
California	3	9 (3 reps. Of each natural)	27 (9 reps. Of each natural)
Colorado Delta, MX	3	9 (3 reps. Of each natural)	27 (9 reps. Of each natural)
Rio Sonoyta, MX	1	- 3 of either tier 2 or 3 -	

Specifications:

Tier 2 populations will receive a high degree of protection and will be long-term populations. A tier 2 population will be considered to be successfully established and count toward recovery if it has survived for 10 years and has required only minor management to persist. Minor management may include:

habitat-

- 1) minor vegetation removal
- 2) fencing
- 3) drawing off excess water for wildlife and livestock

populations-

- 4) population monitoring
- 5) management for other native species
- 6) pupfish transfers for genetic maintenance

Major management actions which would preclude a population from being considered successful would include:

habitat-

- 1) new or modified water supply
- 2) dredging
- 3) major vegetation removal
- 4) habitat (re)construction
- 5) exotic fish introduction or control

populations-

- 1) restocking pupfish

- 2) supplemental stockings of pupfish (for reasons other than genetic protocol)

Tier 3 populations may experience major management activities. Management will not preclude counting populations as contributing towards recovery. The specified total number of populations must be achieved and continuously maintained for 10 years.

Amended recovery criteria

Desert pupfish (*Cyprinodon macularius*) will be considered for downlisting when:

- (1) Naturally occurring populations in the United States and Mexico are established and secure. These include seven Management Units at 14 known locations:
 - (a) San Felipe Creek/San Sebastian Marsh, California;
 - (b) The rest of the Salton Sink (upper Salt Creek, and shoreline pools and irrigation drains of Salton Sea) California;
 - (c) El Doctor (3 localities) and Santa Clara Slough (2 localities), Sonora;
 - (d) Laguna Salada, Baja California; and
 - (e) Cerro Prieto (2 localities), Baja California, Mexico;
 - (f) Rio Sonoyta, Sonora;
 - (g) Quitobaquito Spring, Arizona;
- (2) Populations of Desert Pupfish are reestablished and secure within probable historical range according to specifications detailed in task 2 of this plan and Table A below (which include the persistence of a reestablished, secure population for a minimum of 10 years);
- (3) A protocol for exchange of genetic material among reestablished populations is developed and implemented to ensure maintenance of natural levels of allelic genetic diversity; and
- (4) Population and genetic monitoring plans as outlined below in the stepdown of this plan are devised and implemented to routinely assess status of all populations.

“Secure” populations are defined as formal protection of habitat and water rights by methods such as land and water rights acquisition, legislation, or management agreement, and maintenance of a genetically pure, self-sustaining, stable or increasing (viable) population. Until additional information becomes available, a viable population (Lacy 1987, Ryman and Utter 1987, Soule 1987, Templeton 1990) will include not fewer than 500 overwintering adults or existing numbers, whichever is greater, in a normal sex ratio with in-situ reproduction and recruitment sufficient to maintain that number.

In the United States, formal protection of water and land will be considered to occur when one of the following criteria is met:

- (1) Water rights and habitat associated with each naturally occurring population are in the legal possession of an agency, or organization, or entity whose goals include protection and recovery of endangered species, which possess adequate statutory

- authority to protect those populations against other land and water uses which may adversely affect Desert Pupfish, which has adequate regulations in place to enforce such authority, and which has demonstrated over a period of not less than 10 years adequate capability to protect and manage a viable population of Desert Pupfish; or
- (2) A legally-binding, long-term (>25 years) agreement is in place between the land and water rights owner(s) and an agency, organization, or entity such as described above, which provides sufficient legal rights to the agency or organization to manage a viable population of Desert Pupfish. The efficacy of this agreement should be demonstrated over a period greater than (if not equal to) 10 years.

In Mexico, formal protection of land and water will be considered to occur when secure populations comparable to that defined for the United States is achieved (See Table A, below).

Locally adjacent Desert Pupfish populations are considered separate only if a discrete catastrophic event (e.g., invasion by exotic fishes, habitat destruction, etc.) is likely to impact only one population. Unless demonstrated otherwise on a case-by-case basis, the presence of non-native fishes is considered a threat to Desert Pupfish population viability.

Table A below differs from Table 3 of the Recovery Plan in that it is based on genetic information (Echelle 2008, Echelle et al. 2007, Koike 2007, Loftis et al. 2009), and not just the remnant natural populations. However, the only difference between the two criteria is that the Salton Sink populations have now been separated into two groups: San Felipe Creek and San Sebastian Marsh as one group, and the second group contains all other populations in the Salton Sink (e.g. Salt Creek, irrigation drains, Salton Sea). Table A was constructed to contain a similar number of Tier 2 and Tier 3 populations required for downlisting. We also used the 1:3 ratio of Tier 2 to Tier 3 populations in Table 3.

Table A. Reestablishment specifications (number of populations) required to downlist the Desert Pupfish within historical range. There are 130 replicates.

Management Unit	Tier 2	Tier 3
San Felipe Creek/San Sebastian Marsh, California	6	18
Other Salton Sink populations, California	6	18
Laguna Salada, Baja California	6	18
Cerro Prieto, Baja California	6	18
El Doctor/Cienega de Santa Clara, Sonora	6	18
Rio Sonoyta, Sonora	6 of either Tier	
Quitobaquito Springs, Arizona	6 of either Tier	

Additionally, we also change the definition of a captive population to that below. The definition applies to all replicated populations used to meet downlisting or delisting criteria.

Captive population: populations established outside of or within historic range in aquaria, pools, or ponds at a location that has a mailing address.

Delisting Recovery Criteria

Existing recovery criteria

None

Amended recovery criteria

In addition to achieving all of the downlisting criteria specified above, Desert Pupfish (*Cyprinodon macularius*) will be considered for delisting when:

- (1) Populations of the seven Desert Pupfish Management Units (Table B, below; Echelle et al. 2007:13) are reestablished and secure within the historical range of the species according to specifications detailed in task 2 of the Recovery Plan and, at least two of these populations are in a large riverine system, such as in the Colorado, Gila, Hardy, Santa Cruz, San Pedro, or Salt Rivers

Justification: The genetic work conducted by the Anthony Echelle Lab (Oklahoma State University) on Desert Pupfish was instrumental in providing a clear representation of the phylogenetic relationships of natural and reestablished Desert Pupfish populations (Echelle 2008, Echelle et al. 2007, Koike 2007, Loftis et al. 2009). Thus, the Management Units they identified represent the best available information on the subject. The genetic Management Units largely mirror the metapopulations in the Recovery Plan (Service 1993:13). Requiring that two replicated populations be established in a larger river within historical range (Service 1993:2) not only addresses the two main threats of loss of water and non-native species, but also greatly enhances the resilience, redundancy, and representation of the species.

Certain larger rivers will likely retain flowing water for the foreseeable future as they transport water for human use. For example, by Treaty, the United States must deliver one million acre-feet annually to the Republic of Mexico from the Colorado River. Because the point of measurement is at the international border, the river channel is the most efficient conveyance. The Verde and Salt rivers provide a significant portion of the water used in the Phoenix metropolitan area. For pupfish to maintain a viable population in a large riverine system, problematic non-native fish must be removed or adequately controlled.

The creation of additional populations to replicate the genetic Management Units will increase resilience, redundancy, and representation. Additionally, populations in large

rivers would add the one historical ecological niche that is currently not occupied. A population of Desert Pupfish in a large river would allow the species to expand, contract, and potentially access new areas as they did historically. Large habitats in and of themselves provide resilience and representation.

- (2) A population which meets all other requirements (Service 1993:19) to qualify as a Tier 2 population must persist a minimum of 20 years (as opposed to 10 years as described in conditions defining a Tier 2 population).

Justification: In Arizona, conservation and management of the Gila Topminnow *Poeciliopsis occidentalis* and Desert Pupfish are comingled. Hundreds of topminnow reestablishments have been attempted in Arizona, and most of those were extirpated in less than 10 years (Weedman 1999, Voeltz and Bettaso 2003). Many reestablished Desert Pupfish populations in Arizona also persisted for less than 10 years (Weedman and Young 1997, Robinson and Mosher 2018). Like the Recovery Plan, the draft revised Gila Topminnow Recovery Plan required reestablished Tier 2 populations to persist for 10 years before a population could count towards delisting to give reestablished populations adequate time to become established. Requiring a population to persist for 10 years before counting it towards downlisting criteria accounts for the difficulty in reestablishing populations and insures that the best sites remain occupied. Requiring a reestablished population to persist for at least 20 years ensures that threats have been eliminated or substantially reduced and that maintenance of the best habitats can be achieved over the long term before the protections of the Act are removed.

- (3) The specified total number of populations (Table B) must be achieved and continuously maintained for 20 years.

Justification: Similar to the justification above, the total number of reestablished populations is never constant since reestablished populations continue to be extirpated and attempts to establish new populations are implemented. Requiring that these populations be maintained for 20 years ensures the downlisting criteria are truly met given the fluctuations in populations. The Gila Topminnow again provides an example of the prudence of this criteria. The criteria for the number of reestablished populations identified in the recovery plan for downlisting was met for several years, and a downlisting proposal was drafted (Simons et al. 1989; Duncan, *in review*, Service files). In the intervening years, the number of reestablished Gila topminnow populations fell below the threshold required to downlist the species. The downlisting proposal was terminated, and the Desert Fishes Recovery Team, the Service and other partners working on the conservation of the Gila Topminnow realized the 1984 Gila Topminnow recovery plan was inadequate and needed revision. Through population restoration efforts with small, endemic fishes, we have learned that a period of time is required for a reestablished population to be considered an enduring, viable population.

Table B. Reestablishment specifications (number of populations) required to delist the Desert Pupfish in California, Baja California, Sonora, and Arizona. There are 177 population replicates required.

Management Unit	Tier 2	Tier 3
San Felipe Creek/San Sebastian Marsh	6	27
Other Salton Sink populations, California	6	27
Laguna Salado	6	27
Cerro Prieto, Baja California	6	27
El Doctor/Cienega de Santa Clara, Sonora	6	27
Rio Sonoyta, Sonora	6 of either Tier	
Quitobaquito Springs, Arizona	6 of either Tier	

The objective to establish 27 Tier 3 populations is derived from the criteria for downlisting in the recovery plan, but applied to each management unit, instead of nine replicates of each natural population in California and the Colorado Delta. An additional 50 percent of Tier 3 replicates was considered reasonable to recover the species. Tier 3 sites are a lower quality than Tier 2 sites. However, they are more numerous and also give management agencies additional flexibility in how they approach the recovery of the species. Management Units (MUs) overlap with all previously delineated metapopulations except for the top two MUs, which were previously considered to be one metapopulation or Management Unit. At the time of the 1993 Recovery Plan, the Quitobaquito pupfish was not included in the downlisting criteria. Including the Quitobaquito pupfish here accounts for the seventh Management Unit/metapopulation in the table above. Delisting criteria are delineated by Management Units, which reflect genetic units, similar to the metapopulations used to delineate downlisting criteria. For Quitobaquito Springs (and possibly also for Rio Sonoyta), replicating existing habitat is limited by the availability of sites that meet the replication criteria. Therefore, replication of this population must occur in locations outside of the Quitobaquito Springs Management Unit, and only six of either Tier are required due to realistic habitat limitations.

All classification decisions consider the following five factors: (1) is there a present or threatened destruction, modification, or curtailment of the species' habitat or range; (2) is the species subject to overutilization for commercial, recreational scientific or educational purposes; (3) is disease or predation a factor; (4) are there inadequate existing regulatory mechanisms in place outside the ESA (taking into account the efforts by states and other organizations to protect the species or habitat); and (5) are other natural or manmade factors affecting its continued existence. When delisting or downlisting a species, we first propose the action in the *Federal Register* and seek public comment and peer review. Our final decision is announced in the *Federal Register*.

Rationale for Recovery Criteria

The numbers of replicated populations that are required for down- and delisting reflect the conditions aquatic species face in arid North America, including recovering a species that also occurs in Mexico. The criteria in the Recovery Plan regarding security of pupfish populations and sites address both threats; attaining criteria for populations is a surrogate for measuring effectiveness of managing for non-native species and habitat loss in terms of water availability/contamination for Desert Pupfish. Conservation actions are often taken to control and reduce the spread of non-native aquatic species, though not everywhere the pupfish occurs. Some non-native species are notoriously difficult to remove once they become established, and both legal and illegal restocking of non-native species are expected to continue. Non-native species currently co-occurring with Desert Pupfish include Green Sunfish, Tilapia, Sailfin Mollies, and Largemouth Bass. While we know what non-native species are present in existing Desert Pupfish populations, predicting which non-natives will invade other pupfish populations is not possible with any certainty. Lastly, new non-native species are discovered almost annually within the historical range of the Desert Pupfish, and it is not possible to predict their arrival or effects.

Replenishing waters in desiccated areas specifically needed for recovery to replicate conditions where the species previously occurred and removing non-natives in critical areas required for the species recovery present significant technical and political challenges. In these areas needed to delist the pupfish, there are many sources for recontamination with non-natives, effectively requiring areas to be treated and re-treated, and data on tracking non-natives are lacking. Replenishing depleted waters removed from aquatic habitats such as the Salton Sea is highly unlikely, and the effective removal of non-native species in the remaining aquatic habitats in Arizona, California, and Mexico would involve a concerted effort by citizens and governments, given these habitats are constantly re-contaminated.

We have information concerning these threats, and have a general understanding of the impacts of the threats to Desert Pupfish, yet we do not have the extent of quantitative information we would like to develop recovery criteria to depict how many individuals and populations are needed in terms of resilience, representation, and redundancy for the species' long-term viability. However, we have used the best available scientific and commercial information to develop the modified recovery criteria discussed above.

In conclusion, although reestablishment of new Desert Pupfish populations occurs, new populations will not abate the major threats. Many of these sites are small in size, are disjunct, and may not persist for a long time. The species' former status of living in a diversity of connected aquatic environments, from large rivers (lower Colorado) to ciénegas (small, natural water holes associated with springs) across the landscape, without the presence of non-natives, will be exceedingly difficult to replicate.

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